ENDOCRINE AND REPRODUCTIVE CHANGES IN DAIRY HEIFERS AS AFFECTED BY GROWTH RATE AND MELENGESTROL ACETATE

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ABSTRACT

ENDOCRINE AND REPRODUCTIVE CHANGES IN DAIRY HEIFERS AS AFFECTED BY GROWTH RATE AND MELENGESTROL ACETATE

BY

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This study was conducted to determine the effects of a normal and high level of nutrition alone or with the synthetic progestagen melengestrol acetate (MGA) on body growth, levels of certain anterior pituitary hormones in the pituitary and blood, development of the reproductive tract and mammary gland, and subsequent reproductive and lactational performance of 140 Holstein heifers. Heifers were raised under uniform conditions from 2 weeks to 2.5 months of age at which time they were randomly assigned to 14 groups consisting of 10 heifers each. MGA was fed beginning at 2.5 months of age or after first estrus. One hundred heifers were slaughtered either at 2.5 months of age, at first estrus, or at breeding size, while 40 heifers fed a roughage ration only between pregnancy diagnosis and parturition were kept to obtain data on breeding and lactational performances.

Heifers fed the high level of nutrition exhibited first estrus at a significantly younger ($P<0.01$) age than those fed the normal level (7.5 $\pm$ 0.1 vs 8.7 $\pm$ 0.2 months), but there was no significant difference ($P>0.10$) in body weight (255 $\pm$ 4 vs 250 $\pm$ 5 kg) or withers height (108.6 $\pm$ 0.6 vs 109.2 $\pm$ 0.7 cm) at first estrus. These data emphasize that first estrus is

associated more with physical size than with calendar age. At breeding size (120 cm withers height), heifers fed the high level of nutrition were 11.4 ± 0.4 months old while those fed the normal level were 12.5 ± 0.2 months old ($P<0.01$). MGA fed with either the normal or high levels of nutrition at the rate of 0.45 mg per heifer per day did not significantly affect the ages at breeding size, indicating that MGA did not affect skeletal growth. However, MGA increased body weight gains, but only when fed with the high level of nutrition ($P<0.01$). Heifers fed the high level of nutrition with MGA gained faster ($P<0.05$) after about 5.5 months of age than heifers fed the high level alone. The time from first estrus to breeding size (about 3.5 months) was not significantly different ($P>0.10$) for heifers fed the two nutritional levels without or with MGA, indicating that level of nutrition or addition of MGA did not affect rate of skeletal growth after first estrus.

Uterine weights, nucleic acids concentrations, and epithelial cell heights were not affected by level of nutrition, but these parameters indicated that uterine hypertrophy was associated with MGA feeding. Ovarian weights were not affected by nutritional level, but more large diameter follicles were present on the ovaries of heifers fed MGA. Level of nutrition had no effect on parenchymal tissue weights or nucleic acids concentrations and contents of the mammary gland. MGA did not affect mammary parenchymal tissue weights but caused significantly greater ($P<0.01$) concentrations and contents of nucleic acids in the heifers at breeding size. Paired adrenal weights were not significantly different ($P>0.10$) for groups fed the two levels of nutrition without or with MGA. However, at breeding size, MGA caused a significant decrease ($P<0.05$) in the width of the glucocorticoid producing fasciculata zone of the cortex. No large differences in pituitary weights or pituitary or plasma concentrations of

LH, FSH, and prolactin resulted at first estrus or breeding size from feeding the two levels of nutrition without or with MGA. In all treatment groups, correlation coefficients between pituitary concentration and plasma concentration of LH and prolactin, and between pituitary content and plasma concentration of these two hormones were not significant ($P>0.05$).

The interval between MGA withdrawal and occurrence of estrus was considerably longer, though not significantly different ($P>0.10$), for heifers fed MGA from 2.5 months than for those that received MGA after first estrus only (19.7 vs 7.7 days). However, once estrous cycles commenced they were of normal length (17-24 days) for all MGA treated animals. While heifers fed the high level of nutrition without or with MGA were younger at breeding size than those fed the normal level without or with MGA, there were no significant differences ($P>0.10$) among treatment groups in ages at conception or services required per conception. At parturition, the level of nutrition or MGA fed prior to conception produced no significant differences ($P>0.10$) in body weights or withers heights of the dams, birth weights of the calves, or in the subjective dystocia ratings. Birth weights of the calves sired by the two bulls were not significantly different ($P>0.10$). There were no significant differences ($P>0.10$) between the treatment groups in actual milk production weights for the first 60 days of lactation or extended 305 day milk production values.

ENDOCRINE AND REPRODUCTIVE CHANGES IN DAIRY HEIFERS AS AFFECTED BY GROWTH RATE AND MELENGESTROL ACETATE

By

Donald Edward Pritchard

A THESIS

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Dedicated to my grandfather, Oliver Edward, and to my father, Earl Stanley, whose dreams and sacrifices encouraged and permitted me to obtain my college education.

IT'S ALL IN THE STATE OF MIND

If YOU think you are beaten, you are;
If you think you dare not, you don't;
If you think you'd like to win, but you can't;
It's almost a "cinch" you won't;
If you think you'll lose, you've lost;
For out in the world you'll find
Success begins with a fellow's will -
It's all in the state of mind.
FULL many a race is lost
Ere even a race is run,
And many a coward fails
Ere even his work's begun.
Think big and your deeds will grow
Think small and you fall behind.
Think that you can, and you will;
It's all in the state of mind.
If YOU think you are outclassed, you are;
You've got to think big to rise;
You've got to be sure of yourself before
You can ever win a prize.
Life's battle doesn't always go
To the stronger or faster man;
But sooner or later, the man who wins
Is the fellow who thinks he can.

Author Unknown.

AUTOBIOGRAPHICAL SKETCH

of

Donald Edward Pritchard

Born in Aurora, Illinois, on March 9, 1942, I grew up on a dairy-grain farm about one hour's drive west of Chicago, Illinois. My primary and secondary education was obtained from the Hinckley-Big Rock Consolidated School District. I attended the University of Illinois from September, 1960, to March, 1966, and received a B.S. and M.S. in dairy science during that time. In March of 1966 I was awarded a graduate research assistantship by the Department of Dairy, Michigan State University. This position allowed me to study for my Ph.D., which was completed in December, 1969.

ACKNOWLEDGMENTS

In acknowledging all those who have assisted me during my Ph.D. program, I first must thank the Department of Dairy here at Michigan State University. Without its interest and financial assistance, my studying for a doctoral degree would not have been possible. My advisor, Dr. Louis Boyd, and Dr. Harold Hafs both receive my sincerest appreciation for their advice, support, assistance, and encouragement. Were it not for them, I would have terminated my program the first year. I also thank Drs. Paul Reineke, John Gill, and Karl Wright for their advice and guidance, and for serving on my graduate committee.

My research program required the assistance of numerous people. Dr. R. G. Zimbelman and his staff in the TUCO products division of the Upjohn Company were most generous in helping to finance the study and in supplying the melengestrol acetate. Mr. Dennis Armstrong, the M. S. U. dairy herd manager, was most helpful in procuring the heifers and in providing the facilities and labor to house and care for them. Drs. H. A. Tucker and J. T. Huber deserve recognition for the many hours of advice and assistance they rendered in conducting various aspects of the study. And the technical assistance and loyal dedication of Marianne Holfelner is appreciated. I am also indebted to my student contemporaries for their advice and many hours of assistance provided in taking the monthly body measurements, observing the heifers for estrus, and slaughtering the heifers. Members of the group included Roger Purchas, Bill Thatcher, Art Hackett, Robert Wettemann, Lloyd Swanson, Jim Koprowski, Linda Miller,

Wayne Oxender, and Dean Peterson.

The gifts of purified LH and the antibody to LH from Drs. Niswender and Midgley, and the hormones from Squibb Laboratories and the Endocrinology Study Section of the National Institutes of Health are recognized and appreciated.

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INTRODUCTION

During this century mankind has made tremendous progress in all facets of science. Industrial production has reached levels that at one time were unimaginable. Space technology has now attained man's age old dream of flying to the moon. Eradicating many diseases and controlling others that have plagued man for centuries have extended our life expectancy by decades. Equally important as these areas is the progress that has occurred in the agricultural industry. Mechanized farming, crop yields per acre, production per animal, and output per man make the agriculture of yester-years appear strangled with inefficiency. Because of gigantic advancements in food production, spurred by Land Grant universities, approximately 95 percent of our population is free to pursue other industrial and technical endeavors.

Dairymen have also shared in the benefits of science. Besides having automated equipment for handling the feed and waste products, milk is now removed from the cow, transported, processed, and never exposed to the air until the consumer pours it from a container. Feed additives which provide a nonprotein source of nitrogen and estrous cycle control are examples of progress in dairy production. And surely artificial insemination and the use of frozen semen have been a real bonanza to genetic progress.

We now know how to feed cows, how to milk cows, how to manage cows, and how to breed cows to increase their milk producing ability. But

while these research findings and applied procedures have been filtered out to dairymen at various rates over time, little progress has resulted in having dairy heifers freshen at an earlier age. Official records show that, on the average, heifers today are about 30 months of age at first parturition, which is a few months older than what Eckles found in a 1915 survey. Many dairymen still feed and breed their heifers in the same manner they did many years ago. This means that heifers are about two and a half years old before they become a productive unit. Needless to say, this time period is wasteful and costly. It should be reduced by feeding heifers so they will grow faster and breeding them according to body size rather than age.

Recent studies by Sinha (1967) indicate that mammary gland development, as measured by deoxyribonucleic acid (DNA) determinations, was about the same for both 9- and 16-month old heifers. This finding suggests that at least in terms of maximal mammary development before conception, there would be no advantage to delay breeding of heifers beyond 9 months of age. However, Desjardins (1966) and Hackett (1968) found that uterine nucleic acids approximately doubled between 12 and 17 months of age in Holstein heifers. This fact suggests that the reproductive tract of 12-month old Holstein heifers is not fully developed and perhaps not completely ready to support pregnancy. However, epithelial cell heights of all portions of the tubular genitalia did not change greatly between the two ages and this characteristic may be more important than the DNA changes. This finding on epithelial cell heights implies that the reproductive tract of 12-month old heifers may be capable of supporting pregnancy successfully.

Thus, armed with the desire to have heifers calve sooner and the previous observations on mammary gland and reproductive tract development

as an incentive, this study was initiated. My immediate purposes were to determine if Holstein heifers could grow to the usual breeding size (320-385 kg) by 12 months of age, and to study the physiological effects of rapid growth on certain endocrine, mammary gland, and reproductive tract changes at the beginning, at puberty, and at the end of this growing period. My ultimate aim was to determine if growing heifers as fast as possible and then breeding them according to body size rather than age is a feasible and practical approach to raising herd replacements. With these goals driving me, I enthusiastically proceeded to conduct this study, a combination of basic and practical research.

REVIEW OF LITERATURE

A. Growth Rate of Dairy Heifers

1. Influence of Nutritional Level

Numerous studies have been conducted to determine the influence of underfeeding and overfeeding on growth rate of dairy heifers. The work of Eckles (1915), which was later expanded upon by Eckles and Swett (1918), is classic in this field. Eckles divided 40 heifers into two groups; one of which received a heavy ration from birth to first calving and the other group a light ration. The heavy ration consisted of whole milk during the first 6 months and all the grain and hay the animals would consume up to first calving. The light - fed group received skim milk during the first 6 months, and hay or pasture only from that age to first calving. His conclusions are generally still acceptable today: a) the heavy ration accelerated skeletal growth, especially during the period of most rapid development; b) later in the growing period , heifers receiving the heavy ration became excessively fat; c) the animals receiving the light ration grew less rapidly ,but continued growing for a longer period of time; and d) the level of nutrition fed growing heifers had a greater effect upon body weight than upon the rate of skeletal growth.

Reed et. al., (1924) compared an all roughage ration with roughage plus grain for heifers after 6 months of age,and found satisfactory development of Holstein heifers only when grain was included in the ration. Herman and Ragsdale (1946) overfed growing heifers and noted

that they were characterized by a heavy, course build which in dairy heifers is objectionable and costly. Hansson (1956), in a series of experiments in Sweden, fed heifers at levels ranging from 51 to 124 percent of the Swedish normal feeding program. Heifers receiving the highest levels of feeds gained more than twice as much as the heifers fed the lowest levels during the period of 1 to 19 months of age. The great retardation in rate of growth of heifers on the extremely low level of nutrition had no serious effect on growing capacity after the level of feeding was increased.

Crichton et. al., (1959,1960a) fed heifers for 44 weeks on either a high or low plane of nutrition, then reversed the nutrition level on half of the heifers in each group until 2 months before parturition. In heifers kept continuously on the restricted nutritional level, they noted that late maturing characters such as live weight and heart girth were affected most while height and length which are earlier maturing characters were affected least. Height was more affected than length in the heifers which had their rations reversed from a high to a low nutritional plane. Using identical twin heifers, Swanson (1957,1967) has for several years studied the effects of nutritional level on growth. His findings concur with those of other investigators who have used unrelated animals.

Cornell workers raised dairy heifers at various nutritional levels in an extensive study of the causes and prevention of reproductive failures in dairy cattle. Sorensen et al., (1959) and Reid et al., (1964) reported on the growth rate of heifers included in the experiment. The low and high levels of feed consumption were 61 and 129 percent of the medium level which amounted to 93 percent of the total digestible nutrients recommended by Morrison (1956). After 80 weeks, heifers on

the low nutrient level weighed about 350 pounds less and were about 10 and 30 centimeters shorter in height and length, respectively, than heifers on the medium nutrient level. Meanwhile, heifers on the high nutrient level were only about 200 pounds heavier and 3.5 centimeters taller and longer than the medium level group. These findings indicate that at feeding levels used in this study, low nutrient intake retards growth more than high nutrient intake accelerates it.

One of the most recent studies evaluating the influence of nutrition level on growth in Holstein heifers was reported by Gardner and Garcia (1966). Starting at 6 weeks of age, 24 heifers were fed grain and alfalfa hay free choice, while 24 control heifers were limited to 4 pounds grain per day and alfalfa hay free choice. All heifers were changed to roughage only after pregnancy verification. Heifers fed grain free choice grew 40 percent faster than controls in body dimensions. Evidence from this study tends to negate the notion that heifers fed rations of high caloric value utilize excess calories for fattening rather than growth.

Most of the studies cited in this section were reviewed in detail by Schultz (1969).

2. Effect of Gonadal Steroids

To my knowledge, no one has attempted to stimulate growth in dairy heifers with gonadal steroids. So the literature review which follows describes the growth stimulatory effects of gonadal steroids in beef cattle. From such studies one can obtain indications of the responses that might result if dairy heifers were given the gonadal steroids. An excellent review was presented by Casida et al., (1959) in a publication by the National Research Council (NRC) of the National Academy of Sciences.

The following evidence on the effects of testosterone and the estrogen-like compounds was obtained from this NRC publication.

Use of testosterone to stimulate growth in heifers has proven to be effective only in certain studies. Apparently, intramuscular injections of about 1 mg per kg of body weight per week are required to cause an increase in feed efficiency and rate of gain. The detriments to using testosterone in heifers are that it produces a marked masculine behavior and appearance, effective results require intramuscular administration, and the cost per animal is greater than for the synthetic estrogens or progestagens.

The estrogen-like compound used most often as a growth promoter has been diethylstilbestrol (DES). Although it is most effective in steers, it does increase weight gain in heifers by about 0.01 to 0.35 pounds daily. Other orally active estrogens which have been used are dienestrol and hexestrol. They increase rate of gain to approximately the same degree as DES. Estrogens, like testosterone, also improve feed efficiency. Despite the beneficial effects of estrogens, they do cause undesirable effects. Relaxation of the lumbar ligaments, producing the typical nymphomaniac stance, is objectional to many cattlemen. Furthermore, extreme hyperemia and swelling of the external genitalia, an increased incidence of vaginal prolapse, and mammary development and teat growth may result from the estrogens.

While numerous studies have been conducted with various estrogens and androgens to improve the performance of feedlot heifers, little attention had been given until the early 1960's to the possible use of progestagens. Perhaps this is because the progestagens were not considered anabolic. Only recently have potent and orally active synthetic progestagens become available for growth promotion. Raun et al., (1965)

were among the first to study the effects of a synthetic progestagen on growth in cattle. In their study, heifers fed chlormadinone acetate (CAP) gained 13.3 percent faster than the control heifers. Bloss et al., (1966) obtained significantly greater weight gains and feed efficiencies from feeding 0.35 to 0.50 mg melengestrol acetate (MGA) daily to beef heifers. Burroughs et al., (1966) found that MGA improved live weight gains by 15 percent over controls. But, Newland and Henderson (1966) and Young et al., (1969) reported no beneficial effects on growth rate from feeding MGA. Still, the unpublished summaries of over 100 trials conducted by the Upjohn Company, the developer of MGA, in cooperation with universities and feedlots throughout the country show an increase of at least 10 percent in weight gain by MGA heifers over controls (Zimbelman, 1968). These findings lend credence to the effectiveness of the progestogen as a growth stimulant.

3. Effect on Age at Puberty

That age at sexual maturity is influenced to a considerable extent by the ration is an accepted fact among animal husbandrymen (Casida, 1959, Reid, 1960). Eckles (1915) noted that heifers receiving a heavy ration mature sexually at an age from 2 to 4 months younger than those receiving a light ration. In his study, heavy fed Holstein heifers had their first estrus at an average age of 8.7 months, while light fed heifers exhibited first estrus at 12.4 months of age. These ages are somewhat younger than those observed by other workers. Reed et al., (1924) observed first estrus at 18.5 months for heifers fed an all forage ration from 6 months of age, while heifers that received grain in addition to forage exhibited first estrus at 13 months of age. The heifers of Hansson (1956) that were fed 43, 62, 81, or 119 percent of

the normal growing feed requirements, exhibited first estrus at 13.3, 12.5, 10.9 and 10.6 months of age, respectfully. Feeding the normal ration allowed first estrus to occur at 10.4 months of age. In the study of Crichton et al., (1959), heifers fed the high plane of nutrition exhibited first estrus when 12.4 months old, while the low plane heifers were delayed until 15.8 months. The group that was switched from a low to a high nutrition plane at 44 weeks of age showed first estrus at 14.7 months, whereas the group that went from the high to the low level was retarded to 18.4 months. It is interesting to note that going from a high to a low nutrition level slows the attainment of sexual maturity more than does a continual low nutrition level.

Sorensen et al., (1959) found striking differences in the average age at first estrus in heifers fed three levels of nutrition. Fifteen heifers on the high feeding level came into estrus at 8.7 months of age, whereas 10 heifers on the medium feeding level averaged 11.4 months. Only 3 of the 5 heifers on the low feeding level showed estrus before they were slaughtered at 80 weeks of age, and they averaged 15.5 months of age. Gardner and Garcia (1966) increased growth rate through accelerated feeding to the extent that the heifers exhibited first estrus when they were 7.7 months old. Control heifers were 9.7 months old at first estrus. Desjardins (1966) fed 24 heifers for a normal growth rate and observed first estrus at an average age of 6.9 months.

Since there is such variation in age at first estrus among the studies cited, it is apparent that differences in nutrition levels and /or accuracy and method of detecting first estrus existed among the experiments. It is important to remember, as Sorensen et al., (1959) stated, that there is more of a tendency for heifers to come in first estrus at a given skeletal growth rather than at a certain weight or age.

4. Effect on Conception and Dystocia

Few studies using different nutrition levels for rearing heifers have reported the effects on conception and dystocia. Reid (1960) cited a New Zealand report which stated that heifers fed a high level of energy while growing required more services per conception than heifers on a low nutritional plane. He also stated that in the Cornell study the percentage of heifers conceiving at first service was 79, 68, and 58 for the low, medium and high nutritional levels, respectively. Thus, this study and others cited by him suggest that nutrition level affects either fertilization rate or embryonic mortality. However, other studies have shown no difference attributable to feeding level on conception rate (Eckles, 1915, Reed et al., 1924, Joubert, 1954, Reid et al., 1964, Hibbs and Conrad, 1965, Gardner and Garcia, 1966). The number of services required per conception in all of these studies ranged from about 1.0 to 2.0. Since the literature contains differing opinions, no conclusion can be made on the nutrition level effect on conception.

It is not possible or correct to relate feeding level during the growing period to dystocia at first parturition. Rather, it is more a matter of relating size of heifer at parturition to dystocia. Heifer size at parturition may, in turn, be related to feeding level during rearing. It is well established that if a heifer does not have sufficient skeletal size at parturition, she will encounter a certain degree of dystocia (Wickersham and Schultz, 1963, Swanson and Hinton, 1964, Reid et al., 1964, Hibbs and Conrad, 1965, and Gardner and Garcia, 1966). Thus, it is important that heifers be fed adequately before

parturition to ensure the skeletal growth necessary to eliminate or minimize dystocia.

5. Effect on Subsequent Lactational Performance

Ultimately, the ability of a dairy heifer to produce milk (and progeny) determines her value. Thus, if factors other than genetic potential, such level of nutrition during the growing period, influence milk production they should be considered by dairymen.

Eckles (1915) considered heifers receiving a heavy ration until first parturition to be slightly inferior in milk production to those receiving a light ration. Turner (1932) concluded that the most efficient milk production would be obtained by breeding heifers to calve at 20 to 24 months of age. This would mean that heifers should be fed so they would grow large enough by parturition to minimize calving problems.

In a study by Herman and Ragsdale (1946), milk production of heifers which received the "rapid growth ration" until parturition was disappointing to them and remained so for the second and third lactations. Swanson and co-workers in a series of papers (Swanson and Spann, 1954, Swanson, 1957, 1960, 1967, Swanson et al., 1967) concluded that fattened heifers or heavy feeding until first parturition will result in lower milk production for the first two lactations than that of normal and light fed animals. In their studies, the light fed heifers produced the most milk. Hansson (1956) also found that as the level of feeding until first parturition increased from 60 to 80, 100, 120, or 140 percent of the normal recommended level, the average yield of 4 percent fat corrected milk (FCM) for all lactations declined. Those heifers reared at the

60 percent feeding level were actually the best milk producers.

Crichton et al., (1960b) raised heifers at a high or low nutrition level for the first 44 weeks of age, at which time half of each group was switched to the other nutrition level. They found that feeding these rations until first parturition resulted in no differences in milk production among the groups over 3 lactations. This finding does not agree with Swanson's contention that overfeeding during the growth period results in a lowered level of production. But, it supports his finding that heifers reared on below standard feed levels milk just as well as control and heavy fed heifers. Reid et al., (1964) also found no differences in milk production during the first four lactations of cows reared on a low, medium, or high nutrition level until first parturition. Gardner and Garcia (1966) found that heifers fed for accelerated growth until conception produced about 2200 fewer pounds of milk than the controls during the first lactation, but both groups produced at the same level in the second lactation. These first lactation results could be a result of age at calving as discussed in the next section; the accelerated heifers were 19.7 months and the controls were 36.7 months old.

Reviews of this topic are presented by Burt (1956) and Schultz (1969).

6. Effect of Age at First Calving on Lactational Performance

The effect of age at first calving upon subsequent lactational performance has been studied for several years. Eckles (1915) was one of the first to make such a study. He found that Jerseys and Holsteins bred to calve at 20 to 24 months of age produced slightly less milk and butterfat in the first lactation than heifers bred to calve at 30 to 34

months of age. Turner (1932) examined official breed association records for age at first parturition and the subsequent production yield. He found an increase of about 1400 pounds in average yearly milk yield of Holstein heifers calving at 30 months of age as compared to 24 months. Delaying first calving until after 30 months of age resulted in practically no additional increase. However, Turner stated that because of the additional costs incurred by delaying first calving beyond 24 months of age, the most efficient milk production would be obtained by breeding heifers to calve at 20 to 24 months of age. Wickersham and Schultz (1963) noted that the average first lactation (305 day, 4% FCM) yields of heifers which calved at about 20, 24, and 28 months of age were not significantly different, although the oldest age group produced about 1500 pounds more milk. Hibbs and Conrad (1965) and Gardner and Garcia (1966) also found that heifers freshening at about 20 months of age produced less milk the first lactation than heifers which were about 27 months old at first calving. Thus, the findings of these researchers show most conclusively that as the age at first freshening increases up to about 30 months, the first lactation yield also increases.

However, in evaluating the effects of early calving on production, a truer picture is obtained if lifetime production is examined rather than production during the first lactation only. A 1953 English Milk Marketing Board study, as cited by Salisbury and VanDemark (1961), showed that after five lactations there was little difference in total milk production between heifers freshening for the first time at 24 or 36 months of age. Chapman and Dickerson (1936) and Hansson (1941, as cited by Salisbury and VanDemark, 1961) determined the amount of butterfat produced to a specified age and found the cows calving at an early age produced considerably more than those that calved at an older age.

And Salisbury and VanDemark (1961) presented lifetime milk production data showing that the later-calving cows never catch up with the earlier calvers in total milk produced to any particular age. From these studies it appears quite conclusive that although earlier calving heifers produce less milk their first lactation, total yield during their productive life, or to any specified age, will be greater than that for later calving heifers.

B. Reproductive Tract Development

1. Changes Associated with Nutrition and Age

Sorensen et al., (1959) slaughtered Holstein heifers at 1, 16, 32, 48, 64, and 80 weeks of age after they had been on either a low, medium, or high nutritional plane. The most striking changes in the reproductive organs were those that took place in the uterus at puberty. The weight of the uterus and the length of the oviducts, uterus, and vagina increased greatly at about the time of first estrus. These increases occurred between 16 and 32 weeks in the high plane heifers, 32 and 48 weeks in the medium plane heifers, and 48 and 64 weeks in the low plane heifers. Once estrous cycles were initiated, uterine growth continued at a slower rate in all groups. As expected, the degree of uterine epithelial development reflected the degree of sexual maturity. At a given age, the high plane heifers had the thickest endometrium and most endometrial glands, followed by the medium plane heifers and the low plane heifers. The height of the surface epithelium increased from approximately 14 to 36 microns at first estrus. Marked increases in ovarian weight occurred at about the time of first estrus in the heifers fed the high (8.7 mo.) and medium planes (11.4 mo.) of nutrition. Although mature ovarian follicles developed earlier and the onset of estrus and ovulation occurred earlier in heifers on the

high level of feeding, ovarian function after first estrus was not affected by age of heifers or their nutritional level.

Desjardins and Hafs (1969) slaughtered Holstein heifers at monthly intervals from birth through 12 months of age. They determined nucleic acids, protein, endometrial cell height, weight, and length of the tubular genitalia as indices of growth and function. Relative to the values at birth, uterine weight, ribonucleic acid (RNA), and protein increased more rapidly after 6 months than before this age. The relative increase in DNA to 10 months was only about two-thirds as great as the increases in uterine weight and RNA, suggesting hypertrophy of uterine cells concurrent with hyperplasia. Uterine epithelial cell height was stimulated at birth and then regressed. It did not return to the value at birth (20.9 microns) until 9 months of age (24.3 microns) but by 12 months had increased to 33.0 microns. The increase in endometrium thickness did not occur until about 2 months after first estrus and 3 months after changes in uterine weight, RNA, and protein content. Ovarian weight increased nearly four times more rapidly than body weight from birth to 5 months, but plateaued from 5 to 8 months. From 8 to 12 months of age, growth rate of the ovaries was comparable to that for the body. No follicles were visible on the ovaries at birth, but by 4 months of age the number of small and large follicles reached a maximum, after which it decreased to 8 months, and then remained relatively constant thereafter. Since stage of estrous cycle at slaughter was not constant in this experiment, it is possible that differences in stages of the estrous cycle among the age groups contributed considerable variation to the reproductive criteria observed.

2. Effects of Ovarian Steroids

Certain physiological effects of ovarian steroids on the reproductive processes are well established. Estrogens, secreted primarily by ovarian follicles, cause growth and vascularization of the uterus, while progesterone, secreted primarily by ovarian corpora lutea, promotes growth of the uterine endometrium and glands and suppresses estrous cycles. Hisaw and Hisaw (1961) discussed the effects of estrogens and progesterone on the reproductive tract.

The subject of estrous sychronization has been reviewed by Ulberg (1955), Hansel (1959), and Lamond (1964). The fact that estrous cycles in cattle can be regulated with progesterone was demonstrated by Ulberg et al., (1951) and Ulberg and Lindley (1960). They found that estrus and ovulation could be inhibited by daily injections of as little as 12.5 mg progesterone. Estrus occurred 2.5 to 9.5 days after the 14-day injection period. An injection of 0.5 to 10.0 mg of estradiol benzoate 3 days after the last injection of progesterone reduced the variation in the onset of estrus. Conception rate, however, was reduced by the progesterone injections with the higher dosages being more detrimental.

During the past 10 years orally active progesterone analogues have been developed since progesterone itself is inactivated when administered orally. Pincus and Merrill (1961) described some of the earliest work on oral progestagens developed to inhibit ovulation in women. The first synthetic progestagen studied quite extensively in cattle was medroxyprogesterone acetate (MAP) (Barnes et al., 1959, Hansel and Malven, 1960, Hansel et al., 1961, Hansel, 1961, Nelms and Combs, 1961, Zimbelman, 1961, Collins, 1961, Anderson et al., 1962, and Zimbelman, 1963). These studies showed that estrus and ovulation were inhibited during the oral administration period. After withdrawal,

heifers came into estrus, ovulated, and most of the studies showed conception rate to be nearly normal after cycle synchronization. Zimbelman (1963), however, reported a first service conception rate of 51 percent. After two services it was 76 percent while that of controls after two services was 74 percent.

Another compound which has received some attention is chlormadinone acetate (CAP). Wagner et al., (1963) and Van Blake et al., (1963) found this synthetic hormone to be extremely potent in inhibiting estrus and ovulation in cattle when fed for 15 to 20 days. Heifers came into estrus 4 to 6 days after the drug was withdrawn. Although conception rates were somewhat reduced at first service, the percent of heifers pregnant after two services was the same for treated and controls.

An orally active synthetic progestagen presently being studied quite extensively is melengestrol acetate (MGA). It is an analogue of medroxyprogesterone acetate with enhanced capacity to promote endometrial proliferation, maintain pregnancy, and delay estrus activity (Duncan et al., 1964). Zimbelman and Smith (1966a) found the minimal effective oral dose required to inhibit estrus and ovulation in cattle to be about 0.4 mg daily, while that for MAP was 180 mg orally daily (Zimbelman, 1963). For progesterone given subcutaneously, the minimal effective dose was 12.5 mg daily (Ulberg et al., 1951). These dosage level differences emphasize the potency of MGA. Zimbelman and Smith (1966a) reported that conception rate at first insemination averaged 42 percent for the various dose levels used, but after two services it was 82 percent. In other studies, Zimbelman and Smith (1966a) and O'Brien et al. (1968) found that 0.4 mg MGA daily for 18 days caused increased ovarian weights due to an increased incidence of a detectable follicle which increased in size with time on MGA.

Follicular fluid weight also increased, a finding also reported by Young et al. (1969). Zimbelman and Smith (1966a) concluded from the cervical mucous fern patterns and increased adrenal gland weights that the follicles were secreting estrogen even though estrus and ovulation were inhibited by MGA treatment. Zimbelman (1966) reported that MGA caused elevated pituitary luteinizing hormone (LH) content, suggesting that LH was not being released. However, there was no effect on follicle stimulating hormone (FSH) content in MGA fed heifers which would agree with the increased follicle size and inhibited LH release. This finding would seem to explain the increased incidence of large persistent follicles which do not ovulate in MGA fed cattle.

A search of the literature revealed only one study on the histology of the reproductive tract after progestagen administration. Smallwood and Sorensen (1969) administered MAP to heifers in an effort to determine some of the possible causes of lowered conception rate at first service. While they could make no definite conclusions, they noted that cystic follicles were found in several heifers and the surface epithelium of the uterus was separated from the stratum compactum in numerous cases. Perhaps these findings explain part or all of the lowered conception rate observed at first service after progestagen administration. Certainly additional study of this problem is needed.

C. Mammary Gland Development

1. Influence of Nutritional Level

Although Herman and Ragsdale (1946) did not measure the effect of nutrition on the mammary gland directly, they observed that the heavy fed heifers had a great deal of fat deposition in the udder before freshening. Swanson and his associates have studied this topic more extensively than

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anyone else (Swanson and Spann, 1954, Swanson, 1957, 1960, Swanson and Hinton, 1964, Swanson, 1967, Swanson et al., 1967). They conclude that fattening heifers causes large fat deposits in the mammary gland. The fat deposits, which are different from the normal fat pad, inhibit the development of the lobule-alveolar system, and in turn, lower the milk producing ability of the fattened heifers. Cross sections of udders from fattened animals showed incomplete development of the lobule-alveolar system. Sorensen et al., (1959) attempted to quantify the mammary development in heifers fed a low, medium, or high nutrient intake by using the method of Swett (1947). They found that udder development measured by this method was markedly affected by the level of feeding, with higher development ratings being associated with higher levels of feeding. The size of the excised mammary glands was closely related to development and feeding level at 16 weeks of age.

2. Changes Associated with Age

Presentations by Folley (1952), Cowie and Folley (1961), and Raymaud (1961) review mammary gland development during embyronic and fetal stages of life as well as growth of the mammary gland after birth. Based on gross observation and histology, it is generally accepted that the bovine mammary gland grows in size up to the time of puberty due to fat infiltration and ductular development. No appreciable lobule-alveolar growth occurs prepuberally. After puberty and especially during the last half of pregnancy, ductular and lobule-alveolar growth is greatly accelerated.

Only the study of Sinha and Tucker (1969) was found in which quantitative measurements were made of the changes in the bovine mammary gland with age. They measured changes in mammary gland weight, nucleic acids, lipid, and collagen between birth and 12 months of age, and

during the various stages of the estrous cycle in 16-month old heifers. Deoxyribonucleic acid content increased little between birth and 2 months of age. But between the second and third months, DNA content increased 15-fold and continued to increase almost linearly until 9 months of age. Between 9 and 12 months DNA content did not change. Mammary RNA and hydroxyproline (measure of collagen) followed patterns similar to mammary DNA from birth to 12 months of age. But, hydroxyproline values were more variable and the changes were not as great as for the nucleic acid changes. Mammary DNA and RNA values in the cycling 16-months old heifers were greatest on the day of estrus and lowest on day 20 of the cycle. Per 100 kg body weight, the mammary DNA value of 16-month old heifers was no greater than the value for 9- month old heifers, suggesting that a major portion of puberal mammary growth was largely completed by 9 months of age.

3. Effects of Steroids

The importance of ovarian hormones in growth of the mammary gland has been known and accepted for many years. The early work of Turner and coworkers (1939) clearly showed that estrogens stimulate duct growth, whereas a combination of estrogen and progesterone are needed for lobule-alveolar development. Following this initial report, numerous researchers attempted to develop the mammary gland and initiate lactation with exogenous hormones. Most investigators have used estrogen or a combination of estrogen and progesterone and have attained varying degrees of success (Turner, 1939, Folley, 1952, Cowie and Folley, 1961, Jacobsohn, 1961). Sud et al., (1968) for example, obtained mammary development in open heifers similar to that in 5- month pregnant heifers by injecting either 200 mg progesterone and 800 ug estradiol-17 Beta, or 100 mg progesterone and 400 ug estradiol-17 Beta

three times weekly for 20 weeks.

The effects of adrenal steroids on mammary development have been studied quite extensively in mice and rats (Jacobsohn, 1961). In general, most studies have shown that duct growth will result from low level adrenal steroid injections, but estrogens are usually necessary to obtain lobule-alveolar development. Kumaresan et al., (1967) injected corticosterone during pregnancy in rats and found a 23 percent increase in DNA and a 52 percent increase in RNA over that of pregnant controls, but subsequent lactational performance was not tested. Apparently no corticoid studies have been conducted on mammary gland development in cattle.

Mammary development has been observed, but never studied quantitatively, in several experiments involving diethylstibestrol administration (Casida et al., 1959). Also, at least one of the synthetic progestagens, melengestrol acetate, was observed to cause mammary development in cattle (Young et al., 1969). However, no studies were found which quantified the mammary development in cattle caused by the synthetic estrogens and progestagens.

D. The Adrenal Glands

1. Structure and Function

Structurally, the adrenal is a compound gland composed of an inner medulla and an outer cortex of different embryological origin (Turner, 1960). The medulla is ectodermal in origin and secretes amine hormones, while the cortex is derived from mesoderm and secretes steroid hormones. Neural innervation of the medulla regulates its secretions, but the cortex is practically devoid of nerves and is regulated by other body hormones. The cortex is composed of three zones; the zona glomerulosa,

zona fasciculata, and zona reticularis, from the exterior to the interior, respectively. The zona glomerulosa is little affected by hypophysectomy and secretes mineralocorticoids that are involved in regulating electrolyte metabolism. The fasciculata and reticularis are highly dependent on an insitu pituitary and secrete glucocorticoids which regulate carbohydrate metabolism.

Desjardins (1966) and Macmillan (1967) studied changes in adrenal weight and cortex zone widths from birth to 12 months of age in Holstein heifers and bulls. Weight of the paired adrenal glands increased linearly from birth to 10 months of age with only slight changes thereafter in both heifers and bulls. Differences in average adrenal weights for the two sexes were usually less than a gram. However, the width of the zona glomerulosa was consistently greater in bulls than heifers from 2 to 11 months of age. Although the combined width of the zonas fasciculata and reticularis was greater at most ages in bulls than in heifers, the proportional differences were not as great as the differences between the sexes in the width of the zona glomerulosa. The monthly increase in width of the zona glomerulosa in bulls was quite erratic, but showed a general increase from birth to 12 months of age. However, in heifers the glomerulosa width declined from birth to 6 months and then increased to a value at 12 months which was similar to the value at birth. The monthly increase in combined fasciculata-reticularis width was erratic in both heifers and bulls, but increased in both sexes by about 50 percent from birth to 12 months of age. Detailed discussions of the histological structure of the adrenal are presented by Elias (1948), Weber et al., (1950), and Nicander (1952).

2. Effects of Ovarian Hormones

It has been known for many years that ovarian hormones affect the

adrenal glands. Ellison and Burch (1936), for example, found increased adrenal weights and increased width of the fasciculata and reticularis zones of the cortex in animals that had received estrogen. The synthetic estrogens, notably diethylstilbestrol, also cause adrenal weight increase, as noted by Clegg and Cole (1954), Cahill et al., (1956), and Casida et al., (1959). Turner (1960) speculated that estrogens are acting by blocking the synthesis of adrenal corticoids. Since the lowered level of corticoids in the blood would act to stimulate increased pituitary adrenocorticotrophin (ACTH) release, adrenal hypertrophy would result. Guyton (1961) offered a different theory on the action of estrogens. He thought the estrogens were acting directly on the pituitary to cause ACTH release, rather than indirectly as Turner (1960) postulated. Both concur, though, that the pituitary is necessary to produce an estrogen effect on the adrenals.

At least one synthetic progestagen, melengestrol acetate, also affects the adrenals. Duncan et al., (1964) found that adrenal weights of male rats fed MGA decreased. But, Zimbelman and Smith (1966b) and Bloss et al., (1966) found that adrenal weights of intact heifers increased following long-term MGA administration. Obviously, more investigations are needed on the relationship between progestagens and the adrenals.

E. The Gonadotropins and Prolactin

1. Concentrations Associated with Age

The literature is almost devoid of studies on the changes in pituitary and blood concentrations of gonadotropins and prolactin with advancing age in the bovine. In 1935 Bates et al., reported the potency of pituitary prolactin in seven classes of cattle; embryos, veal calves, adult steers, adult bulls, and either open, early pregnant, or late pregnant cows.

Prolactin content was highest in embryos by two to three fold. Values for steers and bulls were greater than for nonpregnant cows, while the value for cows increased as the stage of pregnancy advanced. Reece and Turner (1937) also reported values for pituitary prolactin in cattle of various ages. They found that total pituitary content and the content per gram of anterior pituitary increased steadily with age in females. Values for bulls were generally lower than those for heifers, while lactating cows had higher values than dry cows. Desjardins et al., (1966) found similar prolactin concentrations in five 12-month old Holstein heifers and 42 nonpregnant mature Holstein cows. Sinha and Tucker (1969) reported that pituitary prolactin values showed no significant differences between birth and 12 months of age in Holstein heifers killed at monthly intervals. However, the values were somewhat elevated at 3 and 4,and 8 and 9 months of age. These elevated values at 3 and 4 months corresponded to the times when there was a shift to an increased rate of mammary growth, and the latter increase occurred when mammary growth was the greatest.

Bates et al., (1935) found that FSH potency was the lowest in steers, about 25 percent greater in embryos, bulls, and nonpregnant cows, 40 percent greater in veal calves and cows in late pregnancy, and 75 percent greater in early pregnant cows than in steers. From a recent study of Holstein heifers from birth to 12 months of age, Desjardins and Hafs (1968) found that anterior pituitary concentration of FSH was greatest at 1 month of age; it declined at 2 months and fluctuated only slightly thereafter to 12 months of age. Pituitary LH concentration was considerably greater than that for FSH. LH levels increased rapidly from 1 to 3 months, fluctuated considerably between 3 and 6 months, had a greatly increased peak value at 7 months, and then fluctuated at prepeak values

from 8 through 12 months. The peak at 7 months occurred at the time of puberty in the heifers.

Hackett and Hafs (1969) measured pituitary FSH and LH at various stages of the estrous cycle in 16-month old heifers. Averaging all values of the cycle showed FSH concentration to be only about one-fourth and LH about one-third the values found by Desjardins and Hafs (1968) in 12-month old heifers. Desjardins et al., (1966) also measured pituitary LH and FSH levels in 42 nonpregnant mature cows. The level of pituitary LH which they found was only one-third that reported by Desjardins and Hafs (1968) in heifers at 12 months of age. In contrast, pituitary FSH concentration was similar in the 12-month old heifers and cows.

2, Effects of Ovarian Steroids

While an interaction of the estrogens and progesterone with pituitary gonadotropins and prolactin definitely exists, knowledge of specific action is far from complete. One fairly definite fact is that the estrogens cause an increase in content and release of pituitary prolactin (Meites, 1966). Beyond this, the relationships are much more conditional. The effects obtained in various experiments have been dependent, among other things, on the dosage levels of estrogens and progesterone, length of the injection period, age of test animals, and the species used in the studies. Thus, results have varied from an inhibitory effect, to no effect, to a stimulatory effect (Greep, 1961, and Flerko, 1966). Consequently, only a general scheme of the mechanism of interaction thought to exist between the ovarian hormones and the pituitary gonadotropins and prolactin are presented. According to current concepts, a feedback mechanism operates whereby the pituitary release of FSH and LH is controlled by the levels of estrogen and progesterone in the circulation (Turner, 1960). Very low levels of

estrogens, coming from the immature follicles or extragonadal sources, stimulate the pituitary (probably by way of the hypothalamus) to augment its release of FSH. When the blood estrogen level becomes high, indicating that the ovarian follicle is mature, it acts to inhibit further FSH release and promotes an increase in the rate of LH release (cyclic LH release) above its usual continuous secretion level. Apparently this continual release of LH is needed along with FSH for significant estrogen production by the follicles. Under the influence of rising LH titers, the follicle matures, lutein changes occur in the walls of the mature follicle, and some progesterone along with large quantities of estrogens are secreted. The increasing levels of LH are in some manner involved in promoting ovulation. Once ovulation occurs, there is an immediate fall in the level of circulating estrogens. The ruptured follicle becomes transformed into a corpus luteum either spontaneously, or under the influence of LH or prolactin, depending on the species, and commences to secrete progesterone. Further release of LH above the base line secretion level is prevented by the high levels of progesterone. FSH, however, is released in quantities sufficient to cause follicle growth midway through the cycle. Although estrogen is secreted by these follicles, since the follicles do not grow to the mature ovulatory size because LH is lacking, estrogen production is not great enough to promote the behavioral signs of estrus. Since the level of LH is insufficient to promote follicle maturation and ovulation, the follicle regresses in size and becomes atretic. Progesterone level remains high until the corpus luteum begins degenerating in function and structure. When this occurs, cyclic LH release is again possible and the estrous cycle is repeated (Turner, 1960).

Even though the advent of synthetic estrogens such as diethylstil-

bestrol occurred several years ago, their effect on pituitary gonadotropins and the hypothalamus remains unknown. But, this void of knowledge with regard to the progestagen, melengestrol acetate, has been filled by the findings of Zimbelman (1966). He found no consistent effect of MGA on pituitary FSH, and therefore concluded that the corpus luteum was more effective than MGA in the control of follicular development. In intact pregnant heifers, MGA caused an increase in pituitary LH which was interpreted to mean that MGA inhibited LH release. However, this effect of MGA on pituitary LH was not evident in either bilaterally ovariectomized heifers with a low LH content or in unilaterally ovariectomized heifers with increased follicular development in the absence of a corpus luteum. Zimbelman (1966) concluded that these results are consistent with the concept of two hypothalamic centers for LH release, but only the center controlling cyclic LH release appears affected by MGA. Furthermore, it would seem necessary to conclude that a low level of LH release occurs during MGA treatment to allow enlarged follicle development and estrogen production. Additional studies are required to confirm and further clarify the effects of progestagens on the pituitary and hypothalamus.

MATERIALS AND METHODS

A. Experimental Design

This experiment was designed to determine the effects of two levels of nutrition fed alone and in combination with the synthetic progestagen melengestrol acetate (MGA), during the period of most rapid postnatal development in Holstein heifers. Parameters measured were body growth, levels of certain anterior pituitary hormones in the pituitary and blood, development of the reproductive tract and mammary gland, and subsequent reproductive and lactational performance.

For the study 140 heifers were purchased when less than 2 weeks old. They were fed and handled similarly for the first 2.5 months to be assured they were all growing well. According to previous randomized assignments, at 2.5 months of age the heifers were divided into 14 groups of 10 heifers each. A designation of the treatments each group received is shown in Table 1. Two levels of nutrition were fed (Tables 2 and 3); a normal level designed to allow for a normal growth rate, and a high level formulated and fed to promote a maximal growth rate. Besides designating the nutritional level, the group to which each heifer was assigned also determined if and when she would receive MGA, and if and when she would be slaughtered. MGA was fed at the rate of 0.30 mg per heifer per day starting at 2.5 months of age or after first estrus to determine its prepuberal versus only postpuberal effects.

One hundred heifers in designated groups were slaughtered either at 2.5 months of age, first estrus or at breeding size to obtain measure-

TABLE 1.--Experimental treatments beginning at 2.5 months of age for 140 Holstein heifers.

Group[a]	Nutrition treatments		Age at slaughter
	Level	MGA	
1	Normal	None	-[b]
2	High	None	-
3	High	From 2.5 mo.	-
4	High	From first estrus	-
5	Normal	None	Breeding size
6	High	None	Breeding size
7	High	From 2.5 mo.	Breeding size
8	High	From first estrus	Breeding size
9	Normal	From 2.5 mo.	Breeding size
10	Normal	None	First estrus
11	High	None	First estrus
12[c]	High	From 2.5 mo.	First estrus
13[d]	High	From first estrus	First estrus
14	Normal	None	2.5 mo.

[a]Each group contained 10 heifers.

[b]Heifers in groups 1-4 were not slaughtered. They were bred and retained for reproductive, dystocia, and subsequent lactational performance studies.

[c]Heifers were slaughtered when their group 11 pairmates were slaughtered.

[d]Group included for statistical balance. Received same ration as group 11.

TABLE 2.--Description of nutrition levels.

Normal level	High level
(Per day)	(Per day)
0.9 kg of a 12% protein grain mix	free choice to a maximum of 4.5 kg per day of a 20% protein grain mix
Free choice for both levels: (1) Corn silage (2) Alfalfa-grass hay (3) Trace mineralized salt	

TABLE 3.--Composition of experimental grain mixes.

Ingredient	Amount per 100 kg	
	12% protein mix	20% protein mix
Ground shelled corn (kg)	83.3	61.7
Soybean meal (50% protein) (kg)	9.7	29.3
Molasses (kg)	5	7
Dicalcium phosphate (kg)	1	1
Trace mineralized salt (kg)	1	1
Vitamin A (IU)	660,000	660,000
Vitamin D (IU)	880,000	880,000
Auromycin (mg)	22,000	4,400

ments of the various parameters at those physiological ages. Forty heifers were kept to obtain data on reproductive efficiency, dystocia, and lactational performance.

To obtain experimental design balance to the groups slaughtered at first estrus, a group (Group 13) destined to receive the high nutritional level without MGA until first estrus and then MGA in addition thereafter was included. However, since this groups was slaughtered at first estrus, they never received MGA and consequently received the same feeding treatment up to slaughter as the heifers fed the high level without MGA (Group 11). In analyzing the results, data from these two groups were combined.

Because heifers fed MGA would be in a proestrus hormonal condition when they were slaughtered 48 hours after MGA withdrawal, heifers that had not been fed MGA were slaughtered 17 to 20 days after an estrus so they would be in a similar hormonal condition. Since heifers fed MGA from 2.5 months of age were not expected to exhibit estrous cycles, such heifers fed the high nutritional level plus MGA and scheduled for slaughter at first estrus were paired by body weight at 2 weeks of age with heifers that were to be fed the high nutritional level without MGA. Thus, when a heifer fed the high level without MGA was slaughtered after her first estrus, her high level pair-mate fed MGA was also slaughtered.

To measure growth rate, height at the withers and body weight were taken once a month in the morning before the heifers were fed. Height and weight were also recorded on the day before slaughter and at parturition. The criterion for breeding size was that heifers be 120 cm tall at the withers. After a heifer had reached 118 cm at the withers, height measurements were taken every 2 weeks to obtain a more precise estimate of

the date she reached 120 cm at the withers.

In each of the groups kept for breeding, five randomly selected heifers were bred artificially to Zeldenrust Royal Pontiac, registration number 1397753, and 5 to Wis Symbol, registration number 1189593. These two bulls were used since a previous study by Boyd and Hafs (1965) had shown that Pontiac sired calves that weighed about 5 kg more at birth than those sired by Symbol, and we wished to determine if this difference in calf size at birth would be reflected in the degree of dystocia encountered by the dams.

B. Management of Experimental Animals

Holstein heifers from production tested dams and registered sires were purchased when less than 2 weeks old from dairymen near Madison, Wisconsin. They were transported by truck to the M.S.U. dairy barn, and were examined and treated if necessary by a veterinarian upon arrival. Individual health record sheets were kept for each heifer. They were weighed on the second day after arrival. Calves were purchased in two lots of 40 and two lots of 30. Lot 1 arrived on April 12, 1967; lot 2 on July 8, 1967; lot 3 on September 27, 1967; and lot 4 on April 12, 1968. Heifers in lots 1 and 2 were randomly assigned at 5 per group to groups 1 - 8. Those in lots 3 and 4 were, likewise, assigned to groups 9 - 14 (see Table 1). From arrival until 2.5 months of age, the calves were kept in individual 4' by 6' pens. By 3 weeks of age, the heifers received 8 kg whole milk per day and were fed water, excellent quality alfalfa hay, and a 16 percent protein grain mixture free choice. Grain and hay consumption increased gradually, so that by 2.5 months, when switched to the nutritional treatments, the heifers were consuming about 2.3 kg of grain and 0.9 kg of hay per day. At 2.5 months of age

the heifers were moved to loose housing dry lot facilities and penned communally according to the nutritional treatments.

Commencing when the heifers were about 5.0 months old, they were observed for estrus signs twice daily at approximately 8:00 a.m. and 5:00 p.m. Any proestrus signs such as mucous discharge from the vulva, or bawling and general restlessness, as well as the often observed post-estrus bleeding were recorded. A heifer was recorded in estrus when she would stand to be mounted by other heifers. She was also considered in estrus if she would not stand but displayed estrus symptoms such as swollen and inflamed vulva, attempted to mount other heifers, and general uneasiness.

Starting at 6 months of age the ovaries of the heifers were palpated per rectum each month to detect corpora lutea, as evidence of ovulation. Unexpectedly, some heifers receiving 0.30 mg MGA per day showed signs of ovulation as per rectal palpation. When this happened, each heifer in the entire group was then increased to 0.45 mg daily. Heifers fed MGA from 2.5 months of age and slaughtered at first estrus received 0.30 mg daily for about 3.5 months, and the 0.45 mg level for about 0.6 months. Heifers that had been fed MGA from 2.5 months of age to breeding size received the lower dose for about 5.4 months and the higher dose for about 4.2 months. Meanwhile, those heifers fed MGA only after first estrus received the 0.30 mg dose for about 0.8 months and the 0.45 mg dose for about 3.8 months.

When heifers receiving MGA reached breeding size the drug was withdrawn from their ration and they were fed only the high nutrition level. Heifers that were not slaughtered at breeding size were bred artificially in the late afternoon if first observed in estrus that morning, and in the morning of the following day if first observed in

estrus in the afternoon. Heifers on the high level of nutrition were switched to the normal level when they were diagnosed pregnant by palpation, i.e. about 50 - 60 days after conception.

As the experiment progressed, it became obvious that conception rate was lower than expected. Therefore, heifers on the high nutritional level which failed to conceive by the fifth service were switched to the normal level on the day of the fifth service to prevent excessive fattening. A heifer was bred a maximum of 10 times to the bull she was previously assigned, and if still not pregnant she was bred an eleventh time to a different bull. Any heifer that did not become pregnant to the eleventh service was slaughtered and the reproductive tract examined macroscopically and microscopically to determine possible causes for the infertility.

Pregnant heifers were sold to Driggs Dairy at Palmyra, Michigan, where they were kept in a dry lot loose housing barn along with the regular herd heifers through the remainder of pregnancy. They received corn silage and hay free choice. After parturition they were placed in a free stall barn with the regular milking herd and milked in a parlor. Corn silage, alfalfa haylage, and alfalfa-grass hay were fed free choice, and either 2.3 or 7 kg of a 14 percent protein grain mixture per day was fed each cow, depending on her level of milk production. The heifers were weighed and measured at the withers about 10 days before the expected day of calving and within 3 days after calving. At parturition, weight and sex of the calf, health and condition of the dam and calf and a subjective rating of dystocia were recorded. Dystocia was rated from 1 to 4 according to increasing degree of difficulty at parturition. A rating of 1 indicated the heifer had a normal delivery; 2 indicated the heifer encountered a more than normal amount of straining but did

not require assistance; 3 indicated that assistance was required to deliver the calf; and 4 indicated the calf was born dead due to the difficult delivery, or that a Cesarean section was required to deliver the calf. Daily milk weights for the first 60 days of lactation were used to estimate each heifer's milk producing ability.

C. Slaughter Procedures

On the day of slaughter, heifers were transported about 8 miles to the Van Alstine Packing Company near Okemos, Michigan at approximately 6:30 a.m. Usually, slaughter began at about 7:00 a.m., and was completed by 8:30 a.m.

The procedure followed at slaughter was as follows:

(1) On the afternoon prior to slaughter each heifer was weighed and height at withers recorded. A sample of urine, usually less than 500 ml, was collected and stored at -15°C. until analyzed for estrogens.
(2) At slaughter each animal was stunned in the forehead with a captive-bolt gun and exsanguinated. Two litres of mixed venous and arterial blood were collected in a cold heparinized glass jar and immediately stored at 4°C. It was later centrifuged and the plasma frozen in 10 ml samples for hormonal analyses.
(3) Within 10 minutes after stunning, the top of the skull was sawed off, the brain displaced, and the pituitary and hypothalamus dissected free. The whole pituitary was weighed and then the two lobes were weighed separately. The anterior pituitary was placed in a polyethylene bag on Dry Ice. The hypothalamus, median eminence and pituitary stalk were diced, immersed in a minimum volume of 0.1N hydrochloric acid and placed on Dry Ice. Both the anterior pituitary and the hypothalamus were stored at -15°C. until analyzed for hormone content and releasing factors, respectively.
(4) The mammary gland was removed and halved down the medial suspensory ligament. A representative sample of the right rear quarter was placed in Bouin's fixative for subsequent histological examination. The two halves were wrapped separately in heavy paper and within one and one half hours were stored at -15°C. for later nucleic acid analyses.
(5) The reproductive tract was removed and the ovaries were dissected from the rest of the tract and weighed. Follicles were measured for surface diameter and recorded as ranging from 4-9mm, 10-15mm, 16-20mm,>20mm in size. Size of corpora lutea, presence of ovulation points and any other noteworthy observations were also recorded. The ovary containing the largest follicle was bisected through the follicle. A section of the follicle wall was placed in cold 2 %

glutaraldehyde fixative for 4 hours and then stored at 4°C. in 0.1M phosphate buffer until further preparation for electron microscopy studies. After dissecting the uterus from the rest of the tract, it was weighed. A section of the right horn was put in Bouin's fixative for histological examination, and a 20- to 30-gram piece from the left horn was placed in 0.25 M sucrose and put on Dry Ice until stored at -15°C. for later nucleic acid analyses.

(6) The adrenal and thyroid glands were removed, weighed, and a sample of each placed in Bouin's fixative for later histological examination. The adrenal section was taken midway down the lobe on the right adrenal gland. The remaining adrenal tissue was put in 0.25 M sucrose, held on Dry Ice and later stored at -15°C. until analyzed for corticoid content.

D. Assays of Anterior Pituitary Hormones

1. Homogenization of the Anterior Pituitaries

The anterior pituitaries which had been stored at -15°C. were partially thawed at room temperature, weighed to the nearest 0.1 mg, diced, and homogenized in a Servall Omni-mixer in 10 ml of cold 0.85% saline for about 2 minutes. The volume of the homogenate was adjusted to a final concentration of 50 mg anterior pituitary equivalent per ml. The homogenate was centrifuged in a Servall Superspeed centrifuge type SS-1 for 15 minutes and the supernatant fluid frozen in plastic vials for later FSH, LH, GH, and prolactin analyses.

2. Follicle Stimulating Hormone (FSH) Bioassay

Anterior pituitary FSH content was measured by the immature rat ovarian weight augmentation assay of Steelman and Pohley (1953). Because bovine pituitaries contain little FSH relative to LH or relative to pituitary FSH potencies in other species (Macmillan, 1967), doses of 40 and 80 mg equivalents of anterior pituitary tissue were used whenever possible. Sixteen pituitaries had to be assayed using doses of 30 and 60 mg equivalents, and 6 pituitaries using 25 and 50 mg equivalents due to a lack of pituitary material. The 40-80 mg level unknowns were compared to 40 and 80 μg levels of ovine NIH-FSH-S5, shile the 30-60 and 25-50 mg

level unknowns were compared to 30 and 60 ug levels of ovine NIH-FSH-S5. The unknowns were also compared to a 20 IU dose of human chorionic gonadotropin (Squibb Follutein Chorionic Gonadotropin).

Female Sprague-Dawley rats (from Spartan Research Animals, Haslett, Michigan) were injected subcutaneously between 7 and 8 a.m., 12 and 1 p.m., and 5 and 6 p.m. on days 22,23, and 24 of age. Besides receiving either the unknown or known amounts of FSH, each rat was also injected with 20 IU of human chorionic gonadotropin. For each assay, five female rats were used at each unknown dose level and seven rats for each standard dose level. The total dose was given over a three day period in 9 injections. Between 8 and 11 a.m. on day 25 of age, the rats were killed, both ovaries removed, trimmed, and weighed. Later, the potencies were estimated by the slope ratio procedure of Bliss (1952).

3. Prolactin Radioimmunoassay

Plasma and anterior pituitary prolactin levels were measured by a radioimmunoassay technique developed by Dr. H. A. Tucker and J. A. Koprowski from our laboratory. The methods were essentially those of Niswender et al., (1968, 1969a, 1969b). Briefly, the procedure consisted of the following steps:

(1) Antibodies to NIH-B_1 prolactin were prepared in guinea pigs by emulsifying approximately 2 mg of hormone in 2 ml of 0.85 percent NaCl and 2 ml of Freund's complete adjuvant. Each guinea pig received this mixture subcutaneously once every 2-3 weeks except that incomplete adjuvant was used after the initial injection. Serum was collected via heart puncture at 2- to 3-week intervals after the ninth week.
(2) A sheep (wether) was immunized with guinea pig gamma globulin. Forty to 50 mg gamma globulin (Fraction II, Pentex Inc., Kankakee, Illinois) were emulsified in 2.5 ml 0.85 percent NaCl and 2.5 ml Freund's complete adjuvant. Subcutaneous injections of gamma globulin emulsified in incomplete adjuvant were continued at 3-week intervals for 9 weeks and serum was collected at monthly intervals thereafter.
(3) The methods used in radioiodination and in the radio-immunoassays were essentially those of Niswender et al., (1969b) except that ^{125}I was used.

(4) Cross reactivity of the prolactin antibody with other hormones was checked against NIH-bovine LH, TSH, GH, and ovine FSH. Positive responses did not occur at levels up to 100 mug of GH per tube and up to 1000 mug of each of the other three hormones per tube. Minimal and maximal amounts of prolactin actually measured on a per tube basis were 0.2 and 4 mug. Thus, the assay was specific at the levels used. The immunoassay produced parallel dose response curves between the standards and bovine sera or pituitary extracts.
(5) The plasma samples were diluted 1:6 while the anterior pituitary homogenates were diluted 1:500 with 1 percent bovine serum albumin in phosphate buffered saline.

4. Luteinizing Hormone (LH) Radioimmunoassay

Anterior pituitary and blood plasma levels of LH were determined by the radioimmunoassay procedure of Niswender et al., (1969b), employing a few minor changes. It was essentially the same basic procedure described for the prolactin radioimmunoassay. While the plasma samples were not diluted, the pituitary homogenates were diluted 1:7500 with phosphate buffered saline containing 1 percent egg white albumin. Those changes in the procedure included:

(1) using ^{125}I for radioiodination

(2) using purified bovine LH (LER 1072-2)

(3) using goat anti-rabbit gamma globulin from Nutritional Biochemicals Corporation as the second antibody

Drs. Niswender and Midgley (1968) supplied the LH antiserum and the purified bovine LH.

E. Determination of Nucleic Acids in the Uterus and Mammary Gland

The nucleic acid content of the uteri and mammary glands was determined using the procedure of Schmidt and Thannhauser (1945) as modified by Tucker (1964). An outline of the procedure was:

(1) If tissue frozen, thaw at room temperature.
(2) Dissect mammary parenchymal tissue from hide and fat and weigh it.
(3) Grind parenchymal tissue with a meat grinder to obtain a representative sample for analyses.
(4) Dice the uterus to obtain a representative sample for analyses.

(5) Weigh out 15 to 20 grams of the tissue.
(6) Suspend in cold distilled water at a 1:20 dilution.
(7) Homogenize for 2 minutes in a Waring blender at top speed.
(8) Put duplicate 2 ml samples into 16 ml plastic centrifuge tubes, add 10 ml 95% ethyl alcohol, stopper, and shake for 12-18 hours.
(9) Centrifuge at 17,000 rpm for 20-30 minutes in a Sorvall with a SM-24 rotor and discard supernatant.
(10) Add 5 ml anhydrous ether, centrifuge for 10-15 minutes, and discard supernatant.
(11) Dry tubes until tissue forms a hard pellet.
(12) Add 10 ml methanol:chloroform (2:1), stopper, and shake for 18-24 hours.
(13) Centrifuge for 10-15 minutes and discard supernatant.
(14) Add 10 ml anhydrous ether, stopper, and shake for 18-24 hours
(15) Centrifuge for 10-15 minutes and discard supernatant.
(16) For steps 16 and 17, keep tubes in ice water when not in centrifuge. Add 5 ml ice-cold 10% trichloracetic acid, mix well, centrifuge for 5-10 minutes, and discard supernatant. Repeat this step.
(17) Add 5 ml ice-cold ethanol saturated with sodium acetate, mix well, centrifuge for 5-10 minutes, and discard supernatant.
(18) Add 2 ml 1N potassium hydroxide, mix well, stopper, and store at 37°C. for 15 hours.
(19) Cool tubes in ice-water, add 0.3 ml ice-cold 6N hydrochloric acid and 5 ml ice-cold 10% perchloric acid, mix well, centrifuge for 10-15 minutes, pour supernatant into 25 ml calibrated test tube.
(20) Add 5 ml ice-cold 5% perchloric acid, mix well, centrifuge for 10-15 minutes, pour supernatant into 25 ml calibrated test tube from step 19. Repeat this step. Keep all tubes cold.
(21) Bring volume of calibrated test tube up to 20 ml with 5% perchloric acid, mix well, take 3 ml and combine with 3 ml fresh (<1 hour old) orcinal reagent.
(22) Cap tube with a marble and heat in a water bath for 30 minutes at 100°C. Allow to cool.
(23) Read optical density at 670 mu on a Beckman DB spectrophotometer. Adjust to 0 optical density with a mixture of 3 ml 5% perchloric acid and 3 ml orcinal reagent that has been heated to 100°C. for 30 minutes. The optical densities obtained are compared with a standard curve prepared with highly purified yeast RNA.
(24) To the precipitate in step 20, add 5 ml ice-cold 5% perchloric acid, mix well, heat in a water bath for 15 minutes at 70°C., cool to 5°C., centrifuge for 10-15 minutes, pour supernatant into 25 ml calibrated test tube.
(25) Add 5 ml ice-cold 5% perchloric acid to precipitate, mix well, centrifuge for 10-15 minutes, pour supernatant into calibrated test tube from step 24. Repeat this step.
(26) Bring volume of calibrated test tube up to 25 ml with 5% perchloric acid, mix well, and read optical density at 268 mu on a Beckman DB spectrophotometer. Adjust to 0 optical density with 5% perchloric acid. The optical densities obtained are compared with a standard curve prepared with highly polymerized DNA.

To convert the optical density values obtained into mg of DNA and RNA in the entire mammary gland or uterus, the following mathematical steps were

used:

$$\text{Slope of the standard curve} \times \text{Final diluted volume} \times \text{Optical density} \times \text{Total weight of the organ} \times \frac{1}{100}$$

F. Histological Technique

After fixed in Bouin's fluid for 48 or more hours, the adrenal and uterine tissues were cleared by placing them in the following solutions in the order listed: 50, 70, 80, and 95 percent ethanol, 95 percent ethanol: methyl salicylate (1:1), and methyl salicylate. Tissues were left in each solution for at least 24 hours. Infiltrating the tissues with melted paraffin followed, with the tissues being in the paraffin baths for at least 3 hours (1 and 1/2 hours in each of two baths). The tissues were then transferred to molds and imbedded in paraffin blocks. Sectioning the tissue blocks into 8- to 10-micron sections with a microtome, and placing the sections on a warmed glass slide coated with egg white albumin followed. After drying for 48 hours, the slides were stained using the following steps: 15 min. in Xylene; 5 min. in 95% EtOH; 5 min. in 95% EtOH; 5 min. in 70% EtOH; 5 min. in 35% EtOH; 5 to 20 minutes in Harris' Hematosylin depending on the type of tissue; 2 minutes in distilled water; 5 min. in 35% EtOH; 5 min. in 70% EtOH; a few seconds in acidulated 70% EtOH if over stained; 15 to 45 seconds in alkalinated 70% EtOH if desire greater differentiation; 2 to 3 min. in Eosin; 5 to 10 minutes in 95% EtOH; 10 min. in xylene; put a few drops of Permount on the slide, add a cover slip and allow to dry.

The uterine slides were examined at 400 power and the endometrial cell height measured with a calibrated occular micrometer. Nearly all the adrenal slides contained complete cross sections. These were projected by means of a Bioscope with a 2 power ocular onto paper on a table 122 cm

below the slide. Tracing the medulla and the three cortex zones on the paper was quite easy with this apparatus. Ten locations around the medulla on the tracing from each slide which typified the cortex structure were selected. Lines perpendicular to the medulla were drawn and the cortex zone widths measured at these locations. The values obtained were then corrected for the distance projected and converted into millimeters.

RESULTS AND DISCUSSION

A. Body Growth

1. From 2 Weeks to 2.5 Months

Group means for the body measurements taken at 2 weeks and at 2.5 months of age are presented in Table 4. The overall mean and its standard error for weight at 2 weeks of age was 46 $\pm$ 1 kg. At 2.5 months it was 96 $\pm$ 1 kg and the withers height was 86.6 $\pm$ 0.3 cm. Although variation in weight existed among the group values at each age, all heifers started on the treatments at 2.5 months of age at about the same weight and height.

2. From 2.5 Months to First Estrus

That the different levels of nutrition fed without MGA influenced body growth up to the time of first estrus is indicated by the data in Table 5 and Appendix I. At first estrus, the 30 normal level heifers were 8.7 $\pm$ 0.2 months old with a range of 6.0 - 11.5 months, while the 60 high level heifers were 7.5 $\pm$ 0.1 months old with a range of 5.7 - 10.1 months. The age and body size values recorded at the estimated time of first estrus for the 30 heifers fed the high level plus MGA from 2.5 months are shown in Table 5 and will be discussed later.

The ages at first estrus for the normal and high level heifers were significantly different ($P<0.01$) and agree with the findings of previous studies that heifers fed a higher than normal nutritional level will be younger at first estrus.

TABLE 4.--Growth from 2 weeks to 2.5 months of age for Holstein heifers.

Group[a]	Weight		Withers height
	2 weeks	2.5 months	2.5 months
	----------(kg)----------		(cm)
1	50	97	86.2
2	48	95	86.6
3	47	96	86.6
4	46	99	84.1
5	48	98	86.8
6	49	101	86.8
7	48	92	87.7
8	47	98	86.1
9	45	97	89.2
10	42	90	85.0
11	41	93	86.9
12	44	96	86.8
13	44	94	88.1
14	52	95	85.9
Mean ± SE	46 ± 1	96 ± 1	86.6 ± 0.3

[a]Ten heifers in each group.

TABLE 5.--Growth to first estrus for Holstein heifers fed different nutrition treatments.[a]

Nutrition treatment	Number heifers	First estrus			From 2.5 months to first estrus	
		Age	Weight	Withers height	Daily gain	Increase in withers height
		(mo)	(kg)	(cm)	(kg)	(cm)
Normal	30	8.7 ± 0.2[c]	250 ± 5	109.2 ± 0.7	0.83[c]	23.2 ± 0.9
High	60	7.5 ± 0.1	255 ± 4	108.6 ± 0.6	1.08	22.1 ± 0.6
High + MGA from 2.5 mo.[b]	30	7.4 ± 0.2	263 ± 10	107.9 ± 1.0	1.11	20.8 ± 0.9

[a]Values are means and their standard errors.

[b]Values at first estrus of high treatment pairmates.

[c]Significantly different from the other values ($P<0.01$).

However, the ages in this study were younger than those in most of the previous studies cited in the literature review (Eckles, 1915, Reed et al., 1924, Hansson, 1956, Crichton et al., 1959, Sorensen et al., 1959, Gardner and Garcia, 1966). A multitude of factors such as climate, inheritance, improved rations or better methods of detecting estrus could have caused the age differences between this and previous studies. The heifers in this study had earlier communal contact in loose housing pens than heifers in some of the previous studies which may have prompted the development of estrus behavioral patterns at an earlier age.

Although age at first estrus was different for heifers fed the two levels of nutrition, body weight and height at withers (Table 5) did not differ significantly at first estrus ($P>0.10$). These findings substantiate the contention that heifers exhibit first estrus at a relatively constant physiological age, as indicated by body size, rather than at a certain calendar age. That heifers fed the normal level of nutrition were significantly older but not heavier or taller at first estrus implies that the high level heifers grew faster than the normal level heifers from 2.5 months to first estrus. This in fact did occur as indicated by the nonsignificant difference ($P>0.10$) in withers height increase and the significant difference ($P>0.01$) in daily gain from 2.5 months to first estrus for heifers fed the two levels of nutrition without MGA (Table 5). The normal level heifers gained 0.83 kg per day which is very close to the value of 0.87 kg for daily gain by Holstein heifers from 70 to 260 days of age as reported by Matthews and Fohrman (1954) in the Beltsville growth standards. Morrison (1959) gives values in his growth standards which calculate to be 0.84 kg gain per day between 2.5 and 8.7 months of age. Thus, by these two standards the normal level heifers in this study grew at a normal rate up to first estrus.

Also presented in Table 5 are growth values for the 30 heifers receiving the high level plus MGA from 2.5 months. Since the progestagen inhibited estrous cycles in these heifers, the values shown in Table 5 were taken when the high level without MGA pairmates exhibited their first estrus. Thus, these data are estimations of relative values which might have existed at first estrus and are not absolute values. Heifers fed the normal level plus MGA from 2.5 months did not have contemporary normal level without MGA pairmates, so estimated first estrus values were not available. Although the high level plus MGA values in Table 5 for weight and daily gain were not significantly different ($P > 0.10$) from the high level without MGA values, analysis of the data on a monthly basis revealed that after 5.5 months of age the heifers fed MGA gained significantly faster ($P < 0.05$) than those not receiving the drug. This finding supports the contention of Zimbelman (1968), that MGA causes greater weight gains beginning 1 to 2 months before first estrus. Since MGA apparently increases weight gains through the action of the estrogens from the persistent ovarian follicles (Zimbelman and Smith, 1966b), this action implies that the ovaries commence a certain degree of activity before first estrus. I think this is probably an acceptable explanation, as puberty evolves over a period of time rather than occurring suddenly (Donovan and van der Werff ten Bosch, 1965).

Data extracted from Table 5 for the heifers that were not slaughtered at first estrus are shown in Table 6 and Appendix I. Although combined with the high level values in Table 5, the data for heifers fed the high level up to first estrus but designated to also receive MGA after first estrus are presented separately in this table. Though

TABLE 6.--Growth to first estrus for Holstein heifers fed different nutrition treatments but not slaughtered at first estrus.[a]

Nutrition treatment	Number heifers	First estrus			From 2.5 months to first estrus		
		Age	Weight	Withers height	Daily gain	Increase in withers height	Change in weight
		(mo)	(kg)	(cm)	(kg)	(cm)	(kg)
Normal	20	9.1 ± 0.3[c]	250 ± 7	109.6 ± 1.0	0.79[c]	23.1 ± 1.2	154 ± 7
High	20	8.0 ± 0.3	265 ± 8	109.3 ± 1.3	1.04	22.6 ± 1.1	169 ± 8
High + MGA from 2.5 mo.[b]	20	7.8 ± 0.3	279 ± 14	108.7 ± 1.5	1.10	21.5 ± 1.4	179 ± 12
High + MGA from first estrus	20	7.7 ± 0.2	260 ± 4	107.7 ± 0.5	1.08	22.6 ± 1.0	165 ± 6

[a]Values are means and their standard errors.

[b]Values at first estrus of high treatment pairmates.

[c]Significantly different from the other values ($P<0.01$).

the values in Table 6 are slightly different from those in Table 5, the general findings can be interpreted similarly.

3. From First Estrus to Breeding Size

Ages of the heifers when they reached 120 cm at the withers (breeding size) are most interesting. As Table 7 and Appendix I show, heifers fed the high level or the high level plus MGA grew to breeding size by about 11.4 months of age. While this is important in that it shows the potential that is available, a far more important finding is that the normal level heifers grew to breeding size by about 12.5 months of age. These heifers were raised similarly to the way heifers could be raised on commercial dairy farms. And the results indicate the practical progress that can be made in lowering the age of heifers at first breeding and thereby at first parturition. Heifers fed the normal level plus MGA were 12.1 months old at breeding size. This age was not significantly different ($P>0.10$) from the normal level value and indicates no significant growth advantage from feeding MGA with a normal nutritional level.

Body weights at 120 cm withers height (Table 7) for the heifers fed MGA along with the high nutrition level were significantly greater than for heifers fed the other nutrition treatments ($P<0.01$). This confirms the claim that MGA will increase weight gains by about 10 percent in feedlot heifers fed a heavy concentrate ration. However, those heifers fed the normal level plus MGA treatment weighed the same and were the same age at 120 cm withers height as the normal level heifers. Thus, no increase in body weight or rate of skeletal growth due to MGA occurred when a low level of grain was fed. Furthermore, MGA did not increase the rate of skeletal growth when fed with the high nutrition level; heifers fed the high level reached 120 cm at the withers at the

TABLE 7.--Growth to breeding size (120 cm withers height) for Holstein heifers fed different nutrition treatments.[a]

Nutrition treatment	Number heifers	Breeding size		Change from first estrus			
		Age	Weight	Age	Weight	Height	Daily gain
		(mo)	(kg)	(mo)	(kg)	(cm)	(kg)
Normal	20	12.5 ± 0.2[b]	350 ± 5	3.4	100 ± 8	10.4 ± 1.0	1.02
Normal + MGA from 2.5 mo.	10	12.1 ± 0.4	353 ± 10	-	-	-	-
High	20	11.4 ± 0.4	352 ± 7	3.4	87 ± 11[d]	10.7 ± 1.2	0.89[d]
High + MGA from 2.5 mo.	20	11.3 ± 0.3	385 ± 8[c]	3.5	107 ± 13	11.3 ± 1.6	0.99[e]
High + MGA from first estrus	20	11.4 ± 0.3	387 ± 8[c]	3.7	127 ± 8	12.3 ± 0.9	1.15

[a]Values are means and their standard errors.
[b]Significantly greater than values for the high and high + MGA treatments ($P<0.01$).
[c]Significantly greater than values for the normal, normal + MGA, and high treatments ($P<0.01$).
[d]Significantly less than values for the high + MGA treatments ($P<0.025$).
[e]Significantly less than value for the high + MGA from first estrus treatment ($P<0.05$).

same age as did those heifers fed the high level plus MGA. From these data it is apparent that MGA does not affect skeletal growth, and that it stimulated body weight gain only when fed with large amounts of grain. Analysis of the body weight data revealed a significant interaction ($P<0.01$) between MGA and grain level, which supports such a conclusion.

The time spans between first estrus and breeding size were not significantly different ($P>0.10$) among the treatments (3.4 to 3.7 months). This implies that the normal level heifers grew just as fast after first estrus as those fed the high level without MGA. And indeed this did occur as evidenced by the weight and height changes and the daily gains during this period (Table 7).

Analysis of the data comprising Table 8 and shown in Figure 1 revealed that the body weight curves for heifers receiving the normal level plus MGA and the high level plus MGA were linear. But, the curves for heifers fed the normal and high levels without MGA were not linear ($P<0.01$). To explain these results becomes difficult since they do not agree with the Morrison (1959) or Beltsville (1954) standard growth curves, or the values obtained by Sorenson et al., (1959) for heifers receiving a high nutritional level. According to these studies cited, Holstein heifers gain in body weight at a linear rate through at least the first 12 months of life. The monthly weights shown in Table 8 and Figure 1 reveal that the normal level without MGA heifers started gaining at a faster rate after about 6.5 months of age. Meanwhile, heifers fed the high level without MGA declined in their rate of gain after about 8 months of age. The cause (or causes) for this phenomenon is unknown.

Apparently MGA compensated for the decline in rate of gain by high

TABLE 8.--Body weights by age for Holstein heifers fed different nutrition treatments but not slaughtered at first estrus.[a]

Nutrition treatment						
Age	Normal Weight	High Weight	High + MGA from 2.5 mo. Weight	High + MGA from first estrus Weight	Age	Normal + MGA from 2.5 mo. Weight
(mo)	(kg)	(kg)	(kg)	(kg)	(mo)	(kg)
0.5	49	49	48	46	0.5	45 (10)
2.5	96	96	100	95	2.5	97 (10)
3.0	106	110	109	105	3.3	117 (10)
3.4	114	120	117	118	4.3	140 (10)
4.4	133	150	150	150	5.3	168 (10)
5.4	155	185	185	184	6.3	197 (10)
6.3	174	212	213	215	7.3	217 (10)
7.9	221	265	276	273	8.2	245 (10)
8.7	248	291	300	298	9.2	275 (5)
9.8	268	310 (18)	328 (19)	326 (18)	9.7	284 (10)
10.7	292 (19)	331 (17)	356 (18)	357 (18)	11.1	325 (5)
11.7	315 (18)	356 (15)	381 (15)	391 (14)	12.1	352 (8)
12.5	344 (17)	380 (12)	403 (12)	415 (12)	12.9	382 (5)
13.6	374 (16)	405 (12)	436 (11)	448 (10)		

[a]Twenty heifers at each value except for values followed by number in parentheses. Values are means.

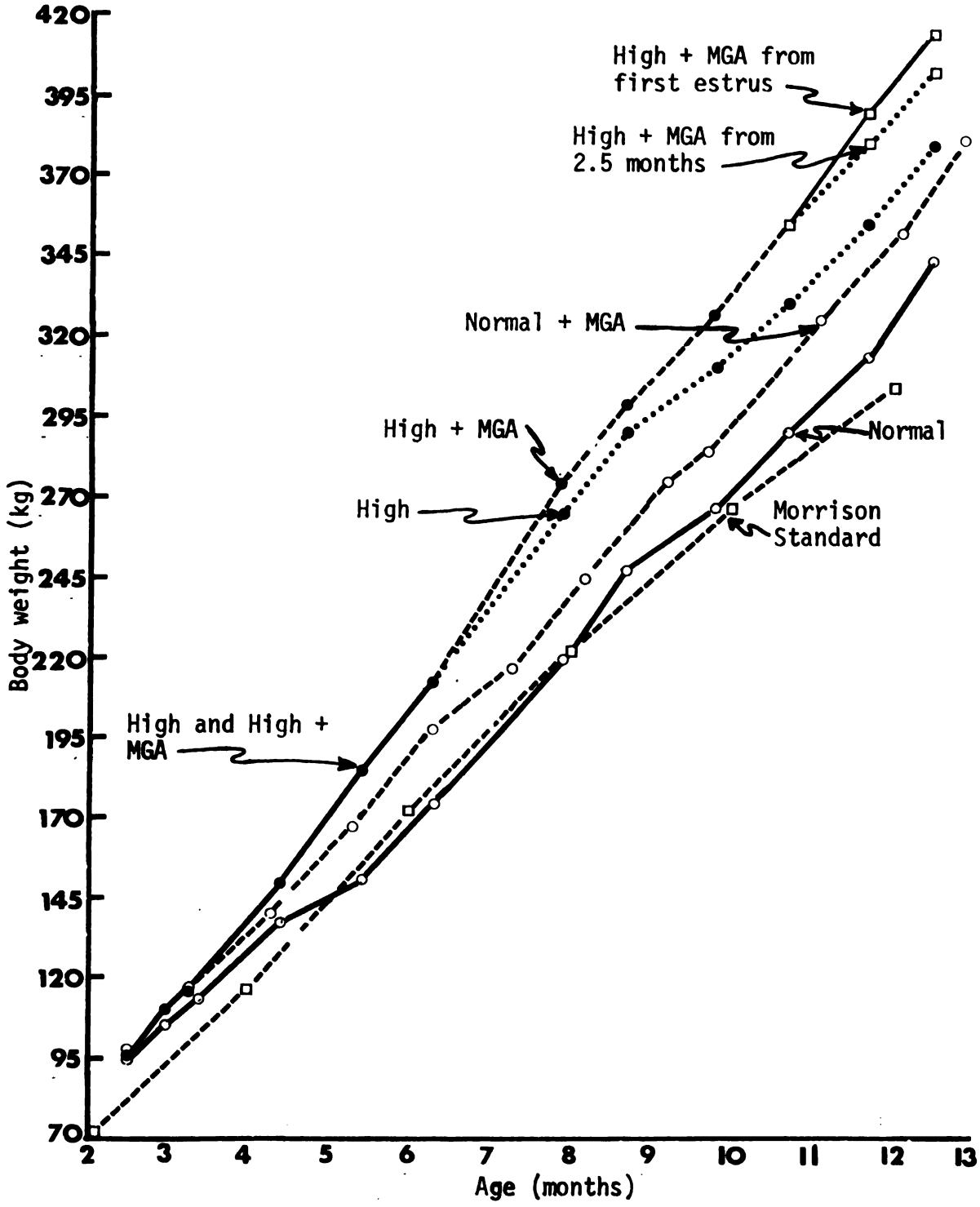

Figure 1.--Body weights by age for Holstein heifers fed different nutrition treatments but not slaughtered at first estrus.

level heifers as indicated in Tables 6 and 7 and Figure 1. Heifers receiving the high level and the high level plus MGA after first estrus gained nearly the same amount between 2.5 months and first estrus (169 kg vs 165 kg, respectively), but between first estrus and breeding size, a nearly equal time period for the two groups, heifers receiving MGA gained about 50 percent more ($P<0.025$) than heifers receiving just the high level (127 kg vs 87 kg, respectively).

Between 2.5 and 4.4 months of age, heifers fed the high level without MGA had a faster rate of skeletal growth than those fed the normal level without MGA. As the data in Table 9 show, after about 4.4 months of age heifers on all nutritional treatments increased in withers height at the same rate. From these findings, it is apparent that nutritional level has little influence on rate of skeletal growth.

Photographs taken at breeding size which are representative of the heifers fed the normal level, high level, and high level plus MGA treatments are shown in Figure 2.

4. Body Size at Slaughter

The ages, body weights, and withers heights at slaughter for heifers on the various nutritional treatments are presented in Table 10 and Appendix I. Study of the first estrus slaughter data and the monthly body size of heifers slaughtered at this physiological age (Table 11) shows the growth stimulating effect of the high nutritional level. MGA did not produce a further increase in weight or height, although as noted earlier, after about 5.5 months of age, heifers fed the high level plus MGA did gain faster than the high level heifers. Although heifers fed the normal level without MGA and slaughtered at first estrus were older ($P<0.01$), body weight and height at the withers were the same for heifers on the normal and high levels without MGA ($P>0.10$). Heifers fed

TABLE 9.--Withers heights by age for Holstein heifers fed different nutrition treatments but not slaughtered at first estrus.[a]

Nutrition treatment						
Normal		High	High + MGA from 2.5 mo.	High + MGA after first estrus		Normal + MGA from 2.5 mo.
Age	Height	Height	Height	Height	Age	Height
(mo)	(cm)	(cm)	(cm)	(cm)	(mo)	(cm)
2.5	86.5	86.7	87.2	85.1	2.5	89.2 (10)
3.0	88.5	89.0	88.7	87.7	3.3	91.9 (10)
3.4	89.3 (10)	91.3 (10)	90.6 (10)	90.9 (10)	4.3	95.7 (10)
4.4	91.9	94.9	94.3	94.0	5.3	99.9 (10)
5.4	96.2	99.3	98.7	98.7	6.3	104.0 (10)
6.3	99.6 (10)	103.2	102.5	103.2	7.3	107.3 (10)
7.9	105.3	108.5	108.4	108.8	8.2	110.4 (10)
8.7	108.9	112.1	111.9	112.0	9.2	112.4 (5)
9.8	112.0	114.7 (18)	114.7 (19)	114.5 (18)	9.7	114.1 (10)
10.7	114.7 (19)	117.6 (17)	117.6 (18)	117.4 (18)	11.3	116.5 (8)
11.7	117.7 (18)	119.6 (15)	119.8 (15)	119.0 (14)	12.1	118.2 (8)
12.5	119.3 (17)	120.8 (12)	121.2 (12)	120.7 (12)	12.9	120.1 (5)
13.6	121.5 (16)	122.5 (12)	122.6 (11)	122.8 (10)		

[a]Twenty heifers at each value except for values followed by number in parentheses. Values are means.

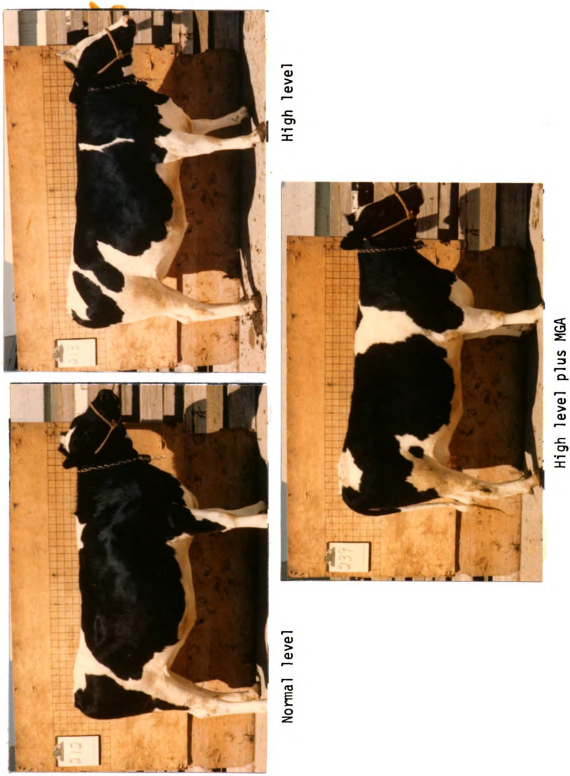

Figure 2.--Photographs at breeding size of Holstein heifers fed normal level, high level, and high level plus MGA nutrition treatments from 2.5 months of age.

TABLE 10.--Age and body size at slaughter of Holstein heifers fed different nutrition treatments.[a]

Nutrition treatment	2.5 months		First estrus			Breeding size		
	Weight	Height	Age	Weight	Height	Age	Weight	Height
	(kg)	(cm)	(mo)	(kg)	(cm)	(mo)	(kg)	(cm)
Normal	95 ± 4	85.9 ± 1.2	8.7 ± 0.3[b]	258 ± 6	110.3 ± 0.8	13.2 ± 0.4[d]	361 ± 6	121.0 ± 0.2
Normal + MGA from 2.5 mo.	-	-	-	-	-	12.5 ± 0.4	359 ± 11	120.4 ± 0.4
High	-	-	7.5 ± 0.2	251 ± 8	110.4 ± 1.0	12.1 ± 0.6	361 ± 10	120.3 ± 0.3
High + MGA from 2.5 mo.	-	-	7.2 ± 0.2	245 ± 9	107.6 ± 0.8[c]	11.7 ± 0.4	396 ± 11[e]	120.7 ± 0.2
High + MGA after first estrus	-	-	-	-	-	11.4 ± 0.5	384 ± 13[e]	120.4 ± 0.4

[a]Values are means and their standard errors for 10 heifers, except high treatment first estrus values which are for 20 heifers.
[b]Significantly different from the other values ($P<0.01$).
[c]Significantly different from the other values ($0.05<P<0.10$).
[d]Significantly different from values for the high and high + MGA treatments ($P<0.025$).
[e]Significantly different from values for the normal, normal + MGA, and high treatments ($P<0.01$).

TABLE 11.--Body size by age for Holstein heifers fed different nutrition treatments and slaughtered at first estrus.[a]

Age	Nutrition treatment: Normal Weight	Normal Height	High Weight	High Height	High + MGA from 2.5 mo. Weight	High + MGA from 2.5 mo. Height
(mo)	(kg)	(cm)	(kg)	(cm)	(kg)	(cm)
0.5	42	--	42 (20)	--	44	--
2.5	90	85.0	94 (20)	87.5 (20)	96	86.8
3.3	113	89.7	121 (20)	91.9 (20)	122	91.6
4.3	137	95.0	149 (20)	98.1 (20)	156	97.0
5.3	162	98.9	184 (20)	102.4 (20)	185	101.7
6.3	193	102.8	222 (20)	106.3 (20)	225	105.2
7.3	205 (8)	105.1 (8)	241 (9)	109.5 (9)	245 (1)	106.0 (1)
8.2	228 (8)	107.4 (8)	241 (2)	110.5 (2)	--	--
9.5	267 (6)	110.6 (6)	295 (1)	114.5 (1)	--	--

[a]Each value represents the mean for 10 heifers except for values followed by number in parentheses.

the high level plus MGA were somewhat younger and smaller than those fed the normal or high level without MGA. The fact they were younger occurred by chance when the heifers were randomly assigned to treatment groups and then paired with the high level heifers on the basis of body weights at 2 weeks of age.

Data on the heifers slaughtered at breeding size (Table 10) were very similar to those presented for all heifers at breeding size in Table 7. The slight differences in measurements exist because of the time lapse between the attainment of breeding size and slaughter. Heifers on MGA were slaughtered 48 hours after withdrawal while all other heifers were slaughtered at 17-20 days post estrus. Heifers on the high level and high level plus MGA were younger at slaughter than heifers on the normal level without MGA ($P<0.025$). Also, the high level plus MGA heifers weighed more than heifers on the other nutritional treatments ($P<0.01$), but no difference existed in withers heights. MGA fed with the normal nutritional level produced no significant difference ($P>0.10$) in age, weight, or withers height. This again points to the interaction between MGA and the high level of grain as discussed previously.

Data on body composition, length and dry weight of the right cannon bone, and thyroid gland weights were collected on the 100 heifers that were slaughtered. Also, acinar cell heights and plasma bound iodine determinations were obtained on certain treatment groups. These data are presented by Roger W. Purchas (1970) as part of his Ph.D. thesis.

B. Reproductive Tract Changes

1. Uterine Weight

Uterine weight increased about four fold from 2.5 months of age to

first estrus (Table 12 and Appendix II). Per 100 kg body weight the uteri more than doubled in weight during this period. Although some uterine weight change is associated with body growth, the majority occurs shortly before first estrus (Desjardins and Hafs, 1969). This would seemingly imply that the ovaries begin steroid hormone secretion before first estrus, since it is accepted that ovarian steroids regulate uterine growth. The various nutritional treatments imposed upon the heifers after 2.5 months of age had no apparent influence on uterine weight at first estrus. However, at breeding size (Table 13 and Appendix II), uteri from heifers fed the normal level and normal level plus MGA weighed significantly less ($P<0.05$) than uteri from heifers fed the high level and high level plus MGA. But uterine weight per 100 kg body weight revealed no significant effect ($P>0.10$) of the various nutritional treatments. Thus, the uteri of heifers fed the high level and high level plus MGA weighed more because the hiefers were heavier.

2. Uterine Nucleic Acids

Uterine nucleic acids (DNA and RNA) increased in total amounts from 2.5 months to first estrus (Table 12 and Appendix II), reflecting the increase in uterine weight during this time period. However, per gram of uterus the picture is different. DNA concentration (mg DNA/g uterus) declined significantly ($P<0.01$) from 2.5 months to first estrus for all treatments. The value for heifers fed the high level plus MGA declined the most, and it was significantly different ($P<0.01$) from the values for heifers fed the normal or high level without MGA. Since uterine weights of heifers fed MGA were not different from those of heifers not fed MGA, hypertrophy of the uterine cells is suggested. Meanwhile, RNA concentration (mg RNA/g uterus) values for all treatment

TABLE 12.--Uterine measurements of Holstein heifers fed different nutrition treatments and slaughtered at 2.5 months or first estrus.[a]

Measurement	2.5 months	Nutrition treatments from 2.5 months to first estrus		
		Normal	High	High + MGA from 2.5 mo.[b]
Weight (g)	29.6 ± 1.5	126.9 ± 8.9	122.5 ± 8.1	116.8 ± 12.0
DNA (mg)	153.9 ± 10.1	599.7 ± 51.1[e]	530.2 ± 28.9[e]	390.7 ± 39.5
DNA (mg/g)	5.2 ± 0.1[c]	4.7 ± 0.2[e]	4.5 ± 0.2[e]	3.5 ± 0.3
RNA (mg)	119.9 ± 9.0	419.2 ± 57.6	432.5 ± 30.0	426.0 ± 41.8
RNA (mg/g)	4.0 ± 0.1[d]	3.4 ± 0.4	3.7 ± 0.2	3.7 ± 0.2
RNA/DNA	0.78 ± 0.03	0.73 ± 0.10[e]	0.81 ± 0.07[f]	1.10 ± 0.07
Cell height (u)	16.6 ± 1.0	25.5 ± 1.5	26.8 ± 1.2	25.4 ± 1.9

[a]Values are means and their standard errors for 10 heifers, except high treatment values which are for 20 heifers.
[b]Values at first estrus of high treatment pairmates.
[c]Significantly greater than the first estrus values ($P<0.01$).
[d]Significantly greater than the first estrus values ($P<0.05$).
[e]Significantly different from value for high + MGA treatment ($P<0.01$).
[f]Significantly different from value for high + MGA treatment ($P<0.05$).

TABLE 13.--Uterine measurements of Holstein heifers fed different nutrition treatments and slaughtered at breeding size.[a]

Measurement	Nutrition treatment				
	Normal	Normal + MGA from 2.5 mo.	High	High + MGA from 2.5 mo.	High + MGA after first estrus
Weight (g)	136.8 ± 7.8[b]	129.3 ± 7.5[b]	151.7 ± 5.5	155.4 ± 12.5	165.8 ± 9.8
DNA (mg)	586.1 ± 19.9	459.5 ± 31.4[c]	640.2 ± 30.3	547.5 ± 27.9[d]	643.8 ± 39.1
DNA (mg/g)	4.3 ± 0.2	3.6 ± 0.2[e]	4.2 ± 0.2	3.7 ± 0.3	3.9 ± 0.2
RNA (mg)	443.6 ± 38.6[b]	450.4 ± 48.6[b]	520.5 ± 28.6[f]	672.2 ± 77.0	667.3 ± 74.0
RNA (mg/g)	3.2 ± 0.1[f]	3.4 ± 0.2[f]	3.4 ± 0.1[f]	4.3 ± 0.3	4.0 ± 0.3
RNA/DNA	0.76 ± 0.06[gf]	1.00 ± 0.09	0.84 ± 0.08[f]	1.21 ± 0.10	1.08 ± 0.09
Cell Height (u)	26.6 ± 0.6[f]	28.6 ± 1.3	27.2 ± 1.4[h]	30.9 ± 2.2	31.5 ± 2.7

[a]Values are means and their standard errors for 10 heifers.
[b]Significantly less than values for the high and high + MGA treatments ($P<0.05$).
[c]Significantly less than all other values ($P<0.05$).
[d]Significantly less than values for the high and high + MGA after first estrus treatments ($P<0.05$).
[e]Significantly less than values for the normal and high treatments ($P<0.05$).
[f]Significantly less than values for the high + MGA treatments ($P<0.05$).
[g]Significantly less than the normal + MGA treatment value ($P<0.05$).
[h]Significantly less than values for the high + MGA treatments ($0.05<P<0.10$).

groups declined significantly ($P<0.05$) from 2.5 months to first estrus. This finding does not support the DNA implication of uterine hypertrophy due to MGA.

DNA concentrations at 2.5 months and at first estrus for the normal level heifers were about the same as values reported by Desjardins and Hafs (1969) for similarly fed heifers of comparable ages. However, their values for RNA concentration were only about one-half the values obtained in this study. Consequently, their RNA/DNA ratios were about half the values found in this study.

Feeding a normal or high nutritional level did not change the RNA/DNA ratios at first estrus from the value obtained at 2.5 months (Table 12 and Appendix II). But MGA fed along with the high level significantly increased ($P>0.05$) the ratio over the value at 2.5 months and the other treatment values at first estrus. These data, like the DNA data, implicate hypertrophy of uterine cells due to MGA. This contention is further supported by the nucleic acid data at breeding size (Table 13 and Appendix II). Uterine DNA concentrations at breeding size were lowest in all treatment groups fed MGA, and singificantly less ($P<0.05$) in heifers fed the normal level plus MGA than in those fed the normal or high level. But unlike the first estrus data, RNA concentrations were elevated significantly ($P<0.05$) in heifers fed the high level plus MGA. There was no effect on RNA concentration in heifers fed the normal level plus MGA. As at first estrus, the RNA/DNA ratios were greater in heifers fed MGA; the normal level plus MGA value being greater ($P<0.05$) than the normal level value, and the high level plus MGA values being greater ($P<0.05$) than the high level value. Thus, the stimulatory effect of MGA on uterine cell size appears quite conclusive, as measured by nucleic acids content.

The high level of nutrition produced no significant change in DNA and RNA concentrations nor in the RNA/DNA ratios from the values for normal level heifers slaughtered at breeding size. Values for these normal level heifers slaughtered at about 13 months of age were similar to those of 17-month old heifers slaughtered by Hackett and Hafs (1969) on day 18 of the estrous cycle. Although the data are similar, the observations do not agree with the conclusions of Hackett and Hafs (1969), because from the standpoint of uterine nucleic acids, 12- to 13-month old Holstein heifers appear as mature at that age as they will be by 17 months of age.

3. Uterus Epithelial Cell Height

Height of the uterus epithelial cells increased by 50 percent between 2.5 months and first estrus in heifers of all treatment groups (Table 12 and Appendix II). No treatment effect was detected at first estrus. The values obtained agree very closely with those of Desjardins and Hafs (1969) for heifers of comparable ages. Values at breeding size (Table 13 and Appendix II) for normal level heifers were less than reported values for 11-and 12-month old heifers(Desjardins and Hafs, 1969); but they were greater than values of Hackett and Hafs (1969) for 17-month old heifers. Animal variation most likely explains these differences since the heifers in all three studies were treated similarly, and slaughtered at the same stage of the estrous cycle.

A treatment effect on the epithelial height at breeding size is suggested by the data in Table 13 and Appendix II. Although the normal level plus MGA heifers had taller uterine epithelial cells than the normal and high level heifers, the difference was not significant ($P > 0.10$). But uterine cell height of heifers fed the high level plus MGA was significantly taller ($P < 0.05$) than those of heifers fed the normal

and high level. Thus, it appears that MGA produced an increase in the uterine epithelial cell height. Whether this increase was due to the direct progestational action of MGA on the epithelium or to the indirect action through the ovary and thus an estrogenic effect is not clear. Perhaps there was synergism between the two possible routes of action. The data of Hackett and Hafs (1969) on Holstein uterine epithelial height during the estrous cycle suggest that estrogen was the principal effector. In their study epithelial height was greatest on the day of estrus and at the time of the mid-cycle follicle. Still, this could mean that both hormones are required, since minimal amounts of progesterone would also be present at these times.

4. Ovarian Changes

Ovarian weight increased approximately 50 percent from 2.5 months to first estrus, and also from first estrus to breeding size (Table 14 and Appendix II). Values at all comparable ages were higher than those reported by Desjardins and Hafs (1969). No treatment effect existed at first estrus, but at breeding size ovaries of heifers fed the high level of nutrition without MGA weighed significantly less ($P<0.025$) than those of heifers fed the other nutritional treatments. This may have resulted from animal variation, or more likely from an unknown cause. MGA did not cause a significant ($P>0.10$) increase in ovarian weights which agrees with the results of Zimbelman and Smith (1966b). However, they found that MGA caused a significant increase ($P<0.05$) in the follicular fluid weight, which was not measured in this study. Average number of follicles by size (Table 14) showed that heifers fed MGA had a higher incidence of larger follicles. This also confirms Zimbelman and Smith's (1966b) data. It therefore appears that MGA increases ovarian follicle size, and the follicles in turn secrete estrogens (perhaps at elevated

TABLE 14.--Ovarian measurements of Holstein heifers fed different nutrition treatments and slaughtered at 2.5 months, first estrus, or breeding size.

Nutrition treatment	Time of slaughter	Paired ovarian weight [a]	No. Follicles by size				
			4-9	10-15	16-20	>20	Total
		(g)	--------(mm)---------				
Normal	2.5 months	6.8 ± 1.4	2.0	0.6	0	0	2.6
Normal	First estrus	10.0 ± 0.9	3.9	0.7	0.1	0	4.6
High	First estrus	11.4 ± 0.8	2.0	0.8	0.3	0	3.1
High + MGA from 2.5 mo.	First estrus [b]	10.2 ± 1.1	3.9	0.4	0.5	0.3	5.1
Normal	Breeding size	15.1 ± 0.8	1.5	1.3	0.5	0	3.3
Normal + MGA from 2.5 mo.	Breeding size	15.4 ± 0.9	2.2	0.7	0.5	0.7	4.1
High	Breeding size	12.9 ± 0.7 [c]	3.3	1.1	0.4	0.1	4.9
High + MGA from 2.5 mo.	Breeding size	15.4 ± 1.8	1.4	0.3	1.0	0.7	3.4
High + MGA after first estrus	Breeding size	17.6 ± 1.3	3.7	0.3	0.6	1.0	5.6

[a] Values are means and their standard errors for 10 heifers, except high treatment first estrus values which are for 20 heifers.

[b] Values at first estrus of high treatment pairmates.

[c] Significantly less than values for all other treatments at breeding size ($P<0.025$).

levels from normal) which according to Bloss et al., (1966) cause the body weight increase observed in feedlot heifers.

C. Mammary Gland Changes

1. Mammary Gland Weight

The left halves of the mammary glands were used to obtain the different parameters shown in Table 15 and Appendix III. Weight of the dissected parenchymal tissue increased about ten fold between 2.5 months of age and first estrus. But between first estrus and breeding size the value increased only about two fold. No detectable difference ($P>0.20$) in mammary gland weight due to nutritional treatment existed at either slaughter age because of the large variation within each treatment group. Although mammary weights of heifers in this study were considerably larger than those obtained by Sinha and Tucker (1969) from heifers of comparable ages, the same growth pattern existed. That is, starting at 2 to 3 months of age up to about 8 or 9 months of age, or near the time of first estrus, the mammary gland grew at a greatly accelerated rate. During this time it had an allometric growth pattern in comparison to the body's rate of growth. Before and after this time interval, the mammary gland grew at about the same rate as the body, or isometrically.

2. Mammary Gland Nucleic Acids

At first estrus heifers fed the high level of nutrition without MGA had less total mammary DNA ($P<0.05$) than heifers fed the other two nutritional treatments (Table 15 and Appendix III). This might be expected since the mammary glands, though not significantly smaller, actually weighed the least. But per gram of tissue, no difference in the DNA values was detectable, although the value for the group fed MGA was the largest, suggesting somewhat greater cell numbers. This indicates no nutritional

TABLE 15.--Mammary gland measurements of Holstein heifers fed different nutrition treatments and slaughtered at 2.5 months, first estrus, or breeding size.[a]

Nutrition treatment	Time of slaughter	Dissected weight	DNA		RNA		RNA / DNA
		(g)	(mg)	(mg/g)	(mg)	(mg/g)	
Normal	2.5 mo.	27 ± 5	64 ± 24	2.0 ± 0.3	65 ± 15	2.5 ± 0.3	1.40 ± 0.14
Normal	First estrus	328 ± 38	796 ± 82	2.5 ± 0.2	534 ± 96	1.6 ± 0.1[d]	0.69 ± 0.09
High	First estrus	238 ± 17	576 ± 53[c]	2.4 ± 0.1	421 ± 36	1.8 ± 0.1[d]	0.78 ± 0.06
High + MGA from 2.5 mo.	First estrus[b]	280 ± 62	817 ± 217	2.8 ± 0.2	778 ± 214	2.6 ± 0.2	0.92 ± 0.07
Normal	Breeding size	707 ± 49	1694 ± 156[df]	2.4 ± 0.2[e]	1445 ± 205[df]	2.1 ± 0.3[e]	0.88 ± 0.10
Normal + MGA from 2.5 mo.	Breeding size	578 ± 66	2191 ± 315[d]	3.7 ± 0.1	1988 ± 394[d]	3.2 ± 0.3	0.87 ± 0.07
High	Breeding size	599 ± 66	1501 ± 250[d]	2.4 ± 0.2[e]	1289 ± 228[d]	2.1 ± 0.2[e]	0.89 ± 0.08
High + MGA from 2.5 mo.	Breeding size	788 ± 36	2564 ± 235	3.2 ± 0.2	2342 ± 186	2.9 ± 0.1	0.94 ± 0.05
High + MGA after first estrus	Breeding size	736 ± 135	2369 ± 385	3.3 ± 0.2	2258 ± 374	3.2 ± 0.2	0.97 ± 0.06

[a]Values are means and their standard errors for left halves of the mammary glands for 10 heifers, except high treatment first estrus values which are for 20 heifers.
[b]Values at first estrus of high treatment pairmates.
[c]Significantly less than values for other treatments ($P<0.05$).
[d]Significantly less than values for high + MGA treatments ($P<0.01$).
[e]Significantly less than values for MGA treatments ($P<0.01$).
[f]Significantly less than value for normal + MGA treatment ($P<0.20$).

treatment effect on cell concentration up to this age. In a reversal pattern, total RNA values were not significantly affected by the nutritional treatments, but RNA concentration was; heifers fed MGA had a significantly ($P<0.01$) greater RNA concentration. Therefore, it would appear that protein synthesis had been stimulated by MGA. Although this fact is suggested, RNA to DNA ratios revealed no detectable differences ($P>0.20$). Thus it would seem that MGA fed with the high level of nutrition from 2.5 months of age to first estrus had stimulated cellular growth to a certain extent and most likely protein synthesis on a cellular basis.

Effects of MGA on nucleic acids become quite conclusive after studying data obtained when the heifers reached breeding size (Table 15 and Appendix III). In each of the two groups fed the high level plus MGA, 7 of the 10 heifers exhibited mammary proliferation, whereas only 5 of 10 heifers in the group fed the normal level plus MGA showed a response. Total mammary DNA of heifers fed the high level plus MGA was about 60 percent greater ($P<0.01$) than the value for heifers fed just the high level, while the value for heifers fed the normal level plus MGA was about 30 percent greater ($P<0.20$) than the value for heifers fed just the normal level. No apparent beneficial effect resulted from commencing MGA feeding at 2.5 months of age rather than after first estrus. Total RNA showed trends similar to those for DNA, with the value being about 78 percent greater ($P<0.01$) when MGA was given with the high level of nutrition than when it was not, and about 38 percent greater ($P<0.20$) when given with the normal level than when it was not.

Examining the data on a concentration basis led to the same findings. Both DNA and RNA per gram of tissue were significantly greater ($P<0.01$) for the groups fed MGA, indicating more cells and more protein synthesis

per gram of tissue. RNA to DNA ratios were not significantly different, implying the same degree of protein synthesis occurring per cell for the heifers on the various nutritional treatments. The values obtained were about 50 percent greater than the value reported by Sinha and Tucker (1969) for 12-month old Holstein heifers. A difference among the treatment groups in the type of mammary protein being synthesized was suggested by observations at slaughter. Most of the heifers fed the normal or high level had only a small quantity of a nearly clear fluid in the mammary gland, while in the glands of most of the heifers fed MGA there was a rather large volume of a cloudy, milky-looking substance. Whether this secretion and the nucleic acids changes were caused by progesterone and estrogen activity or by corticoid activity of MGA, or by certain pituitary hormones, or by synergism of several of these hormones is not known.

Certain structural differences were also observed in the glands. The parenchymal tissue appeared pinker and the duct system seemed more developed in mammary glands of heifers fed MGA than in the glands of heifers not fed MGA. Examination of the histological sections revealed that the degree of ductular development was greater in heifers fed MGA. However, no satisfactory method was found to quantitatively measure the development.

Representative rear view pictures of the mammary glands as they appeared at breeding size on heifers fed the normal level, high level, and high level plus MGA are shown in Figure 3.

D. Adrenal Gland Changes

1. Adrenal Weights

Adrenal weights increased about 80 percent from 2.5 months to first estrus as shown in Table 16 and Appendix III. There was no significant effect ($P>0.10$) of the various nutritional treatments on actual adrenal weights or weight per 100 kg body weight at first estrus. By the time the

Figure 3.--Rear view photographs of mammary glands on Holstein heifers at breeding size that were fed normal level, high level, and high level plus MGA nutrition treatments from 2.5 months of age.

TABLE 16.--Adrenal gland measurements of Holstein heifers fed different nutrition treatments and slaughtered at 2.5 months, first estrus, or breeding size.[a]

Nutrition treatment	Time of slaughter	Paired adrenal weight	Cortex width	Width of cortex zones		
				Glomerulosa	Fasciculata	Reticularis
		(g)	(mm)	----------(mm)----------		
Normal	2.5 mo.	6.1 ± 0.3	1.38 ± 0.04	0.33 ± 0.02	0.65 ± 0.03	0.41 ± 0.02
Normal	First estrus	11.5 ± 0.5	1.72 ± 0.07	0.31 ± 0.02	1.02 ± 0.05[e]	0.39 ± 0.02
High	First estrus	11.7 ± 0.4	1.61 ± 0.06	0.30 ± 0.01	0.89 ± 0.04	0.42 ± 0.03
High + MGA from 2.5 mo.	First estrus[b]	10.4 ± 0.5	1.68 ± 0.11	0.31 ± 0.02	0.89 ± 0.07	0.41 ± 0.03
Normal	Breeding size	13.7 ± 0.4	2.10 ± 0.15[c]	0.34 ± 0.03	1.26 ± 0.09[c]	0.60 ± 0.05[c]
Normal + MGA from 2.5 mo.	Breeding size	14.3 ± 0.9	1.71 ± 0.10[d]	0.35 ± 0.02	0.93 ± 0.07	0.44 ± 0.03
High	Breeding size	15.9 ± 0.9	2.20 ± 0.15[d]	0.33 ± 0.01	1.37 ± 0.13[d]	0.50 ± 0.03
High + MGA from 2.5 mo.	Breeding size	15.6 ± 0.7	1.77 ± 0.09	0.29 ± 0.01	1.05 ± 0.08	0.43 ± 0.04
High + MGA after first estrus	Breeding size	14.3 ± 0.7	2.01 ± 0.07	0.35 ± 0.02	1.12 ± 0.08	0.54 ± 0.06

[a]Values are means and their standard errors for 10 heifers, except high treatment first estrus values which are for 20 heifers.
[b]Values at first estrus of high treatment pairmates.
[c]Significantly greater than value for normal + MGA treatment ($P<0.05$).
[d]Significantly greater than values for high + MGA treatments ($P<0.05$).
[e]Significantly greater than values for other treatments ($0.05<P<0.10$).

heifers had grown from first estrus to breeding size the adrenals increased another 2 to 4 grams in weight, depending on the nutritional treatment. Again no significant differences were detected among nutritional treatments in the actual adrenal weights at breeding size. But, when expressed per 100 kg body weight, adrenals of the heifers fed the high level plus MGA weighed significantly less ($P<0.01$) than those of heifers fed the high level.

These adrenal weight data contrast with Zimbelman and Smith's (1966b) data. They found that long term MGA administration increased adrenal weights when compared with controls. This they took as further supportive evidence that the ovarian follicles of MGA fed heifers were secreting estrogens. Such evidence is not available from this study. Adrenal weights obtained in this study agree fairly well with those of Desjardins and Hafs (1966) for heifers of comparable ages.

2. Adrenal Cortex Histology

Total width of the cortex increased between 0.2 and 0.3 mm from 2.5 months of age to first estrus for all nutritional treatment groups (Table 16 and Appendix III). No significant difference ($P>0.10$) among the group values existed at first estrus. But this was no longer true at breeding size, as heifers fed MGA had adrenal cortex widths that were significantly smaller ($P<0.05$) than their normal and high level controls (Table 16 and Appendix III). Perhaps this observation was reflected in the decreased per unit weights as discussed in the previous section.

From the data on widths of the various cortex zones at breeding size (Table 16 and Appendix III), it is evident that MGA decreased the fasciculata zone width ($P<0.05$) in both the normal level plus MGA and high level plus MGA heifers. Also, the reticularis zone width of the normal level plus MGA heifers was significantly less ($P<0.05$) than for the normal level heifers. The width of the glomerulosa zone did not change due to

nutritional treatments or with increasing size of the heifers. Desjardins (1966) found this also, although his values were somewhat smaller.

Since the fasciculata and reticularis zones secrete glucocorticoids, and since MGA has glucocorticoid activity (Duncan et al., 1964), it is tempting to speculate that MGA acts directly on the adrenal cortex to decrease the fasciculata zone width, or that perhaps MGA causes a reduction in the release of ACTH from the anterior pituitary. Preventing ACTH release could be by direct action on the anterior pituitary or via affecting release of corticotropin releasing factor from the hypothalamus. Because of the lowered circulating levels of ACTH to stimulate the fasciculata and reticularis zones, these zones would decrease somewhat in width and perhaps function. Data supporting this last statement about a functional decline are presented by Roger W. Purchas (1970). He measured the adrenal and plasma corticoids of the heifers fed the normal level, high level, and high level plus MGA and slaughtered at breeding size. Heifers fed MGA had significantly lower ($P<0.05$) cortisol concentrations in both the adrenals and plasma than the heifers fed either the normal or high nutritional levels without MGA. One might then conclude that MGA, through a positive feedback mechanism, causes the glucocorticoid secreting adrenal cortex zones to decrease in both width and function. However, Purchas (1970) found a nonsignificant correlation between adrenal cortex width and adrenal corticoid content which would not support such a conclusion. Why MGA caused a decreased fasciculata width when fed with both levels of nutrition, but a decreased reticularis width when fed with only the normal level is not known. Perhaps there is an interaction between level of nutrition and MGA in the manner by which MGA affects the adrenal cortex.

E. Changes in the Pituitary Weight and Hormones

1. Pituitary Weight

Weights of the total pituitary and its two parts as influenced by body size and treatments are shown in Table 17 and Appendix IV. Weight of the total pituitary increased about 50 percent between 2.5 months and first estrus, with the nutritional treatments producing no detectable difference ($P>0.05$). Between first estrus and breeding size there was only a 20 to 30 percent increase in total pituitary weight in all nutritional treatment groups. Again no significant differences existed ($P>0.10$) between the treatment values, although values for heifers fed MGA were the largest. These observations agree with those of Zimbelman (1966) who reported that MGA does not significantly affect pituitary weights. Total pituitary weight values obtained in this study also agree closely with those reported by Desjardins (1966) for heifers of comparable ages.

Anterior pituitary weight changes followed the same pattern of the total pituitary (Table 17 and Appendix IV). However, at breeding size, heifers fed the high level plus MGA had significantly heavier ($P<0.025$) anterior pituitaries than heifers fed the high level without MGA. Even per 100 kg body weight this difference was still significant ($P<0.10$). The value for heifers fed the normal level plus MGA was not significantly different ($P>0.10$) from that of the normal level without MGA. Thus, it appears that feeding heifers a high level of nutrition plus MGA resulted in enlarged anterior pituitaries when compared with values for high level ones.

There appears to be no simple explanation for the elevated anterior pituitary weights. Zimbelman (1966) found elevated levels of LH in the pituitaries of heifers fed MGA. So, an immediate explanation for the increased anterior pituitary weights is that they are associated with increased LH content. However, as will be discussed in the next section,

TABLE 17.--Pituitary weights of Holstein heifers fed different nutrition treatments and slaughtered at 2.5 months, first estrus, or breeding size.[a]

Nutrition treatment	Time of slaughter	Total pituitary	Anterior pituitary	Posterior pituitary
		----------(g)----------		
Normal	2.5 mo.	0.99 ± 0.08	0.76 ± 0.06	0.18 ± 0.02
Normal	First estrus	1.64 ± 0.10	1.26 ± 0.09	0.33 ± 0.01
High	First estrus	1.48 ± 0.04	1.10 ± 0.03	0.34 ± 0.01
High + MGA from 2.5 mo.	First estrus[b]	1.47 ± 0.07	1.11 ± 0.07	0.33 ± 0.01
Normal	Breeding size	1.72 ± 0.10	1.35 ± 0.07	0.34 ± 0.03
Normal + MGA from 2.5 mo.	Breeding size	1.80 ± 0.08	1.39 ± 0.07	0.37 ± 0.02
High	Breeding size	1.74 ± 0.07	1.26 ± 0.05	0.40 ± 0.03
High + MGA from 2.5 mo.	Breeding size	1.92 ± 0.14	1.54 ± 0.13[c]	0.35 ± 0.02
High + MGA after first estrus	Breeding size	1.93 ± 0.10	1.51 ± 0.08[c]	0.34 ± 0.02

[a]Values are means and their standard errors for 10 heifers, except the high treatment first estrus values which are for 20 heifers.

[b]Values at first estrus of high treatment pairmates.

[c]Significantly greater than value for high treatment ($P<0.025$).

in this study anterior pituitary LH values were not affected by MGA. Since the fasciculata zone of the adrenals was reduced in width as already discussed, perhaps ACTH release was inhibited and the pituitary build up of ACTH was reflected in the anterior pituitary weight.

Posterior pituitary weights nearly doubled from 2.5 months of age to first estrus (Table 17 and Appendix IV), but values at breeding size were not changed from those at first estrus. Furthermore, the different nutritional treatments produced no differences in posterior pituitary weights at first estrus or breeding size. Since the posterior pituitary is derived from neural tissue which undergoes mainly prenatal growth, this finding might have been expected. Still, Desjardins (1966) found that heifer posterior pituitary weights generally increased from 1 to 12 months of age, suggesting more postnatal growth than observed in this study.

2. Hormones in the Anterior Pituitary

Data on the pituitary gonadotropins and prolactin are presented in the study while growth hormone data are presented by Roger W. Purchas (1970).

Levels of the gonadotropins and prolactin in the anterior pituitary are shown in Table 18 and Appendix IV. LH concentration values generally increased from 2.5 months to first estrus, with some additional increase within a nutritional treatment from first estrus to breeding size. The values at first estrus did not differ significantly ($P>0.10$) among the nutritional treatment groups. Breeding size values also showed little effect of nutritional treatment, except the value for heifers fed the normal level without MGA was slightly greater ($P<0.10$) than the other treatment values. Since all heifers were slaughtered at a similar stage of the estrous cycle, this difference was not expected. The LH concentration values were generally smaller than those reported by Desjardins (1966) and Zimbelman (1966). But the fact that they used an LH bioassay whereas a

TABLE 18.--Hormones in the anterior pituitary of Holstein heifers fed different nutrition treatments and slaughtered at 2.5 months, first estrus, or breeding size.[a]

Nutrition treatment	Time of slaughter	Concentration			Content		
		LH	FSH	Prolactin	LH	FSH	Prolactin
		(ug/mg)			(ug)		
Normal	2.5 mo.	1.55 ± 0.16	1.05 ± 0.26	0.13 ± 0.02	1061 ± 109	760 ± 218	92 ± 15
Normal	First estrus	1.71 ± 0.18	0.60 ± 0.11	0.14 ± 0.03	2000 ± 202	778 ± 177	177 ± 40
High	First estrus	1.46 ± 0.15	0.69 ± 0.14	0.15 ± 0.01	1527 ± 167	681 ± 125	157 ± 15
High + MGA from 2.5 mo.	First estrus[b]	1.74 ± 0.22	0.60 ± 0.19	0.14 ± 0.01	1868 ± 193	651 ± 192	148 ± 14
Normal	Breeding size	2.15 ± 0.29[c]	0.79 ± 0.18	0.17 ± 0.02[e]	2772 ± 413	996 ± 233	226 ± 36
Normal + MGA from 2.5 mo.	Breeding size	1.44 ± 0.13	0.29 ± 0.07[d]	0.18 ± 0.03[e]	1933 ± 187	400 ± 110[d]	260 ± 55
High	Breeding size	1.68 ± 0.23	0.80 ± 0.11	0.15 ± 0.02	2051 ± 320	948 ± 119	180 ± 17
High + MGA from 2.5 mo.	Breeding size	1.76 ± 0.40	0.77 ± 0.20	0.14 ± 0.01	2456 ± 539	1091 ± 267	203 ± 32
High + MGA after first estrus	Breeding size	1.24 ± 0.10	0.58 ± 0.14	0.11 ± 0.01	1721 ± 144	859 ± 262	152 ± 25

[a]Values are means and their standard errors for 10 heifers, except high treatment first estrus values which are for 20 heifers.
[b]Values at first estrus of high treatment pairmates.
[c]Significantly greater than the other treatment values ($P<0.10$).
[d]Significantly less than the other treatment values ($P<0.05$).
[e]Significantly greater than values for high + MGA treatments ($P<0.10$).

radioimmunoassay was used in this study could explain the differences.

Anterior pituitary content of LH increased between 50 and 100 percent from 2.5 months to first estrus, and like the concentration values increased an additional 20 to 30 percent within a nutritional treatment from first estrus to breeding size. Total content increases were thus a result of both concentration and anterior pituitary weight increases with increasing age. No significant differences ($P>0.10$) in content due to the nutritional treatments were detected at first estrus or breeding size.

FSH concentration values decreased markedly from 2.5 months to first estrus, with no significant ($P>0.10$) nutritional treatment differences present at first estrus (Table 18 and Appendix IV). Desjardins (1966) also found a precipitous drop in FSH concentration between 2 and 3 months of age. Perhaps this FSH decline is associated in some manner with the gradual processes involved in sexual maturation and the occurrence of first estrus. From first estrus to breeding size, the values remained relatively the same, with perhaps a slight increase within a particular nutritional treatment. While the values at breeding size are about one half the magnitude of Zimbelman's (1966) values for pregnant heifers, the two sets of data agree that MGA does not affect pituitary FSH concentration. However, there is one exception in that heifers at breeding size which were fed the normal level plus MGA had significantly smaller ($P<0.05$) pituitary FSH concentrations than all other nutritional treatment values at both breeding size and first estrus. This finding would suggest a depressing interaction effect of the normal level and MGA on pituitary FSH concentration. Such an interaction does not seem plausible.

Due to increased concentration, the pituitary FSH content at 2.5 months was as large as the first estrus values. Values at breeding size were about 35 percent greater than the first estrus values. No nutritional

treatment produced significantly different values ($P>0.10$) at first estrus or breeding size, except for the normal level plus MGA effect, as already discussed regarding concentration changes.

Pituitary prolactin concentration values were unaffected by the nutritional treatments or body size when comparing the values at 2.5 months and first estrus (Table 18 and Appendix IV). Sinha and Tucker (1969) also found no appreciable difference in pituitary prolactin values in heifers of comparable ages. However, at breeding size heifers fed the normal level and normal level plus MGA had slightly larger ($P<0.10$) prolactin concentrations than heifers fed the high level plus MGA. Since the value for heifers fed the high level without MGA was also somewhat lower than that of heifers fed the normal level or normal level plus MGA, an effect of nutritional level is implied. However, analysis of the data showed such an effect did not exist ($P>0.10$). Maybe heifers fed the normal level and normal level plus MGA were experiencing elevated pituitary prolactin levels concomitant with changes in mammary biochemical parameters, as proposed by Sinha and Tucker (1969) for 12-month old heifers in their study.

Pituitary prolactin content values nearly doubled from 2.5 months to first estrus. But from first estrus to breeding size only about a 25 percent increase resulted. No nutritional treatment effect was detectable at either age though the normal level and normal level plus MGA values were the largest. Thus, the concentration differences observed at breeding size were not significantly reflected in total pituitary prolactin content.

3. Hormones in the Blood Plasma

LH concentration in the blood plasma of heifers fed the normal level without MGA did not differ at 2.5 months and first estrus (Table 19 and Appendix IV). However, heifers fed the high level and high level plus MGA

TABLE 19.--Hormones in the blood plasma of Holstein heifers fed different nutrition treatments and slaughtered at 2.5 months, first estrus, or breeding size.[a]

Nutrition treatment	Time of slaughter	Concentration		Content[g]	
		LH	Prolactin	LH	Prolactin
		------(ng/ml)------		-----(ug)----------	
Normal	2.5 mo.	2.3 ± 0.3	58 ± 9	8 ± 1	196 ± 33
Normal	First estrus	2.4 ± 0.2[c]	92 ± 16[d]	21 ± 2	852 ± 163[d]
High	First estrus	3.0 ± 0.5	42 ± 10	26 ± 3	385 ± 94
High + MGA from 2.5 mo.	First estrus[b]	3.8 ± 0.3[d]	37 ± 7	33 ± 3[h]	315 ± 68
Normal	Breeding size	2.5 ± 0.2	112 ± 12[f]	32 ± 2	1419 ± 151[f]
Normal + MGA from 2.5 mo.	Breeding size	3.3 ± 0.3[e]	62 ± 11	41 ± 4[i]	755 ± 124
High	Breeding size	2.8 ± 0.2	81 ± 16	36 ± 3	1035 ± 219
High + MGA from 2.5 mo.	Breeding size	2.6 ± 0.2	95 ± 17	36 ± 3	1341 ± 253
High + MGA after first estrus	Breeding size	3.2 ± 0.3[e]	79 ± 12	43 ± 5[i]	1075 ± 170

[a]Values are means and their standard errors for 10 heifers, except high treatment first estrus values which are for 20 heifers.
[b]Values at first estrus of high treatment pairmates.
[c]Significantly less than the other treatment values ($P<0.05$).
[d]Significantly greater than the other treatment values and the 2.5 month value ($P<0.01$).
[e]Significantly greater than the normal and high + MGA from 2.5 mo. treatment values ($P<0.05$).
[f]Significantly greater than the normal + MGA treatment value ($P<0.10$).
[g]Estimated by assuming plasma volume to be 3.5 percent of the heifer's slaughter weight.
[h]Significantly greater than the other treatment values ($P<0.10$).
[i]Significantly greater than the normal treatment value ($P<0.10$).

had plasma LH concentrations which were singificantly greater at first estrus ($P<0.05$) than the value of heifers fed the normal level without MGA. Furthermore, the value for heifers fed the high level plus MGA was significantly greater ($P<0.01$) than first estrus values for heifers fed the normal or high level without MGA. This suggests an effect of high level of nutrition which is augmented by MGA. Values at breeding size further suggest that heifers fed MGA had elevated plasma LH concentrations, as two of the three groups fed MGA had elevated LH levels ($P<0.05$). Since heifers fed MGA had not consumed the drug for 48 hours prior to slaughter, perhaps during this time the drug lost its inhibitory action on cyclic LH release as Zimbelman (1966) proposed, thereby explaining the elevated blood levels. Pituitary LH concentration values, however, did not indicate LH release, as discussed previously. Furthermore, all heifers were supposedly slaughtered in a similar hormonal condition, which would not support this explanation.

As an approximation of the total LH content in the blood, plasma volume, as estimated at 3.5 percent of a heifers's slaughter weight, was multiplied by the concentration values. Granted this procedure is subject to error, it provided an estimate of the total blood LH content. With this mathematical calculation, it was found, as shown in Table 19 and Appendix IV, that heifers slaughtered at first estrus which had been fed MGA, and two of the three groups slaughtered at breeding size which had received MGA had greater total plasma LH contents ($P<0.10$) than heifers which had not been fed the drug. These data simply reflect the plasma concentration data, and indicate that more LH was available to the end organs in the heifers fed MGA. However, since all heifers were slaughtered at the same stage of the estrous cycle, the effects of MGA on LH values at other times in the estrous cycle are not known.

Plasma prolactin data as shown in Table 19 present a different picture from that of the LH plasma concentration. Data at first estrus reveal that heifers fed the normal level without MGA had the largest concentration ($P<0.01$) of prolactin. This occurrence might coincide with a period of rapid mammary gland growth. Sinha and Tucker (1969) suggest this explanation for their data on pituitary prolactin concentration of 9-month old heifers, the approximate age of normal level heifers at first estrus in this study. At breeding size the concentration value of heifers fed the normal level without MGA was significantly greater ($P<0.025$) than the value of heifers fed the normal level plus MGA. This finding suggests an inhibitory effect of MGA on plasma prolactin concentration when administered only with the normal nutritional level. Such an action does not seem plausible. It may be that the elevated value at breeding size of heifers fed the normal level without MGA was associated in some way with mammary gland development. Such an explanation supports Sinha and Tucker's (1969) pituitary prolactin data on 12-month old heifers.

Total prolactin content in the blood, like LH content, reflects the concentration data already discussed. That the heifers fed the normal level without MGA had the largest value ($P<0.01$) at first estrus, and a value significantly greater ($P<0.025$) than that of heifers fed the normal level plus MGA at breeding size suggests that prolactin was performing, or at least associated with, some function in heifers fed the normal nutritional level. Whether this function concerned mammary growth or something else is not known.

4. Plasma to Pituitary Hormone Content Ratios

To obtain an indication of the release to storage ratio for LH, total plasma content was divided by the total anterior pituitary content. As shown in Table 20 and Appendix IV, the ratio value increased from two to

TABLE 20.--Plasma to pituitary hormone ratios of Holstein heifers fed different nutrition treatments and slaughtered at 2.5 months, first estrus, or breeding size.[a]

Nutrition treatment	Time of slaughter	LH[b]	Prolactin[c]
Normal	2.5 months	7.64 ± 0.96	2.41 ± 0.43
Normal	First estrus	12.22 ± 1.97[d]	5.52 ± 0.66[e]
High	First estrus	23.29 ± 4.82	2.73 ± 0.60
High + MGA from 2.5 mo.	First estrus	19.11 ± 2.15	2.16 ± 0.42
Normal	Breeding size	13.85 ± 1.97[d]	7.65 ± 1.45[f]
Normal + MGA from 2.5 mo.	Breeding size	23.97 ± 4.09	4.17 ± 0.94
High	Breeding size	23.72 ± 5.32	6.52 ± 1.51
High + MGA from 2.5 mo.	Breeding size	19.84 ± 5.10	7.46 ± 1.47
High + MGA after first estrus	Breeding size	26.25 ± 3.63	8.14 ± 1.43

[a]Values are means and their standard errors for 10 heifers, except high treatment first estrus values which are for 20 heifers.

[b]ug plasma LH ÷ mg pituitary LH.

[c]ug plasma prolactin ÷ ug pituitary prolactin.

[d]Significantly less than the other treatment values ($P<0.10$).

[e]Significantly greater than the other treatment values ($P<0.01$).

[f]Significantly greater than the normal + MGA treatment value ($P<0.10$).

three fold between 2.5 months and first estrus, but then remained about the same within a nutritional treatment between first estrus and breeding size. This suggests that after first estrus occurs, the plasma and pituitary LH contents establish a certain ratio which does not change, at least up to breeding size. The data suggest a nutritional level effect on the LH ratio. At both first estrus and breeding size the value of heifers fed the normal level without MGA was significantly smaller ($P<0.10$) than the other nutritional treatment values, suggesting that a lower ratio is associated with a normal level of nutrition. However, when MGA was fed with the normal level, the hormone content ratio was comparable to that of heifers fed the high nutritional level, without or with MGA, which does not fit the ratio-nutritional level hypothesis.

Plasma-pituitary prolactin ratios (Table 20) show that the first estrus value of heifers fed the normal level without MGA was significantly greater ($P<0.01$) than the other nutritional treatment values. But, at breeding size heifers fed the normal level without MGA had a value which was significantly greater ($P<0.10$) than those fed the normal level plus MGA. The ratio values for each nutritional treatment increased from first estrus to breeding size, which was different from the LH data. Also, the prolactin ratios show generally the reverse patterns of the LH ratios, suggesting different regulatory pathways for the two hormones.

5. Correlations Between Anterior Pituitary and Plasma Hormone Values

To study pituitary and plasma hormone data and find a singificant pattern existing between them would be an endocrinologist's desire. Such findings were hoped for in this study. However, when pituitary LH and prolactin concentrations and also total pituitary contents were correlated with their respective plasma concentration values for the various nutritional treatments at the three different slaughter ages, no significant

correlations existed ($P>0.05$). In fact, of the 36 coefficients calculated only the one between pituitary and plasma prolactin concentrations for heifers fed the high level of nutrition plus MGA after first estrus and slaughtered at breeding size approached significance. It was a positive correlation suggesting that plasma prolactin concentration changed in the same direction as pituitary prolactin concentration. So because of the nonsignificant correlations, no apparent conclusions can be made which relate pituitary and plasma hormone levels.

F. Data on the Bred Heifers

1. Estrous Cycle Data

Lengths of the estrous cycles were recorded for all heifers kept beyond first estrus and not fed MGA. They ranged from 17 to 24 days, the usually accepted normal range, for all heifers except one. She consistently had cycles that ranged from 29 to 32 days in length. A few heifers developed cystic corpora lutea which resulted in abnormally long cycles. If the cysts did not spontaneously recover in 30 to 60 days, the heifers were given a 5000-IU injection of human chorionic gonadotropin intramuscularly to hasten recovery. This treatment seemed beneficial. Cycle lengths were not affected by the normal and high nutritional levels, and the first cycle was of the same length as all subsequent ones.

When heifers fed the high level plus MGA reached breeding size, MGA was withdrawn from their ration. Those that had received MGA from 2.5 months averaged 19.7 days after withdrawal before estrus occurred. Four heifers were in estrus on the second or third day after withdrawal, while the other six did not exhibit estrus until 12, 21, 27, 28, 35, and 64 days after withdrawal. Of the heifers that did not receive MGA until after first estrus, six were in estrus on the second or third day, while the

other four went 5, 10, 16, and 30 days from withdrawal to first estrus. The 10 heifers had a mean interval of 7.7 days between withdrawal and estrus. This value, however, was not significantly different ($P>0.10$) from the value of 19.7 days for heifers fed MGA from 2.5 months. Clearly, some of the long cycles could have resulted from silent or missed estrual periods, but an effect from prepuberal MGA administration is suggested. Still, once heifers started cycling, the cycles were of normal duration. Perhaps commencing long term MGA administration prepuberally results in a longer carryover effect in certain heifers than the approximately 2 to 7 days observed by Zimbelman and Smith (1966a) after a 16-day administration period. Or maybe the pathways for eliminating MGA from the body are not as functional in certain heifers as in others. And it is possible that the hypothalamus requires a longer recovery time in certain heifers. Whatever the cause or causes, the situation demands further investigation.

2. Breeding Data

At first breeding, heifers fed the normal level without MGA were significantly older ($P<0.05$) than heifers fed the other nutritional treatments (Table 21 and Appendix I). Also, heifers fed the high level without MGA were significantly younger ($P<0.05$) at first breeding than those fed the high level plus MGA. Since this significant age difference did not exist at breeding size (Table 21), it apparently resulted from the interval between MGA withdrawal and first breeding as discussed in the previous section. The time interval between breeding size and first breeding (Table 21 and Appendix I) for heifers fed a normal or high level was caused by the time lapse after reaching breeding size until the heifers were observed in estrus and bred. Because of this time interval, withers heights at first breeding exceeded 120 cm. Withers height at first breeding, however, did not differ among the nutritional treatment groups, but body

TABLE 21.--Age and body size at breeding size and first breeding of Holstein heifers fed different nutrition treatments.[a]

Nutrition Treatment	Age at breeding size	First breeding		
		Age	Weight	Height
	----------(mo)----------		(kg)	(cm)
Normal	12.5 ± 0.3[b]	13.1 ± 0.4[c]	363 ± 9	121.2 ± 0.2
High	11.0 ± 0.2	11.3 ± 0.2[c]	358 ± 11	121.0 ± 0.3
High + MGA from 2.5 mo.	11.4 ± 0.4	12.2 ± 0.4	401 ± 12[d]	121.3 ± 0.4
High + MGA after first estrus	11.7 ± 0.4	12.1 ± 0.4	410 ± 6[d]	120.7 ± 0.2

[a]Values are means and their standard errors for 10 heifers.

[b]Significantly different from the other treatment values ($P<0.01$).

[c]Significantly different from the other treatment values ($P<0.05$).

[d]Significantly different from the normal and high treatment values ($P<0.01$).

weight revealed the stimulatory affect of MGA as noted previously. Heifers fed the normal or high level without MGA weighed the same at first breeding.

As emphasized previously in the section on growth, that heifers fed the normal level were bred by about 13 months of age is most exciting. If dairy farmers could be challenged to feed their heifers at a level similar to the normal nutritional level in this study, the dairy industry would benefit greatly.

Breeding data for only the heifers that conceived are shown in Table 22 and Appendix V. Due to apparent infertility, only 36 of the 40 heifers conceived. One heifer fed the high level plus MGA after first estrus finally conceived to the eleventh service when she was about 2 years old. Because of her age, she was excluded from the data in Table 22. The four infertile heifers were bred 10 times to one bull and

TABLE 22.--Age and conception data for Holstein heifers fed different nutrition treatments.[a]

Nutrition treatment	Number heifers	Age at breeding size	Age at first breeding	Age at conception[f]	Services per conception[f]
Normal	9	12.6 ± 0.3[b]	13.3 ± 0.4[d]	14.7 ± 0.7	2.3 ± 0.7
High	9	10.9 ± 0.2[c]	11.3 ± 0.2[e]	13.4 ± 0.7	3.2 ± 0.9
High + MGA from 2.5 mo.	10	11.4 ± 0.4	12.2 ± 0.4	14.7 ± 0.7	3.4 ± 0.7
High + MGA after first estrus	7	12.2 ± 0.4	12.6 ± 0.4	14.6 ± 0.8	3.0 ± 0.8

[a]Values are means and their standard errors.

[b]Significantly different from the high and high + MGA from 2.5 mo. treatment values ($P<0.01$).

[c]Significantly different from the high + MGA after first estrus treatment values ($P<0.01$).

[d]Significantly different from the other treatment values ($P<0.01$).

[e]Significantly different from the high + MGA treatment values ($P>0.05$).

[f]No significant differences in the values ($P>0.10$).

an eleventh time to a different bull. Since they were still not pregnant, they were slaughtered. All four had some reproductive tract disorder which could have caused the infertility. Two cases of chronic endometritis, one extremely fibrous endometrium, and one case where an ovary was surrounded by fibrous tissue may have caused the infertility.

Although age at first breeding differed among the nutritional treatments, age at conception (Table 22 and Appendix V) did not differ significantly ($P>0.10$). This was the result of more services per conception, though the difference was not significant ($P>0.10$), for heifers fed the high level and high level plus MGA. Because of the lack of a significant difference in services per conception among the nutritional treatments, one must conclude the high level and high level plus MGA had no detrimental effect on conception. This agrees with the results of Reid et al., (1964) and several others as cited in the literature review.

However, since heifers fed the various nutritional treatments were of different ages at first breeding but not at conception, an effect of nutritional level on conception is strongly implicated. Perhaps the increased amount of pelvic area fat observed in heifers fed the high level and high level plus MGA that were slaughtered at breeding size also existed in heifers that were bred, and thereby in some manner affected fertility. Since heifers fed the normal level required more services than expected, perhaps the outbreak of IBR in the herd when the hefiers were about 9 months old increased the services required per conception by all nutritional treatment groups. However, heifers were vaccinated for IBR and seemingly recovered within a month with no after effects. Although heifers fed MGA with the high level of nutrition required as many services per conception and as many heifers conceived to the first service as did heifers fed the high level, data from our laboratory using rabbits

(Pritchard et al., 1969) and that data of Quinlivan and Robinson (1969) with ewes suggest that MGA may inhibit sperm transportation at the first service after withdrawal of the progestagen.

3. Parturition Data

Of the 36 heifers that conceived, parturition data were available on only 34 of them. Observations at parturition are shown in Table 23 and Appendix V. In studying the data, it should be remembered that all weights, withers heights measurements, and subjective ratings of dystocia were made by the workmen at Driggs Dairy. Although some reservation may exist as to accuracy, the data are worthy of examination. Suffice it to say at this point that the nutritional treatments produced no significant differences ($P>0.10$) in the data for any category listed in Table 23 and Appendix V.

From the dams' weights before and after parturition and dystocia ratings, it appears that the smaller heifers encountered more difficulty at parturition. However, these heifers were not any smaller in skeletal size as indicated by withers heights. Correlating the dams' weights before and after parturition with dystocia ratings indicated no significant correlation ($P>0.05$). Thus, size of dam had no apparent effect on calving difficulty. Calf birth weight and dystocia rating data hint that the heavier calves were associated with a more difficult parturition. This in fact was true, for the correlation between these two parameters was highly significant ($P<0.01$). When birth weights of the calves are expressed as a percentage of both the pre- and postpartum dam weights, the data suggest that when calf weight as a percent of the prepartum dam weight increases, the calving difficulty rating increases. However, only the correlation coefficient between calf weight as a percent of postpartum dam weight and dystocia rating was significant ($P<0.05$). Thus, one can

TABLE 23.--Some observations at parturition of Holstein heifers fed different nutrition treatments prior to conception.[a]

Criterion	Nutrition treatment			
	Normal	High	High + MGA from 2.5 mo.	High + MGA after first estrus
Number of heifers	9	8	10	7
Age (mo)	23.4 ± 0.7	22.0 ± 0.7	23.5 ± 0.7	23.5 ± 0.8
Weight before parturition (kg)	504 ± 17	540 ± 10	517 ± 19	505 ± 20
Weight after parturition (kg)	460 ± 14	475 ± 15	486 ± 18	460 ± 20
Withers height (cm)	128.7 ± 0.9	129.8 ± 1.1	128.4 ± 0.9	128.6 ± 0.9
Dystocia rating	2.4 ± 0.5	1.6 ± 0.4	1.5 ± 0.2	1.9 ± 0.5
Calf weight (kg)	39 ± 3	40 ± 2	36 ± 2	44 ± 4
Calf weight ÷ dam				
prepartum weight	0.080 ± 0.005	0.075 ± 0.004	0.071 ± 0.006	0.089 ± 0.006
postpartum weight	0.083 ± 0.007	0.087 ± 0.008	0.077 ± 0.007	0.099 ± 0.008

[a]No significant differences in the treatment values for any category ($P>0.10$). Values are means and their standard errors.

conclude from the data in Table 23 and Appendix V, that larger calves, both in actual birth weight and as a percent of the postpartum dam weight, are associated with more difficulty at parturition. These findings are not new and could have been anticipated.

4. Calving Data by Sire

One of the objectives of this study was to determine the effect of sire on calf size and calving difficulty. The calving data as presented in Table 24 give meaningful assistance towards answering our objective. Although the difference was not significant, calves sired by Royal Pontiac weighed more than those sired by Wis Symbol (41 $\pm$ 2 vs 38 $\pm$ 2 kg). This was true for actual birth weights and when the calf weights were expressed as a percentage of the dams' pre- and postpartum weights. Furthermore, this was always the case, whether considering all calves sired by Pontiac or dividing them into male and female calves. Dystocia ratings also revealed that more calving difficulty was encountered with calves sired by Royal Pontiac than those sired by Wis Symbol. The data suggest male calves were associated with a higher incidence of dystocia, presumably because they weighed more. However, only female birth weights and dystocia ratings were correlated significantly ($P<0.05$). This would suggest that perhaps something about male calves besides their weight, perhaps bone structure, influences the degree of difficulty encountered at parturition. These findings illustrate the influence of sire on calf size, as shown by Boyd and Hafs (1965), and thereby show indirectly that the sire a cow is bred to can affect the degree of dystocia she will encounter at parturition.

5. Milk Production Data

The milk production data are shown in Table 25 and Appendix V. Since the mammary nucleic acid data at breeding size (Table 15 and Appen-

TABLE 24.--Calving data by sire.[a]

	Sire	
	Royal Pontiac	Wis Symbol
Birth weight (kg):		
all calves	41 ± 2	38 ± 2
male calves	43 ± 3 (10)[b]	42 ± 3 (5)
female calves	38 ± 2 (8)	36 ± 3 (9)
Dystocia ratings:		
all calves	2.4 ± 0.3[c]	1.3 ± 0.2
male calves	3.0 ± 0.4	1.6 ± 0.4
female calves	1.4 ± 0.3	1.2 ± 0.2
Calf weight ÷ dam prepartum weight:		
all calves	0.078 ± 0.004	0.076 ± 0.005
male calves	0.081 ± 0.006	0.080 ± 0.008
female calves	0.075 ± 0.004	0.073 ± 0.005
Calf weight ÷ dam postpartum weight:		
all calves	0.082 ± 0.006	0.079 ± 0.005
male calves	0.093 ± 0.008	0.086 ± 0.009
female calves	0.082 ± 0.008	0.075 ± 0.006
Dam weight (kg):		
before parturition	519 ± 11	514 ± 15
after parturition	462 ± 10	482 ± 14
Dam withers height	129.0 ± 0.7	127.9 ± 0.6

[a]Values are means and their standard errors.

[b]Number of calves in parentheses.

[c]Significantly different from the other sire value ($P<0.025$).

TABLE 25.--Milk production data of Holstein heifers fed different nutrition treatments prior to conception.[a]

Nutrition treatment	Number heifers	First 60 days of lactation[b]	Extended 305 day production[b]
		----------(kg)----------	
Normal	8	1038 ± 47	4194 ± 158
High	8	979 ± 42	4088 ± 184
High + MGA from 2.5 mo.	9	931 ± 78	3746 ± 292
High + MGA after first estrus	6	1004 ± 50	4056 ± 216

[a]Values are means and their standard errors.

[b]No significant differences in the values ($P>0.10$).

dix III) indicated the heifers fed MGA were commencing pregnancy with considerably more parenchymal tissue, it was anticipated that this difference would result in greater milk production after parturition. But since no significant differences ($P>0.10$) existed among the nutritional treatment groups in the actual first 60 day milk production weights or in the estimated 305 day production values, such an occurrence did not happen.

Perhaps the mammary gland growth that resulted from feeding MGA before the heifers were bred regressed during the first few months of pregnancy. Since MGA was not fed during pregnancy, the hormonal stimulus required for mammary proliferation was perhaps not sufficient the first several months after conception. Consequently, the enlarged glands may have regressed in development, and therefore heifers fed all nutritional treatments entered the latter half of pregnancy with the same degree of gland development. Although the milk production values in Table 25 and Appendix V were not significantly different ($P>0.10$), the data hint that heifers fed the normal level were better milk producers. The lower milk production by heifers fed the high level of nutrition supports the contention of Swanson (1960) that feeding above normal nutritional levels during growth and pregnancy results in lowered milk production. Since heifers in this study were not fed at an elevated level during pregnancy, this may explain why we did not get the dramatic difference in milk production that Swanson obtained. Perhaps it is the feeding of high nutritional levels during pregnancy that is associated with lowered milk production.

G. Nitrogen Balance Trials

Unpublished data of the Upjohn Company show that feedlot heifers fed MGA gain faster than controls, but the increase is due partly to protein deposition and not solely to the accumulation of depot fat. Knowing this,

it was decided to conduct nitrogen balance trials on several of the heifers used in this study to ascertain if indeed heifers fed MGA were retaining more protein from their ration. Postpuberal cycling heifers ranging from 7 to 13 months of age were placed in metabolism stalls a few days after an estrus and allowed to acclimate for 5 to 10 days. Heifers of comparable age and size fed MGA were put in the stalls at the same time. They were fed their regular level of grain plus corn silage and hay ad lib. Data were collected for a 7-day period on the amount of feed consumed, and feces and urine excreted. Samples of the feed offered, feed not eaten, feces, and urine were collected and analyzed for nitrogen content by the Kjeldahl method. The nitrogen values were then converted to protein equivalent.

The data obtained in Trial 1 are shown in Table 26. Heifers fed the normal level plus MGA retained about the same amount of protein daily as heifers fed the normal level without MGA. These data support the growth data which showed little or no stimulatory effect of MGA on weight increase when fed with the normal level of nutrition. But, data for heifers fed the high level plus MGA are very perplexing. At least during the collection period, heifers fed the high level plus MGA retained considerably less protein from the daily ration than heifers fed just the high level. This seemingly conflicts with growth and carcass evaluation data. Also, heifers fed the high level plus MGA excreted more protein in the urine as well as more urine than heifers fed the high level without MGA.

To obtain further data on this phenomenon, an experiment was designed in which each of four heifers was to be fed both the normal and high nutritional levels, without and with MGA. Four regular university herd heifers about 12 months old with the stage of their estrous cycles unknown were placed in the metabolism stalls and allowed 2 weeks to

TABLE 26.--Nitrogen balance data of Holstein heifers fed different nutrition treatments.[a]

Trial 1.

Nutrition treatment	Number heifers	Age	Per day: Protein consumed	Fecal protein	Urine protein	Protein retained	% protein retained	Urine volume
		(mo)	(g)					(g)
Normal	7	9.0	468	176	217	74	15.8	3600
Normal + MGA from 2.5 mo.	6	8.6	493	253	159	82	16.6	3727
High	10	9.2	870	241	403	227	26.1	4685
High + MGA from 2.5 mo.	9	9.7	943	254	529	95	10.1	8953

Trial 2.

Heifer number	Nutrition treatment	Per day: Protein consumed	Fecal protein	Urine protein	Protein retained	% protein retained	Urine volume
		(g)					(g)
1020	Normal	700	300	181	219	31.3	2347
	Normal + MGA	681	231	200	250	36.7	2420
1023	Normal	556	256	400	-100	-18.0	7673
	Normal + MGA	669	275	231	163	24.5	8204
1022	High	1187	437	244	506	42.6	10002
	High + MGA	1194	469	606	119	10.0	12340
1021	High	1294	500	369	425	32.8	2969
	High + MGA	1069	475	337	257	24.0	2561

[a]Values are means.

acclimate to both the stalls and the first ration treatment. Intake and excreta samples were collected for 5 days, followed by 7 days adjustment to the second treatment before another 5-day collection period. Midway through this trial, we were advised by researchers from the Upjohn Company that the data could be invalid because about 3 weeks are required after MGA administration commences for assurance that no corpora lutea are functional. If any were functional, they would prevent follicular growth and hormone secretion. Consequently, the experiment was terminated after collecting data for only two heifers fed the normal nutritional level without and with MGA, and two heifers fed the high nutritional level without and with MGA. However, the data obtained in Trial 2, as shown in Table 26, with each heifer serving as her own control, indicate the same effect of MGA on protein retention as did the results from Trial 1. At this point the data are presented without any logical explanation of the mechanism of action. To speculate, perhaps since glucocorticoids cause an increased elimination of urinary nitrogen (Turner, 1960), and since MGA has glucocorticoid activity, maybe this action is involved. But, elevated urine protein and lowered protein retention were observed only when MGA was fed with the high level of nutrition. Perhaps the amount of protein consumed and MGA have some type of interaction. However, since the number of heifers used in these trials were small, perhaps animal variation caused the results obtained. Obviously additional research on this topic is needed to determine, at least for academic reasons, the effects and mode of action of MGA on protein retention.

SUMMARY AND CONCLUSIONS

This study was conducted to determine the effects of a normal and high level of nutrition alone or with the synthetic progestagen melengestrol acetate (MGA) on body growth, levels of certain anterior pituitary hormones in the pituitary and blood, development of the reproductive tract and mammary gland, and subsequent reproductive and lactational performance of 140 Holstein heifers. Heifers were raised under uniform conditions from 2 weeks to 2.5 months of age at which time they were randomly assigned to 14 treatment groups consisting of 10 heifers each. One hundred heifers were slaughtered either at 2.5 months of age, at first estrus or at breeding size, while 40 heifers fed a roughage ration only between pregnancy diagnosis and parturition were kept to obtain data on breeding and lactational performances.

Heifers fed the high level of nutrition exhibited first estrus at a significantly younger ($P<0.01$) age than those fed the normal level (7.5 ± 0.1 vs 8.7 ± 0.2 months), but there was no significant difference ($P>0.10$) in body weight (255 ± 4 vs 250 ± 5 kg) or withers height (108.6 ± 0.6 vs 109.2 ± 0.7 cm). These data emphasize that first estrus is associated more with physical maturity than with calendar age.

At breeding size (120 cm withers height), heifers fed the high level of nutrition were 11.4 ± 0.4 months old while those fed the normal level were 12.5 ± 0.2 months old ($P<0.01$). MGA fed at the rate of 0.45 mg per heifer per day with either the normal or high levels of nutrition did not significantly affect the ages at breeding size, indicating that MGA did not

affect skeletal growth. However, MGA increased body weight gains, but only when fed with the high level of nutrition ($P<0.01$). Heifers fed the high level of nutrition with MGA gained faster ($P<0.05$) after about 5.5 months of age than heifers fed the high level alone. At breeding size, heifers that had been fed the high level plus MGA weighed about 35 kg more than heifers fed either the normal level, normal level plus MGA, or high level of nutrition.

The time from first estrus to breeding size (about 3.5 months) was not significantly different ($P>0.10$) for heifers fed either level of nutrition without or with MGA, indicating that level of nutrition or addition of MGA did not affect rate of skeletal growth after first estrus.

Uterine weights for the various nutrition groups at first estrus or at breeding size were not significantly different ($P>0.10$) when expressed per 100 kg body weight. Uterine nucleic acids concentrations at both first estrus and breeding size showed no significant differences ($P>0.10$) in the normal and high nutritional level values. But, when MGA was fed with both the normal and high levels, DNA concentration was generally lower and RNA concentration generally higher than when the compound was not fed. These data implicate uterine hypertrophy associated with MGA feeding. Also suggestive of uterine hypertrophy were the increased uterine epithelial cell heights, but only at breeding size, in heifers fed MGA. Ovarian weights were not affected by the nutritional treatments, but more large diameter follicles were present on the ovaries of heifers fed MGA.

Weights of the dissected parenchymal tissue from one half of the mammary gland were not affected by the nutritional treatments at either first estrus or breeding size. Nucleic acids concentrations at first estrus suggested an elevated DNA value and showed a significantly increased ($P<0.01$) RNA value in heifers fed MGA. At breeding size both DNA and RNA

concentration values were significantly increased ($P>0.01$) in heifers fed MGA. MGA fed with the normal level of nutrition increased total mammary DNA content about 30 percent and RNA content about 38 percent over the values for heifers fed the normal level alone. This stimulatory action of MGA was twice as great when it was fed with the high level of nutrition as compared to when it was fed with the normal level.

Neither the paired adrenal weights nor the weights expressed per 100 kg body weight were affected significantly ($P>0.10$) by the various nutritional treatments. However, at breeding size, MGA caused a significant decrease ($P<0.05$) in the width of the glucocorticoid producing fasciculata zone of the cortex. Since MGA is known to have glucocorticoid activity, some direct or indirect regulatory action of MGA on the adrenal cortex is suggested.

The various nutritional treatments produced no significant differences ($P>0.10$) in total, anterior, or posterior pituitary weights at first estrus, but heifers fed the high level of nutrition plus MGA had significantly larger ($P<0.10$) anterior pituitary weights per 100 kg body weight at breeding size than the other nutritional treatment values.

No large dramatic differences in pituitary of plasma concentrations of LH, FSH, and prolactin resulted at first estrus or breeding size from feeding the various nutritional treatments.

Correlations between pituitary concentration and plasma concentration, and between pituitary content and plasma concentration for LH and prolactin for all nutritional treatments at the three slaughter times were not significant ($P>0.05$).

The interval from MGA withdrawal until the heifers came in estrus was considerably longer, though not significantly so ($P>0.10$), for heifers that received the compound from 2.5 months than for those that received

MGA only after first estrus (19.7 vs 7.7 days). Still, once estrous cycles commenced, they were of normal length (17-24 days) for all MGA treated animals.

Heifers that were bred and conceived were 14.7 ± 0.7, 13.4 ± 0.7, 14.7 ± 0.7, and 14.6 ± 0.8 months old at conception for normal level, high level, high level plus MGA from 2.5 months, and high level plus MGA from first estrus nutritional treatment groups, respectively. The number of services required per conception was 2.3 ± 0.7, 3.2 ± 0.9, 3.4 ± 0.7, and 3.0 ± 0.8 for the preceding respective nutritional treatments. These different nutritional treatment values for age at conception or services per conception were not significantly different ($P>0.10$).

At parturition no significant differences ($P>0.10$) were found in body weights, withers heights or subjective dystocia ratings of the dams fed the various nutritional treatments prior to conception. Calf birth weights were not significantly different ($P>0.10$) for the two sires or the various nutritional treatments fed the dams prior to conception. But, the positive correlations between calf birth weight and dystocia rating, and the calf birth weight as a percentage of the dam's postpartum weight and dystocia rating were significant ($P<0.05$).

Neither the actual milk production weights for the first 60 days of lactation nor the extended 305 day values were significantly different ($P>0.10$) for heifers fed the various nutritional treatments prior to conception.

Preliminary nitrogen balance trials revealed no effect on protein retention when MGA was fed with the normal level of nutrition. This agreed with weight gain data. But, feeding MGA with the high level of nutrition resulted in increased urine protein loss and a reduction in the amount of protein retained daily from the ration when compared to high level controls.

These data do not agree with growth and slaughter data.

So, from the data accumulated in this study, certain general conclusions can be made. Growth rate is definitely affected by the level of nutrition; a high level accelerates weight gain but has little influence on rate of skeletal growth; and heifers fed a normal level, that is corn silage and hay free choice plus a small amount of grain daily, can grow to breeding size by 13 months of age. The reproductive tract is sufficiently mature at this age to permit breeding of the heifers. MGA does not affect skeletal growth and it accelerated weight gains only when fed with a high level of grain. No gross effect on hormone levels resulted from feeding MGA or the high level of nutrition. If concention rate were better than obtained in this study, feeding growing heifers a high level of nutrition may be advantageous. But, from this experiment, since all heifers were the same age at first parturition, it is obvious that the high level is not practical. And finally, feeding an above normal level of nutrition without or with MGA prior to concention appears to have no effect on subsequent lactational performance.

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APPENDICES

APPENDIX I.--Age, body weight, and withers height of individual heifers at 2.5 months, first estrus, breeding size, first breeding, or slaughter.

Group No.	Heifer No.	2.5 Months		First Estrus		
		Weight	Withers Height	Age	Weight	Withers Height
		(kg)	(cm)	(mo)	(kg)	(cm)
1	206	74	80.0	8.0	209	102.0
	209	77	84.0	11.5	318	118.0
	212	120	94.0	8.8	270	108.5
	215	77	87.0	9.5	209	109.0
	232	85	87.0	10.7	232	105.5
	250	118	88.0	8.5	270	114.0
	273	85	84.0	9.2	225	109.5
	275	94	84.0	8.3	234	107.0
	284	118	87.0	7.7	234	108.0
	286	121	87.5	8.9	280	111.5
Mean ± SE		97±6	86.2±1.1	9.1±0.4	248±11	109.3±1.4
2	203	91	89.0	6.6	227	104.0
	205	93	84.0	9.7	293	116.0
	207	89	84.5	7.0	236	102.5
	210	98	90.0	7.1	266	111.0
	213	103	85.0	7.8	277	109.0
	257	82	85.0	6.6	234	105.0
	268	126	87.0	9.0	302	112.0
	270	85	81.0	10.1	311	116.0
	283	100	88.0	8.8	277	114.0
	288	81	92.0	8.8	277	116.0
Mean ± SE		95±4	86.6±1.0	8.1±0.4	270±9	110.6±1.6

Breeding Size		First Breeding		
Age (mo)	Weight (kg)	Age (mo)	Weight (kg)	Withers Height (cm)
14.0	382	15.2	426	122.5
13.0	370	13.0	370	120.0
12.1	375	12.6	393	121.0
12.6	323	13.2	336	121.0
12.8	341	13.5	365	122.0
11.2	332	12.0	342	121.5
14.0	323	15.0	357	122.0
12.2	350	12.8	359	121.0
11.4	314	12.3	329	121.0
11.3	355	11.8	348	120.0
12.5±0.3	346±8	13.1±0.4	363±9	121.2±0.2
10.4	343	10.5	347	120.5
10.7	316	11.0	325	121.0
12.0	364	12.2	381	121.0
10.3	341	11.0	355	122.0
11.3	370	12.0	402	122.0
11.7	414	11.7	414	120.0
11.4	361	11.8	372	121.0
11.5	352	11.5	352	120.0
10.7	321	10.7	318	120.0
9.8	300	11.0	314	123.0
11.0±0.2	348±10	11.3±0.2	358±11	121.0±0.3

Group No.	Heifer No.	2.5 Months		First Estrus		
		Weight	Withers Height	Age	Weight	Withers Height
		(kg)	(cm)	(mo)	(kg)	(cm)
3	208	98	87.0	6.5	239	105.0
	211	96	91.5	7.0	282	108.5
	220	86	85.5	7.7	234	103.0
	231	106	85.5	8.5	361	115.5
	236	87	85.0	6.8	227	103.5
	259	91	87.0	9.0	323	115.0
	261	95	88.0	9.1	334	113.0
	269	84	85.5	8.6	275	109.5
	272	105	85.0	9.3	343	117.0
	280	112	86.5	6.7	230	106.0
Mean ± SE		96±3	86.6±0.6	7.9±0.3	290±17	109.6±1.6
4	218	121	90.0	7.0	273	111.0
	222	77	91.5	6.2	245	107.0
	225	82	91.0	6.1	232	106.0
	235	105	85.0	7.8	275	109.0
	239	101	82.0	8.3	291	107.0
	249	92	85.0	8.1	250	106.0
	258	102	86.0	8.3	245	108.5
	274	105	75.0	8.7	243	104.0
	281	89	76.0	9.4	284	109.5
	287	112	80.0	8.5	225	105.5
Mean ± SE		99±4	84.1±1.9	7.8±0.3	256±7	107.3±0.7

Breeding Size		First Breeding		
Age (mo)	Weight (kg)	Age (mo)	Weight (kg)	Withers Height (cm)
10.1	368	10.7	383	121.0
10.3	380	10.7	393	121.0
14.0	416	14.0	416	120.0
12.0	455	13.0	491	122.0
11.8	377	12.8	401	121.0
10.6	370	13.3	409	125.0
11.4	384	12.0	390	121.0
11.8	345	12.8	357	120.5
11.7	395	12.0	406	120.5
9.9	357	10.5	365	121.0
11.4±0.4	385±10	12.2±0.4	401±12	121.3±0.4
10.3	368	10.5	376	120.5
10.0	389	10.1	398	120.5
10.0	366	10.7	384	121.5
11.5	409	12.5	425	122.5
12.0	420	12.5	425	121.0
11.6	417	12.0	439	120.0
13.3	398	13.3	401	120.0
13.0	420	13.0	423	120.0
13.3	409	13.5	409	120.0
12.4	405	13.0	420	121.0
11.7±0.4	400±6	12.1±0.4	410±6	120.7±0.2

Group No.	Heifer No.	2.5 Months		First Estrus		
		Weight (kg)	Withers Height (cm)	Age (mo)	Weight (kg)	Withers Height (cm)
5	221	102	90.0	10.8	255	113.0
	226	120	89.0	8.0	234	107.5
	229	93	85.0	10.9	298	113.5
	238	90	87.0	8.0	234	104.0
	242	88	89.0	8.8	241	104.0
	251	114	79.0	8.8	248	109.5
	263	111	92.0	6.0	211	105.0
	264	88	87.0	10.3	266	116.0
	266	89	86.0	9.1	293	115.0
	278	89	84.0	10.2	236	111.0
Mean ± SE		98±4	86.8±1.1	9.1±0.5	252±9	109.8±1.4
6	204	96	93.0	7.6	280	111.0
	214	85	82.0	8.3	259	104.5
	217	100	84.5	6.5	202	102.5
	223	90	92.5	7.5	305	109.0
	241	118	88.0	7.6	270	104.5
	254	94	81.0	10.0	261	108.0
	255	126	89.0	8.0	259	116.5
	262	77	88.0	9.5	323	119.0
	279	102	81.0	6.6	202	100.0
	285	118	89.5	6.1	232	105.0
Mean ± SE		101±5	86.8±1.4	7.8±0.4	259±12	108.0±1.9

Breeding Size		Slaughter		
Age	Weight	Age	Weight	Withers Height
(mo)	(kg)	(mo)	(kg)	(cm)
12.0	336	13.7	366	123.0
13.5	368	13.8	366	121.0
13.0	398	13.3	382	121.0
13.3	377	14.0	382	122.0
13.5	350	13.5	350	120.0
13.0	364	14.0	393	121.5
11.5	361	11.5	361	120.0
12.0	302	12.0	302	120.0
11.0	341	11.0	341	120.0
14.0	341	15.0	370	121.5
12.7±0.3	354±8	13.2±0.4	361±8	121.0±0.3
10.6	339	11.2	361	121.0
14.3	405	14.3	404	120.0
12.7	348	12.7	348	120.0
10.7	382	11.8	391	121.0
12.0	368	12.5	377	120.5
16.5	373	16.5	373	118.0
9.7	323	10.0	316	121.0
9.7	325	10.2	336	121.5
12.1	382	12.1	382	119.5
9.3	314	9.8	323	121.0
11.8±0.7	356±10	12.1±0.6	361±8	120.3±0.3

Group No.	Heifer No.	2.5 Months		First Estrus		
		Weight	Withers Height	Age	Weight	Withers Height
		(kg)	(cm)	(mo)	(kg)	(cm)
7	202	119	89.0	7.5	289	106.0
	219	96	85.5	6.3	198	103.5
	230	103	85.5	8.5	305	109.0
	237	87	87.0	7.5	261	106.0
	240	92	88.0	7.6	280	103.0
	253	98	85.0	6.0	195	100.0
	256	102	90.0	9.7	368	115.0
	260	80	81.0	6.5	150	97.0
	265	65	90.0	10.0	364	120.5
	282	80	96.0	8.0	311	118.5
Mean ± SE		92±5	87.7±1.3	7.8±0.4	272±23	107.8±2.5
8	201	90	85.0	7.9	280	106.0
	233	95	85.5	7.3	261	105.0
	234	102	89.0	7.8	268	107.0
	243	89	88.0	7.5	264	106.0
	246	91	86.5	7.3	270	106.5
	271	104	84.0	7.8	286	110.5
	276	95	84.0	7.9	248	107.0
	277	78	80.5	8.9	275	109.5
	289	107	88.0	6.9	241	113.0
	290	127	91.0	5.9	248	110.5
Mean ± SE		98±4	86.1±0.9	7.5±0.2	264±5	108.1±0.8

Breeding Size		Slaughter		
Age (mo)	Weight (kg)	Age (mo)	Weight (kg)	Withers Height (cm)
10.7	384	11.3	404	121.5
10.5	355	11.8	366	122.0
11.7	423	12.0	425	120.5
11.5	370	12.0	389	120.0
13.0	464	13.5	473	120.5
11.2	380	11.5	389	121.0
11.5	420	11.5	420	120.0
13.0	359	13.2	368	120.5
9.5	345	10.5	375	121.0
9.3	357	9.3	357	120.0
11.2±0.4	386±12	11.7±0.4	397±11	120.7±0.2
12.3	400	12.3	400	120.0
12.1	409	12.1	409	120.0
10.1	332	11.0	345	121.0
10.3	350	11.3	386	121.5
10.2	361	11.0	386	121.0
11.0	389	11.0	389	120.0
13.5	407	13.5	407	120.0
13.3	450	13.3	450	120.0
9.0	316	9.3	318	121.0
8.5	332	8.9	345	121.0
11.0±0.5	375±14	11.4±0.5	383±12	120.5±0.2

Group No.	Heifer No.	2.5 Months Weight (kg)	2.5 Months Withers Height (cm)	Breeding Size Age (mo)	Breeding Size Weight (kg)
9[a]	294	102	89.0	12.3	393
	305	100	91.0	12.3	373
	312	95	87.0	13.3	389
	318	105	89.0	13.0	375
	322	85	84.0	13.7	370
	344	88	89.0	12.8	343
	357	105	92.0	12.2	339
	364	94	93.0	10.7	302
	368	94	89.0	11.9	350
	369	100	89.0	9.3	298
Mean ± SE		97±2	89.2±0.8	12.1±0.4	353±11

[a]No contempory pairmates not fed MGA, so no first estrus values.

Group No.	Heifer No.	2.5 Months Weight (kg)	2.5 Months Withers Height (cm)	First Estrus Age (mo)	First Estrus Weight (kg)	First Estrus Withers Height (cm)
10	292	108	86.0	6.4	232	106.5
	293	114	86.0	6.1	227	107.0
	300	85	86.0	8.6	282	111.0
	311	73	82.0	8.9	277	111.0
	320	88	84.5	8.4	257	106.0
	354	85	85.0	9.4	255	110.0
	355	100	85.5	8.5	261	108.0
	363	71	84.0	9.1	255	111.0
	366	90	83.0	7.7	248	108.0
	373	85	88.0	7.4	227	107.0
Mean ± SE		90±4	85.0±0.5	8.0±0.3	252±6	108.5±0.6

Slaughter		
Age	Weight	Withers Height
(mo)	(kg)	(cm)
13.0	404	123.0
12.8	382	121.5
13.7	395	121.0
13.0	379	120.0
13.7	370	119.0
12.8	343	119.0
12.8	348	121.0
10.7	302	119.0
12.6	361	121.0
9.6	304	120.0
12.5±0.4	359±11	120.4±0.4

Slaughter		
Age	Weight	Withers Height
(mo)	(kg)	(cm)
7.0	234	107.5
6.7	239	108.0
9.2	273	112.0
9.5	286	112.0
9.0	261	107.0
10.0	268	115.0
9.1	275	112.0
9.7	261	112.5
8.3	252	109.0
8.0	227	108.0
8.7±0.3	258±6	110.3±0.8

Group No.	Heifer No.	2.5 Months		First Estrus		
		Weight	Withers Height	Age	Weight	Withers Height
		(kg)	(cm)	(mo)	(kg)	(cm)
11	298	84	83.5	5.9	182	99.5
	301	87	86.0	5.9	207	105.0
	308	86	82.0	7.1	248	107.0
	309	87	84.0	7.3	216	105.0
	315	91	84.5	5.7	214	102.5
	347	87	87.0	6.6	234	109.0
	350	112	95.0	6.5	252	111.0
	358	98	89.5	6.9	241	108.5
	352	107	91.0	7.2	261	110.0
	371	89	87.0	6.1	220	107.0
Mean ± SE		93±3	86.9±1.2	6.5±0.2	227±8	106.4±1.1
12	291	83	85.5	5.9	191	104.0
	295	98	85.0	5.7	216	107.0
	297	109	87.0	5.7	234	107.0
	302	89	83.5	7.1	264	107.5
	307	93	89.0	7.3	223	105.0
	353	96	86.0	6.2	214	102.5
	359	106	87.5	7.0	270	107.5
	360	108	89.0	6.5	234	104.0
	362	90	86.5	6.9	225	106.5
	365	89	89.0	7.2	255	111.5
Mean ± SE		96±3	86.8±0.6	6.5±0.2	233±8	106.2±0.8

Slaughter		
Age	Weight	Withers Height
(mo)	(kg)	(cm)
6.5	189	101.5
6.5	214	107.0
7.7	259	109.0
7.9	227	106.0
6.3	223	104.0
7.2	243	111.0
7.1	266	112.0
7.5	266	110.0
7.8	250	112.0
6.7	232	109.0
7.1±0.2	237±8	108.1±1.1
6.5	198	105.0
6.3	227	109.5
6.3	245	108.0
7.7	277	109.0
7.9	243	106.0
6.8	225	104.0
7.6	289	108.5
7.1	241	105.0
7.5	227	108.0
8.0	270	113.0
7.2±0.2	244±9	107.6±0.8

Group No.	Heifer No.	2.5 Months		First Estrus		
		Weight	Withers Height	Age	Weight	Withers Height
		(kg)	(cm)	(mo)	(kg)	(cm)
13	299	86	87.0	7.1	259	113.0
	303	75	83.5	10.1	343	118.0
	317	120	96.0	6.4	255	112.0
	319	81	81.0	6.4	214	104.0
	321	96	85.0	7.4	245	106.0
	349	103	89.5	6.4	232	108.5
	351	121	96.0	7.8	307	118.0
	356	110	92.0	6.2	243	110.0
	361	78	86.5	6.9	227	112.5
	367	75	85.0	8.0	230	108.0
Mean ± SE		94±6	88.1±1.6	7.3±0.4	255±13	111.0±1.5
14	296	87	84.0			
	310	89	82.5			
	313	92	85.5			
	314	102	87.0			
	316	93	83.0			
	342	125	95.0			
	343	93	87.0			
	345	107	88.0			
	346	80	81.0			
	348	86	86.0			
Mean ± SE		95±4	85.9±1.2			

Slaughter		
Age	Weight	Withers Height
(mo)	(kg)	(cm)
7.7	270	115.0
10.7	343	119.0
7.0	264	113.0
7.0	236	106.0
8.0	248	108.0
7.0	243	110.5
8.4	318	119.0
6.8	250	112.0
7.5	239	114.5
8.6	236	109.0
7.9±0.4	265±12	112.6±1.4

APPENDIX II.--Weight, nucleic acids, and cell height of the uterus, and ovarian weight and number of follicles for individual heifers.

Group No.	Heifer No.	Uterus				
		Weight	DNA		RNA	
		(g)	(mg)	(mg/g)	(mg)	(mg/g)
5[a]	221	129.0	583.7	4.5	494.6	3.8
	226	160.0	527.7	3.3	560.8	3.5
	229	123.6	567.0	4.6	428.7	3.5
	238	138.6	611.8	4.1	438.9	3.2
	242	123.5	513.1	4.1	392.5	3.2
	251	119.8	545.0	4.5	277.5	2.3
	263	121.0	525.5	4.3	422.6	3.5
	264	112.9	637.0	5.6	347.2	3.1
	266	194.7	706.7	3.6	710.5	3.6
	278	145.0	643.7	4.4	362.8	2.5
Mean ± SE		136.8±7.8	586.1±19.9	4.3±0.2	443.6±38.6	3.2±0.1
6[a]	204	181.7	690.2	3.8	619.6	3.4
	214	131.1	543.1	4.1	454.7	3.5
	217	140.5	705.1	5.0	470.9	3.3
	223	149.5	674.5	4.5	571.4	3.8
	241	134.5	521.2	3.9	444.6	3.3
	254	164.0	796.6	4.9	515.6	3.1
	255	149.3	688.5	4.6	466.1	3.1
	262	174.5	488.2	2.8	720.1	4.1
	279	156.5	670.3	4.3	493.9	3.2
	285	135.4	624.4	4.6	447.6	3.3
Mean ± SE		151.7±5.5	640.2±30.3	4.2±0.2	520.5±28.6	3.4±0.1

Uterus		Ovaries				
	Epithelial Cell Height	Paired Weight	No. Follicles			
RNA/DNA			4-9	10-15	16-20	>20
	(μ)		(mm)			
0.85	26.4	16.4	1	1		
1.06	26.4	15.0	1		1	
0.76	26.4	12.7	1	1		
0.72	24.5	13.4	1	6	1	
0.76	26.4	20.2	1	1		
0.51	26.4	12.2	2	1	1	
0.80	30.2	12.9	3			
0.54	24.5	13.7		2		
1.01	28.3	15.0			1	
0.56	--	18.0	5	1	1	
0.76±0.06	26.6±0.6	15.1±0.8	1.5	1.3	0.5	0
0.90	28.3	14.4		1	1	
0.84	26.4	12.2	2	3		
0.67	22.6	13.5	4		1	
0.85	20.7	16.6	3	2		
0.85	34.0	14.0	3	2		
0.65	24.5	14.0	2	1		
0.68	28.3	10.8	3			1
1.47	28.3	9.4	2		1	
0.74	34.0	11.2	7	1		
0.72	24.5	12.6	7	1	1	
0.84±0.08	27.2±1.4	12.9±0.7	3.3	1.1	0.4	0.1

Group No.	Heifer No.	Uterus				
		Weight	DNA		RNA	
		(g)	(mg)	(mg/g)	(mg)	(mg/g)
7[a]	202	104.2	436.5	4.2	468.8	4.5
	219	199.6	522.0	2.6	868.5	4.3
	230	137.4	665.6	4.8	599.5	4.4
	237	98.9	504.6	5.1	469.7	4.7
	240	193.7	650.2	3.4	1072.4	5.5
	253	166.2	589.6	3.5	817.6	4.9
	256	205.4	632.3	3.1	1001.7	4.9
	260	174.0	486.8	2.8	461.0	2.6
	265	117.3	414.9	3.5	481.0	4.1
	282	156.8	572.4	3.6	482.2	3.1
Mean ± SE		155.4±12.5	547.5±27.9	3.7±0.3	672.2±77.0	4.3±0.3
8[a]	201	179.5	834.8	4.6	896.0	5.0
	233	167.2	629.8	3.8	772.9	4.6
	234	134.3	589.4	4.4	515.0	3.8
	243	171.0	720.3	4.1	941.4	5.5
	246	243.4	800.2	3.3	998.1	4.1
	271	162.4	527.8	3.2	653.4	4.0
	276	137.5	612.0	4.4	359.8	2.6
	277	142.0	660.4	4.6	394.5	2.8
	289	158.2	413.8	2.6	439.5	2.8
	290	162.0	649.1	4.0	702.7	4.3
Mean ± SE		165.8±9.8	643.8±39.1	3.9±0.2	667.3±74.0	4.0±0.3

Uterus		Ovaries				
	Epithelial Cell Height	Paired Weight	No. Follicles			
RNA/DNA			4-9	10-15	16-20	>20
	(μ)		(mm)			
1.07	28.3	12.5	3			1
1.66	37.7	14.3				1
0.90	28.3	14.8	2		1	1
0.93	24.5	13.6	3	1	1	
1.65	39.6	29.1			3	
1.39	37.7	13.5	2	1	1	1
1.58	37.7	15.8			1	1
0.95	32.1	8.4	1		1	
1.16	18.9	20.0	1		1	2
0.84	24.5	11.6	2	1	1	
1.21±0.10	30.9±2.2	15.4±1.8	1.4	0.3	1.0	0.7
1.07	28.3	14.6	3		1	1
1.23	--	21.1	4		1	1
1.38	26.4	19.5	5		1	
1.31	30.2	18.7	2	1	1	1
1.25	30.2	20.9	3	1		1
1.24	49.1	12.1	4		1	1
0.59	20.8	18.2	1	1		2
0.60	28.3	21.3	10			1
1.06	30.2	10.0	1			1
1.08	39.6	19.9	4		1	1
1.08±0.09	31.5±2.7	17.6±1.3	3.7	0.3	0.6	1.0

Group No.	Heifer No.	Uterus				
		Weight (g)	DNA (mg)	DNA (mg/g)	RNA (mg)	RNA (mg/g)
9[a]	294	95.5	353.6	3.7	299.1	3.1
	305	144.3	300.4	2.1	350.5	2.4
	312	111.2	547.5	4.9	287.6	2.6
	318	104.3	432.1	4.1	304.6	2.9
	322	107.0	400.7	3.7	402.3	3.8
	344	157.3	455.4	2.9	635.0	4.0
	357	120.8	427.1	3.5	430.5	3.6
	364	153.7	518.7	3.4	685.0	4.5
	368	149.4	520.8	3.5	678.3	4.5
	369	149.5	638.5	4.3	458.2	3.1
	Mean ± SE	129.3±7.5	459.5±31.4	3.6±0.2	450.4±48.6	3.4±0.2
10[b]	292	142.9	558.3	3.9	639.0	4.5
	293	79.3	370.7	4.7	420.6	5.3
	300	124.8	548.0	4.4	303.2	2.4
	311	138.0	694.5	5.0	794.8	5.8
	320	161.3	852.5	5.3	542.6	3.4
	354	152.5	644.2	4.2	353.3	2.3
	355	110.0	607.2	5.5	238.2	2.2
	363	153.3	775.6	5.1	309.3	2.0
	366	120.3	615.3	5.1	272.5	2.3
	373	86.1	330.8	3.8	318.5	3.7
	Mean ± SE	126.9±8.9	599.7±51.1	4.7±0.2	419.2±57.6	3.4±0.4

Uterus		Ovaries				
RNA/DNA	Epithelial Cell Height (μ)	Paired Weight	No. Follicles (mm) 4-9	10-15	16-20	>20
0.85	32.1	13.4	2	1		1
1.17	30.2	14.1		1	1	1
0.53	34.0	18.9	1		1	
0.70	32.1	18.7	5	1		1
1.00	28.3	17.7		2		1
1.39	22.0	12.4	3		1	
1.01	29.1	17.2	3		1	1
1.32	22.6	10.3	1		1	
1.30	25.4	16.9	1			1
0.72	30.2	15.0	6	2		1
1.00±0.09	28.6±1.3	15.5±0.9	2.2	0.7	0.5	0.7
1.14	28.3	9.6	3	1		
1.13	20.8	3.3	2			
0.55	24.5	9.0	6	1		
1.14	24.5	13.9	4	1		
0.64	17.0	9.8	5	1		
0.55	28.3	8.4	5		1	
0.39	28.3	9.9	1	1		
0.40	30.2	10.5	4	1		
0.44	32.1	13.8	5	1		
0.96	20.8	11.4	4			
0.73±0.10	25.5±1.5	10.0±0.9	3.9	0.7	0.1	0

Group No.	Heifer No.	Uterus				
		Weight	DNA		RNA	
		(g)	(mg)	(mg/g)	(mg)	(mg/g)
11[b]	298	114.0	480.7	4.2	418.5	3.7
	301	94.3	562.8	6.0	406.4	4.3
	308	114.6	482.3	4.2	379.4	3.3
	309	107.4	454.1	4.2	514.0	4.8
	315	200.8	764.9	3.8	839.3	4.2
	347	152.6	675.6	4.4	516.7	3.4
	350	93.4	511.4	5.5	285.2	3.0
	352	119.2	632.9	5.3	387.1	3.2
	358	121.4	628.3	5.2	458.5	3.8
	371	98.8	439.7	4.4	245.1	2.5
Mean ± SE		121.7±10.3	563.3±34.2	4.7±0.2	445.0±51.7	3.6±0.2
12[b]	291	35.0	133.3	3.8	140.7	4.0
	295	104.5	484.6	4.6	483.8	4.6
	297	118.8	521.6	4.4	458.6	3.9
	302	114.6	422.0	3.7	443.8	3.9
	307	102.6	293.4	2.9	418.1	4.1
	353	135.8	384.7	2.8	497.2	3.7
	359	150.5	460.5	3.1	485.1	3.2
	360	119.2	520.5	4.4	457.1	3.8
	362	104.5	267.1	2.6	262.5	2.5
	365	182.8	419.3	2.3	612.7	3.3
Mean ± SE		116.8±12.0	390.7±39.5	3.5±0.3	426.0±41.8	3.7±0.2

Uterus		Ovaries				
RNA/DNA	Epithelial Cell Height (μ)	Paired Weight	No. Follicles 4-9 (mm)	10-15	16-20	>20
0.87	26.4	5.5		2		
0.72	28.3	10.1	5	1		
0.79	20.7	16.3	2	1		
1.13	22.6	12.1	3	1		
1.10	37.7	20.3		1		
0.76	22.6	12.7	1	1		
0.56	24.5	12.5	5	1		
0.60	26.4	10.7	3	1		
0.73	22.6	12.5		1	1	
0.56	30.2	10.5	4			
0.78±0.06	26.2±1.6	12.3±1.2	2.3	1.0	0.1	0
1.06	18.9	6.7	4	1		
1.00	18.9	5.6	1			
0.88	35.8	8.7	5	1	1	
1.05	30.2	14.1	1		1	1
1.42	24.5	11.1	1		1	
1.29	26.4	8.5	11			
0.99	--	14.4	4	1	1	
0.88	24.5	13.4	6	1		
0.98	28.3	6.6	1		1	
1.46	20.7	13.1	5			2
1.10±0.07	25.4±1.9	10.2±1.1	3.9	0.4	0.5	0.3

Group No.	Heifer No.	Uterus				
		Weight	DNA		RNA	
		(g)	(mg)	(mg/g)	(mg)	(mg/g)
13[b]	299	148.2	559.8	3.8	366.2	2.5
	303	188.0	351.9	1.9	518.1	2.7
	317	92.2	595.6	6.5	439.2	4.8
	319	86.5	342.7	4.0	431.8	5.0
	321	93.3	364.5	3.9	574.4	6.2
	349	64.3	288.5	4.5	216.3	3.4
	351	139.2	685.5	4.9	311.2	2.2
	356	180.3	558.8	3.1	398.6	2.2
	361	105.4	658.2	6.2	510.5	4.8
	367	136.0	566.2	4.2	433.7	3.2
Mean ± SE		123.3±13.1	497.2±45.9	4.3±0.4	420.0±33.2	3.7±0.4
14[c]	296	25.4	136.4	5.4	80.2	3.1
	310	28.9	130.4	4.5	118.9	4.1
	313	29.9	156.3	5.2	119.5	4.0
	314	24.7	134.9	5.5	101.0	4.1
	316	31.1	192.1	6.2	140.8	4.5
	342	42.2	224.2	5.3	184.1	4.4
	343	27.9	131.5	4.7	108.7	3.9
	345	27.0	144.6	5.4	117.5	4.3
	346	26.4	123.1	4.7	96.0	3.6
	348	32.0	165.0	5.2	132.9	4.1
Mean ± SE		29.6±1.5	153.9±10.1	5.2±0.1	119.9±9.0	4.0±0.1

[a]Slaughtered at breeding size.

[b]Slaughtered at first estrus.

[c]Slaughtered at 2.5 months of age.

Uterus		Ovaries				
RNA/DNA	Epithelial Cell Height	Paired Weight	No. Follicles 4-9	10-15	16-20	>20
	(μ)		(mm)			
0.65	26.4	19.1			1	1
1.47	37.7	10.7			1	
0.74	32.1	9.7	1	1		
1.26	22.6	11.7		2	1	
1.58	30.2	7.5	4		1	
0.75	20.8	8.4	3	2		
0.45	32.1	11.9			1	
0.71	24.5	9.4	2		1	
0.78	28.3	7.1	3	2		
0.77	18.9	10.0	5			
0.92±0.12	27.4±1.8	10.6±1.1	1.8	0.7	0.6	0.1
0.59	18.9	3.7	2	1		
0.91	15.1	3.8	1			
0.76	20.7	9.6	2			
0.75	11.3	8.4	1			
0.73	15.1	3.2	1	2		
0.82	18.9	16.6	1			
0.83	15.1	2.5	2	1		
0.81	15.1	10.1	5	1		
0.78	18.9	6.2	2			
0.81	--	3.4	3	1		
0.78±0.03	16.6±1.0	6.8±1.4	2.0	0.6	0	0

APPENDIX III.--Weight and nucleic acids of the mammary gland, and weight and cortex zone widths of the adrenals for individual heifers.

Group No.	Heifer No.	Mammary Gland[a]					
		Weight	DNA		RNA		RNA/DNA
		(g)	(mg)	(mg/g)	(mg)	(mg/g)	
5[b]	221	493	1454.0	2.9	820.3	1.7	0.56
	226	547	1525.1	2.8	1566.7	2.9	1.03
	229	936	1751.9	1.9	1224.7	1.3	0.70
	238	806	1323.0	1.6	1148.7	1.4	0.87
	242	837	1438.8	1.7	1754.8	2.1	1.22
	251	861	2723.4	3.2	1401.5	1.6	0.51
	263	649	2065.0	3.2	3063.5	4.7	1.48
	264	593	1898.7	3.2	1590.5	2.7	0.84
	266	562	898.7	1.6	933.2	1.7	1.04
	278	788	1858.3	2.4	948.4	1.2	0.51
Mean ± SE		707±49	1693.7±156.0	2.4±0.2	1445.2±205.2	2.1±0.3	0.88±0.10
6[b]	204	474	1023.6	2.2	1045.1	2.2	1.02
	214	984	2304.2	2.3	1965.0	2.0	0.85
	217	720	2091.3	2.9	2832.5	3.9	1.35
	223	307	573.9	1.9	670.0	2.2	1.17
	241	550	1230.5	2.2	925.5	1.7	0.75
	254	885	2970.9	3.4	1741.5	2.0	0.59
	255	454	686.5	1.5	629.1	1.4	0.92
	262	561	944.6	1.7	902.6	1.6	0.96
	279	590	1980.6	3.4	1544.0	2.6	0.78
	285	464	1201.6	2.6	635.8	1.4	0.53
Mean ± SE		599±66	1500.7±249.6	2.4±0.2	1289.1±228.4	2.1±0.2	0.89±0.08

Group No.	Adrenal Glands				
	Paired Weight	Cortex Width	Zone Widths		
			Glomerulosa	Fasciculata	Reticularis
	(g)	(mm)			
5[b]	11.8	1.89	0.22	1.07	0.60
	15.0	2.02	0.30	1.26	0.46
	11.6	2.51	0.29	1.41	0.81
	13.7	1.48	0.41	1.45	0.62
	15.0	1.52	0.30	0.71	0.51
	15.6	2.67	0.28	1.69	0.70
	13.9	1.91	0.49	1.02	0.40
	13.0	1.93	0.31	1.22	0.40
	12.8	2.07	0.28	1.27	0.52
	14.9	2.98	0.55	1.50	0.93
	13.7±0.4	2.09±0.15	0.34±0.03	1.26±0.09	0.60±0.05
6[b]	17.3	2.24	0.31	1.42	0.51
	16.5	2.91	0.35	2.00	0.56
	14.5	2.86	0.29	2.03	0.54
	22.5	1.82	0.36	0.90	0.56
	15.4	2.45	0.37	1.62	0.46
	13.4	1.64	0.33	0.83	0.48
	11.9	2.05	0.37	1.19	0.49
	13.6	1.77	0.27	1.00	0.50
	16.8	2.49	0.37	1.50	0.62
	17.4	1.81	0.31	1.22	0.28
	15.9±0.9	2.20±0.15	0.33±0.01	1.37±0.13	0.50±0.03

Group No.	Heifer No.	Mammary Gland[a]					
		Weight	DNA		RNA		RNA/DNA
		(g)	(mg)	(mg/g)	(mg)	(mg/g)	
7[b]	202	651	1615.1	2.5	2063.6	3.2	1.28
	219	762	1902.6	2.5	2067.3	2.7	1.09
	230	918	2947.9	3.2	2462.3	2.7	0.84
	237	619	1545.9	2.5	1539.1	2.5	1.00
	240	950	3488.2	3.7	3443.8	3.6	0.99
	253	892	3564.5	4.0	3201.2	3.6	0.90
	256	674	2170.7	3.2	1960.5	2.9	0.90
	260	798	3215.1	4.0	2097.9	2.6	0.65
	265	815	2562.6	3.1	2022.2	2.5	0.79
	282	805	2631.4	3.3	2567.0	3.2	0.98
Mean ± SE		788±36	2564.4±234.8	3.2±0.2	2342.5±186.2	2.9±0.1	0.94±0.05
8[b]	201	718	2034.0	2.8	1887.6	2.6	0.93
	233	1048	3410.9	3.2	4400.0	4.2	1.29
	234	607	2650.3	4.4	2252.5	3.7	0.85
	243	449	1299.9	2.9	1356.2	3.0	1.04
	246	639	2273.5	3.6	2224.1	3.5	0.98
	271	463	2123.9	4.6	2083.0	4.5	0.98
	276	540	2057.0	3.8	1306.5	2.4	0.64
	277	1837	5268.7	2.9	4368.8	2.4	0.83
	289	616	1378.1	2.2	1278.8	2.1	0.93
	290	445	1192.4	2.7	1420.1	3.2	1.19
Mean ± SE		736±134	2368.9±385.3	3.3±0.2	2257.8±374.0	3.2±0.2	0.97±0.06

Group No.	Adrenal Glands				
	Paired Weight (g)	Cortex Width	Zone Widths		
			Glomerulosa	Fasciculata	Reticularis
		(mm)			
7[b]	13.2	1.10	0.28	0.61	0.21
	15.2	1.72	0.36	1.00	0.36
	15.0	1.79	0.27	1.17	0.35
	14.8	1.98	0.32	1.11	0.55
	18.9	1.81	0.26	1.22	0.33
	13.0	1.76	0.32	0.93	0.51
	16.9	2.12	0.31	1.21	0.60
	12.6	1.66	0.28	0.78	0.60
	18.0	2.16	0.27	1.46	0.43
	17.7	1.64	0.27	0.99	0.38
	15.5±0.7	1.77±0.09	0.29±0.01	1.05±0.08	0.43±0.04
8[b]	12.8	1.87	0.31	1.12	0.44
	14.9	1.94	0.40	0.58	0.96
	12.4	1.74	0.29	1.11	0.34
	14.5	2.15	0.31	1.43	0.41
	13.6	2.01	0.33	1.27	0.41
	18.0	2.38	0.50	1.28	0.60
	16.0	1.72	0.33	0.88	0.51
	16.3	2.30	0.38	1.32	0.60
	12.7	1.88	0.31	1.11	0.46
	11.3	2.11	0.31	1.09	0.71
	14.3±0.7	2.01±0.07	0.35±0.02	1.12±0.08	0.54±0.06

Group No.	Heifer No.	Mammary Gland[a]					
		Weight	DNA		RNA		RNA/DNA
		(g)	(mg)	(mg/g)	(mg)	(mg/g)	
9[b]	294	400	1330.4	3.3	942.8	2.4	0.71
	305	739	3103.8	4.2	1989.4	2.7	0.64
	312	450	1339.6	3.0	690.7	1.5	0.52
	318	445	1450.3	3.3	976.3	2.2	0.67
	322	651	2529.1	3.9	1966.0	3.0	0.78
	344	794	3592.1	4.5	3895.4	4.9	1.08
	357	751	3025.8	4.0	3317.9	4.4	1.10
	364	276	962.1	3.5	967.1	3.5	1.01
	368	888	3241.2	3.6	3804.2	4.3	1.17
	369	385	1342.1	3.5	1326.7	3.4	0.99
Mean ± SE		578±66	2191.4±315.4	3.7±0.1	1987.7±394.2	3.2±0.3	0.87±0.07
10[c]	292	306	590.7	1.9	632.1	2.1	1.07
	293	296	669.9	2.3	442.4	1.5	0.66
	300	421	866.7	2.1	623.2	1.5	0.72
	311	574	1375.5	2.4	1299.0	2.3	0.94
	320	423	670.5	1.6	664.6	1.6	0.99
	354	368	906.7	2.5	379.1	1.0	0.42
	355	214	911.5	4.3	366.0	1.7	0.40
	363	262	828.7	3.2	322.1	1.2	0.39
	366	257	741.9	2.9	264.8	1.0	0.36
	373	159	398.3	2.5	357.1	2.2	0.90
Mean ± SE		328±38	796.0±81.8	2.5±0.2	534.0±96.0	1.6±0.1	0.69±0.09

Group No.	Adrenal Glands				
	Paired Weight (g)	Cortex Width	Zone Widths		
			Glomerulosa	Fasciculata	Reticularis
		(mm)			
9[b]	12.6	1.77	0.33	0.89	0.55
	14.1	1.97	0.35	1.16	0.46
	19.4	1.88	0.34	1.17	0.37
	13.1	1.40	0.32	0.79	0.29
	16.8	1.49	0.23	0.76	0.50
	11.9	1.71	0.37	0.94	0.40
	13.3	2.16	0.48	1.17	0.51
	12.7	1.54	0.31	0.81	0.42
	16.5	2.01	0.37	1.06	0.58
	12.3	1.18	0.37	0.52	0.29
	14.3±0.8	1.71±0.10	0.35±0.02	0.93±0.07	0.44±0.03
10[c]	10.3	1.41	0.29	0.86	0.26
	11.1	1.72	0.24	1.09	0.39
	12.2	1.79	0.25	1.08	0.46
	12.9	2.06	0.29	1.37	0.40
	10.7	1.63	0.36	0.94	0.33
	13.2	1.85	0.40	0.94	0.51
	14.0	1.89	0.35	1.15	0.39
	11.0	1.56	0.23	0.91	0.42
	10.2	1.87	0.39	1.05	0.43
	9.6	1.40	0.33	0.81	0.26
	11.5±0.5	1.72±0.07	0.31±0.02	1.02±0.05	0.39±0.02

Group No.	Heifer No.	Mammary Gland[a]					
		Weight	DNA		RNA		RNA/DNA
		(g)	(mg)	(mg/g)	(mg)	(mg/g)	
11[c]	298	308	417.5	1.9	417.0	1.9	1.00
	301	215	662.2	1.9	386.0	1.1	0.58
	308	343	518.6	1.7	776.9	2.5	1.50
	309	308	784.6	2.5	621.4	2.0	0.79
	315	232	508.1	2.2	756.1	3.3	1.49
	347	201	317.2	1.6	178.1	0.9	0.56
	350	233	805.1	3.4	479.3	2.1	0.60
	352	184	447.3	2.4	318.2	1.7	0.71
	358	238	447.1	1.9	345.7	1.4	0.77
	371	216	602.2	2.8	454.7	2.1	0.75
Mean ± SE		248±17	551.0±50.7	2.2±0.2	473.3±60.8	1.9±0.2	0.88±0.11
12[c]	291	167	262.3	1.6	154.8	0.9	0.59
	295	309	810.7	2.6	825.1	2.7	1.02
	297	238	739.4	3.1	969.8	4.1	1.31
	302	804	2512.9	3.1	2527.8	3.1	1.01
	307	231	465.3	2.0	586.6	2.5	1.26
	353	185	646.7	3.5	492.1	2.7	0.76
	359	298	1066.2	3.6	831.0	2.8	0.78
	360	156	426.7	2.7	342.3	2.2	0.80
	362	114	274.3	2.4	216.7	1.9	0.79
	365	293	963.3	3.3	829.7	2.8	0.86
Mean ± SE		279±62	816.8±207.4	2.8±0.2	777.6±213.9	2.6±0.2	0.92±0.07

Group No.	Adrenal Glands				
	Paired Weight (g)	Cortex Width	Zone Widths		
			Glomerulosa	Fasciculata	Reticularis
		(mm)			
11[c]	13.0	1.90	0.29	1.19	0.42
	10.7	1.61	0.32	0.88	0.41
	12.2	1.81	0.34	1.02	0.45
	10.4	2.06	0.27	0.98	0.81
	9.6	1.47	0.26	0.67	0.54
	10.7	1.34	0.23	0.77	0.34
	13.3	1.38	0.34	0.68	0.36
	10.5	1.52	0.29	0.75	0.48
	15.3	1.15	0.22	0.62	0.31
	10.4	1.27	0.25	0.71	0.31
	11.6±0.6	1.55±0.09	0.28±0.01	0.83±0.06	0.44±0.05
12[c]	7.9	1.45	0.26	0.71	0.48
	10.1	1.65	0.33	0.98	0.34
	11.2	1.68	0.34	0.86	0.48
	12.2	2.38	0.40	1.32	0.66
	9.8	1.90	0.40	1.14	0.36
	9.7	2.09	0.29	0.82	0.28
	13.3	1.21	0.31	0.59	0.31
	10.1	1.46	0.24	0.76	0.46
	8.1	1.51	0.27	0.86	0.38
	11.3	1.47	0.28	0.85	0.34
	10.4±0.5	1.68±0.11	0.31±0.02	0.89±0.07	0.41±0.03

Group No.	Heifer No.	Mammary Gland[a]					
		Weight	DNA		RNA		RNA/DNA
		(g)	(mg)	(mg/g)	(mg)	(mg/g)	
13[c]	299	286	695.3	2.4	314.3	1.1	0.45
	303	343	811.3	2.4	424.6	1.2	0.52
	317	120	259.1	2.2	255.1	2.1	0.98
	319	129	302.1	2.3	255.2	2.0	0.84
	321	269	574.6	2.1	521.5	1.9	0.91
	349	151	430.4	2.8	339.1	2.2	0.79
	351	403	1286.5	3.2	577.8	1.4	0.45
	356	185	525.6	2.8	323.4	1.7	0.62
	361	191	371.4	1.9	279.1	1.5	0.75
	367	213	757.8	3.6	400.9	1.9	0.53
Mean ± SE		229±30	601.4±96.7	2.6±0.2	369.1±35.1	1.7±0.1	0.68±0.06
14[d]	296	7	7.7	1.1	13.6	1.9	1.76
	310	44	93.6	2.1	161.1	3.7	1.72
	313	14	25.9	1.8	45.5	3.2	1.76
	314	15	37.0	2.5	53.5	3.6	1.45
	316	33	81.0	2.4	99.7	3.0	1.23
	342	66	270.4	4.1	119.1	1.8	0.44
	343	16	41.4	2.6	36.4	2.3	0.88
	345	27	14.8	0.5	25.3	0.9	1.71
	346	25	37.9	1.5	59.2	2.4	1.56
	348	21	27.0	1.3	40.9	1.9	1.51
Mean ± SE		27±5	63.7±24.5	2.0±0.3	65.4±14.7	2.5±0.3	1.40±0.14

[a]Measurements were made on the left half of the mammary gland.

[b]Slaughtered at breeding size.

[c]Slaughtered at first estrus.

Group No.	Adrenal Glands				
	Paired Weight	Cortex Width	Zone Widths		
			Glomerulosa	Fasciculata	Reticularis
	(g)	(mm)			
13[c]	11.0	1.56	0.27	0.94	0.35
	12.6	1.99	0.26	1.28	0.45
	9.1	2.00	0.38	1.06	0.56
	10.5	1.58	0.37	0.80	0.41
	15.2	1.82	0.37	1.01	0.44
	10.0	1.18	0.29	0.62	0.27
	12.9	1.65	0.34	0.96	0.35
	13.4	1.63	0.26	0.93	0.44
	13.8	1.72	0.33	0.86	0.53
	9.1	1.63	0.34	1.02	0.27
	11.8±0.7	1.68±0.07	0.32±0.01	0.95±0.05	0.41±0.03
14[d]	6.7	1.33	0.36	0.55	0.42
	5.1	1.21	0.25	0.56	0.40
	6.4	1.37	0.29	0.82	0.26
	5.3	1.38	0.39	0.61	0.38
	5.5	1.23	0.22	0.63	0.38
	6.6	1.43	0.34	0.55	0.54
	5.8	1.37	0.34	0.57	0.46
	7.2	1.64	0.48	0.71	0.45
	7.4	1.57	0.32	0.83	0.42
	4.7	1.29	0.30	0.65	0.34
	6.1±0.3	1.38±0.04	0.33±0.02	0.65±0.03	0.41±0.02

APPENDIX IV.--Pituitary weight and anterior pituitary levels of LH, FSH, and prolactin; plasma levels of LH and prolactin, and plasma to pituitary content ratios for LH and prolactin for individual heifers.

Group No.	Heifer No.	Total Pituitary Weight	Anterior Pituitary Weight		Posterior Pituitary Weight
			At Slaughter	After Storage	
		(g)			
5[a]	221	1.85	1.48	1.43	0.37
	226	1.61	1.28	1.23	0.29
	229	1.43	1.19	1.14	0.20
	238	1.43	1.10	1.06	0.28
	242	1.73	1.42	1.37	0.30
	251	2.50	1.90	1.78	0.52
	263	1.69	1.32	1.22	0.38
	264	1.50	1.16	1.10	0.30
	266	1.71	1.25	1.21	0.40
	278	1.72	1.37	1.34	0.32
Mean ± SE		1.72±0.10	1.35±0.07	1.29±0.06	0.34±0.03
6[a]	204	1.72	1.31	1.20	0.32
	214	1.98	1.52	1.47	0.42
	217	1.52	1.09	1.05	0.36
	223	2.23	1.56	1.40	0.51
	241	1.77	1.30	1.25	0.40
	254	1.66	1.17	1.13	0.44
	255	1.55	1.17	1.14	0.32
	262	1.77	1.06	1.03	0.58
	279	1.66	1.33	1.28	0.32
	285	1.56	1.12	1.07	0.37
Mean ± SE		1.74±0.07	1.26±0.05	1.20±0.04	0.40±0.03

Group No.	Pituitary LH		Pituitary FSH		Pituitary Prolactin	
	(µg/mg)	(µg)	(µg/mg)	(µg)	(µg/mg)	(µg)
5[a]	3.345	4800	0.33	474	0.247	354
	3.902	4807	0	0	0.315	388
	1.938	2215	1.26	1440	0.115	131
	1.668	1760	0.68	717	0.196	207
	--	--	--	--	--	--
	1.728	3084	0.78	1392	0.186	332
	1.511	1837	0.46	559	0.140	170
	1.462	1611	1.43	1576	0.077	85
	2.212	2665	0.48	578	0.123	148
	1.625	2171	1.67	2231	0.163	218
	2.154±0.291	2772±413	0.79±0.18	996±233	0.174±0.024	226±36
6[a]	0.702	846	0.89	1072	0.084	101
	1.280	1876	0.80	1173	0.103	151
	2.220	2338	1.51	1590	0.156	164
	2.654	3708	0.71	992	0.168	235
	2.449	3071	0.38	477	0.177	222
	1.171	1317	0.37	416	0.191	215
	--	--	--	--	--	--
	0.958	985	0.83	853	0.179	184
	2.010	2575	0.67	858	0.090	115
	1.636	1744	1.03	1098	0.220	235
	1.676±0.231	2051±320	0.80±0.11	948±119	0.152±0.016	180±17

Group No.	Plasma LH (ng/ml)	Plasma LH (μg)	Plasma Prolactin (ng/ml)	Plasma Prolactin (μg)	Plasma LH ÷ Pituitary LH (μg/mg)	Plasma Prolactin ÷ Pituitary Prolactin (μg/μg)
5[a]	2.9	37	96	1230	7.70	3.47
	2.2	28	47	602	5.82	1.55
	2.0	27	135	1805	12.18	13.78
	2.5	33	149	1992	18.75	9.62
	1.7	21	166	2033	--	--
	3.2	44	94	1293	14.26	3.89
	3.0	38	105	1327	20.68	7.81
	3.0	32	90	951	19.86	11.19
	15.2 omit	181 omit	153	1826	67.91 omit	12.34
	1.9	25	87	1127	11.51	5.17
	2.5±0.2	32±2	112±12	1419±151	13.85±1.97	7.65±1.45
6[a]	3.8	48	12	152	56.74	1.50
	3.8	54	173	2446	28.78	16.20
	2.1	26	60	731	11.12	4.46
	2.9	40	35	479	10.79	2.04
	2.7	36	117	1544	11.72	6.95
	3.8	50	76	992	37.97	4.61
	2.2	24	21	232	--	--
	2.5	29	128	1505	29.44	8.18
	2.3	31	86	1150	12.04	10.00
	2.3	26	99	1119	14.91	4.76
	2.8±0.2	36±3	81±16	1035±219	23.72±5.32	6.52±1.51

Group No.	Heifer No.	Total Pituitary Weight	Anterior Pituitary Weight		Posterior Pituitary Weight
			At Slaughter	After Storage	
		(g)			
7[a]	202	1.63	1.22	1.19	0.35
	219	1.45	1.10	1.06	0.31
	230	1.69	1.34	1.25	0.30
	237	1.79	1.41	1.33	0.34
	240	2.46	2.10	2.06	0.33
	253	1.82	1.39	1.29	0.45
	256	2.78	2.29	2.15	0.50
	260	1.63	1.37	1.31	0.20
	265	2.27	1.86	1.81	0.34
	282	1.66	1.28	1.12	0.34
Mean ± SE		1.92±0.14	1.54±0.13	1.46±0.12	0.35±0.02
8[a]	201	2.20	1.66	1.61	0.44
	233	2.56	1.97	1.76	0.41
	234	2.11	1.53	1.41	0.46
	243	1.74	1.46	1.42	0.22
	246	2.04	1.71	1.62	0.24
	271	1.60	1.18	1.15	0.39
	276	1.90	1.62	1.50	0.25
	277	2.00	1.58	1.48	0.32
	289	1.52	1.16	1.13	0.34
	290	1.62	1.20	1.14	0.33
Mean ± SE		1.93±0.10	1.51±0.08	1.42±0.06	0.34±0.02

Group No.	Pituitary LH		Pituitary FSH		Pituitary Prolactin	
	(μg/mg)	(μg)	(μg/mg)	(μg)	(μg/mg)	(μg)
7[a]	0.914	1087	0.49	583	0.078	93
	1.661	1757	0.43	455	0.225	238
	1.701	2131	1.04	1303	0.163	204
	2.720	3612	1.68	2231	0.134	178
	1.854	3829	0.82	1693	0.086	178
	0.718	928	0.58	750	0.109	141
	0.446	960	0.22	474	0.175	377
	4.876	6373	2.01	2627	0.103	135
	1.148	2076	0.41	741	0.206	372
	1.617	1809	0.05	56	0.100	112
	1.765±0.402	2456±539	0.77±0.20	1091±267	0.138±0.012	203±32
8[a]	--	--	--	--	--	--
	1.418	2503	1.60	2824	0.172	304
	1.285	1807	0.53	745	0.047	66
	1.051	1493	0.75	1066	0.068	97
	1.059	1716	0.50	810	0.079	128
	1.849	2125	0.30	345	0.163	187
	1.282	1923	0.39	585	0.125	187
	0.786	1164	0.07	104	0.136	201
	1.392	1569	0.61	687	0.112	126
	1.040	1191	0.49	561	0.067	77
	1.240±0.101	1721±144	0.58±0.14	859±262	0.108±0.015	152±25

Group No.	Plasma LH (ng/ml)	Plasma LH (μg)	Plasma Prolactin (ng/ml)	Plasma Prolactin (μg)	Plasma LH ÷ Pituitary LH (μg/mg)	Plasma Prolactin ÷ Pituitary Prolactin (μg/μg)
7[a]	3.0	42	57	806	38.64	8.67
	2.5	32	39	500	18.21	2.10
	1.8	27	94	1398	12.67	6.85
	2.1	29	155	2110	8.03	11.85
	1.8	30	164	2715	7.83	15.25
	3.5	48	138	1879	51.72	13.33
	70.1 omit	1030 omit	64	941	1072.92 omit	2.50
	3.9	50	35	451	7.85	3.34
	2.5	33	154	2021	15.90	5.43
	2.6	32	47	587	17.69	5.24
	2.6±0.2	36±3	95±17	1341±253	19.84±5.10	7.46±1.47
8[a]	2.9	41	47	658	--	--
	4.5	64	104	1489	25.57	4.90
	1.5	18	81	978	9.96	14.82
	3.0	41	38	513	27.46	5.29
	1.7	23	17	230	13.40	1.80
	3.0	41	132	1797	19.29	9.61
	4.3	61	119	1695	31.72	9.06
	2.8	44	98	1543	37.80	7.68
	3.9	43	68	757	27.41	6.01
	4.3	52	90	1087	43.66	14.12
	3.2±0.3	43±5	79±12	1075±170	26.25±3.63	8.14±1.43

Group No.	Heifer No.	Total Pituitary Weight	Anterior Pituitary Weight		Posterior Pituitary Weight
			At Slaughter	After Storage	
		(g)			
9[a]	294	1.85	1.30	1.25	0.45
	305	2.21	1.78	1.63	0.39
	312	1.73	1.33	1.27	0.36
	318	1.96	1.41	1.37	0.54
	322	1.87	1.48	1.45	0.31
	344	1.70	1.28	1.26	0.39
	357	1.55	1.22	1.20	0.28
	364	1.54	1.23	1.22	0.27
	368	2.15	1.78	1.77	0.33
	369	1.43	1.06	1.03	0.36
Mean ± SE		1.80±0.08	1.39±0.07	1.34±0.06	0.37±0.02
10[b]	292	1.76	1.35	1.26	0.25
	293	1.60	1.21	1.13	0.35
	300	1.58	1.25	1.13	0.33
	311	2.37	1.89	1.81	0.44
	320	1.82	1.43	1.32	0.37
	354	1.65	1.33	1.30	0.29
	355	1.36	1.09	1.05	0.27
	363	1.26	0.88	0.85	0.35
	366	1.64	1.24	1.10	0.28
	373	1.33	0.97	0.95	0.32
Mean ± SE		1.64±0.10	1.26±0.09	1.19±0.08	0.33±0.01

Group No.	Pituitary LH		Pituitary FSH		Pituitary Prolactin	
	(μg/mg)	(μg)	(μg/mg)	(μg)	(μg/mg)	(μg)
9[a]	1.201	1495	0.44	548	0.115	143
	1.458	2381	0	0	0.252	412
	1.491	1891	0.06	76	0.163	207
	0.547	749	0.34	466	0.073	100
	1.860	2699	0.13	189	0.226	328
	1.959	2459	0.33	414	0.171	215
	1.431	1717	0.55	660	0.182	218
	1.781	2175	0.44	537	0.140	171
	1.311	2317	0.63	1113	0.382	675
	1.410	1449	0	0	0.131	135
	1.445±0.126	1933±187	0.29±0.07	400±110	0.183±0.027	260±55
10[b]	0.785	989	0.76	958	0.104	131
	2.095	2376	0.81	919	0.031	35
	1.520	1725	0.95	1078	0.163	185
	0.704	1636	0.79	1836	0.101	235
	2.000	2630	0.93	1223	0.149	196
	2.454	3195	0	0	0.377	491
	2.040	2146	0.09	95	0.191	201
	1.762	1501	0.30	256	0.121	103
	2.011	2206	0.59	647	0.128	140
	1.687	1599	0.81	768	0.053	50
	1.706±0.176	2000±202	0.60±0.11	778±177	0.142±0.030	177±40

Group No.	Plasma LH (ng/ml)	Plasma LH (µg)	Plasma Prolactin (ng/ml)	Plasma Prolactin (µg)	Plasma LH ÷ Pituitary LH (µg/mg)	Plasma Prolactin ÷ Pituitary Prolactin (µg/µg)
9[a]	3.1	44	70	990	29.43	6.92
	5.4	72	65	869	30.24	2.11
	2.5	35	15	207	18.51	1.00
	3.2	42	42	557	56.07	5.57
	3.4	44	18	233	16.30	0.71
	2.8	34	93	1116	13.83	5.19
	3.4	41	81	987	23.88	4.53
	4.9	52	96	1015	23.91	5.94
	2.1	27	25	316	11.65	0.47
	2.2	23	118	1256	15.87	9.30
	3.3±0.3	41±4	62±11	755±124	23.97±4.09	4.17±0.94
10[b]	3.0	25	81	663	25.28	5.06
	1.4	12	28	233	5.05	6.66
	2.5	24	179	1704	13.91	9.21
	2.7	27	168	1682	16.50	7.16
	3.4	31	121	1105	11.79	5.64
	1.7	16	73	685	5.01	1.40
	2.1	20	91	876	9.32	4.36
	2.8	26	75	685	17.32	6.65
	2.1	19	76	670	8.61	4.79
	1.9	15	27	215	9.38	4.30
	2.4±0.2	21±2	92±16	852±163	12.22±1.97	5.52±0.66

Group No.	Heifer No.	Total Pituitary Weight	Anterior Pituitary Weight		Posterior Pituitary Weight
			At Slaughter	After Storage	
		(g)			
11[b]	298	1.13	0.83	0.80	0.28
	301	1.26	0.92	0.85	0.30
	308	1.35	1.05	0.99	0.28
	309	1.54	1.15	1.11	0.38
	315	1.36	0.98	0.95	0.33
	347	1.47	1.07	1.04	0.36
	350	1.52	1.14	1.03	0.33
	352	1.50	1.22	1.07	0.36
	358	1.65	1.09	1.05	0.38
	371	1.51	1.03	0.89	0.41
Mean ± SE		1.43±0.05	1.05±0.03	0.98±0.03	0.34±0.01
12[b]	291	1.09	0.77	0.74	0.31
	295	1.28	0.92	0.89	0.30
	297	1.48	1.13	1.09	0.28
	302	1.75	1.45	1.38	0.30
	307	1.58	1.15	1.09	0.38
	353	1.33	0.95	0.91	0.32
	359	1.77	1.42	1.36	0.33
	360	1.65	1.24	1.19	0.41
	362	1.30	0.98	0.95	0.29
	365	1.45	1.04	1.01	0.38
Mean ± SE		1.47±0.07	1.11±0.07	1.06±0.06	0.33±0.01

Group No.	Pituitary LH		Pituitary FSH		Pituitary Prolactin	
	(μg/mg)	(μg)	(μg/mg)	(μg)	(μg/mg)	(μg)
11[b]	1.024	823	0.24	193	0.083	67
	1.366	1164	0.32	273	0.084	72
	1.737	1720	0.72	713	0.085	84
	1.618	1791	0.13	144	0.184	204
	0.580	550	0.73	693	0.300	285
	1.734	1805	0	0	0.140	146
	2.124	2188	0.61	628	0.179	184
	2.354	2507	0.38	405	0.043	46
	0.937	1078	0.70	806	0.089	102
	1.120	1001	2.37	2119	0.149	131
	1.459±0.176	1463±200	0.62±0.21	597±190	0.123±0.024	132±23
12[b]	--	--	--	--	--	--
	0.945	837	1.74	1542	0.155	137
	1.485	1623	0.53	579	0.090	98
	1.058	1459	0.35	483	0.135	186
	2.321	2523	0.21	228	0.104	113
	3.094	2819	0	0	0.151	138
	1.539	2095	0.58	789	0.082	112
	1.554	1851	1.29	1536	0.160	191
	1.915	1812	0	0	0.146	138
	1.772	1795	0.69	699	0.217	220
	1.743±0.219	1868±193	0.60±0.19	651±192	0.138±0.014	148±14

Group No.	Plasma LH		Plasma Prolactin		Plasma LH ÷ Pituitary LH	Plasma Prolactin ÷ Pituitary Prolactin
	(ng/ml)	(μg)	(ng/ml)	(μg)	(μg/mg)	(μg/μg)
11[b]	3.9	26	7	46	31.59	0.69
	1.0	7	29	216	6.01	3.00
	4.0	36	85	770	20.93	9.17
	2.9	23	24	191	12.84	0.94
	4.5	35	18	140	63.64	0.49
	2.1	18	25	213	9.97	1.46
	2.0	19	31	289	8.68	1.57
	2.6	24	18	168	9.57	3.65
	1.5	13	34	297	12.06	2.91
	2.5	20	24	195	19.98	1.49
	2.7±0.4	22±3	29±7	252±62	19.53±5.46	2.54±0.81
12[b]	2.7	19	48	331	--	--
	2.0	16	42	334	19.12	2.44
	3.8	33	20	171	20.33	1.74
	5.3	51	91	882	34.96	4.74
	4.3	37	44	374	14.67	3.31
	4.9	39	23	181	13.83	1.31
	4.0	40	26	262	19.09	2.34
	3.4	29	12	101	15.67	0.53
	4.5	36	33	262	19.87	1.90
	2.7	26	27	255	14.48	1.16
	3.8±0.3	33±3	37±7	315±68	19.11±2.15	2.16±0.42

Group No.	Heifer No.	Total Pituitary Weight	Anterior Pituitary Weight		Posterior Pituitary Weight
			At Slaughter	After Storage	
		(g)			
13[b]	299	1.49	1.21	1.16	0.27
	303	2.08	1.63	1.52	0.43
	317	1.34	0.90	0.81	0.39
	319	1.72	1.28	1.20	0.41
	321	1.79	1.32	1.28	0.42
	349	1.57	1.20	1.16	0.33
	351	1.39	1.03	1.00	0.31
	356	1.53	1.18	1.15	0.30
	361	1.31	1.02	1.00	0.25
	367	1.13	0.80	0.78	0.30
Mean ± SE		1.54±0.09	1.16±0.07	1.11±0.06	0.34±0.02
14[c]	296	1.00	0.72	0.66	0.28
	310	0.72	0.57	0.52	0.09
	313	0.90	0.65	0.60	0.23
	314	0.90	0.68	0.65	0.16
	316	0.79	0.59	0.58	0.13
	342	1.51	1.24	1.14	0.22
	343	1.08	0.85	0.82	0.15
	345	1.14	0.89	0.84	0.21
	346	0.71	0.57	0.55	0.12
	348	1.10	0.85	0.74	0.22
Mean ± SE		0.99±0.08	0.76±0.06	0.71±0.05	0.18±0.02

[a]Slaughtered at breeding size.

[b]Slaughtered at first estrus.

[c]Slaughtered at 2.5 months of age.

Group No.	Pituitary LH		Pituitary FSH		Pituitary Prolactin	
	(μg/mg)	(μg)	(μg/mg)	(μg)	(μg/mg)	(μg)
13[b]	1.917	2214	0.54	624	0.113	131
	1.788	2721	0.63	959	0.107	163
	2.387	1926	2.43	1961	0.165	133
	0.532	641	0.35	421	0.192	231
	0.498	636	0.59	754	0.176	225
	1.664	1922	0.23	266	0.184	213
	2.527	2530	1.17	1171	0.223	223
	0.658	755	0.20	229	0.221	253
	2.155	2151	0.33	329	0.147	147
	0.547	427	1.19	928	0.125	97
	1.467±0.260	1592±278	0.77±0.22	764±167	0.165±0.013	182±17
14[c]	1.486	982	0.23	152	0.106	70
	1.550	803	0.90	466	0.292	151
	2.084	1209	0.61	354	0.091	53
	1.371	894	0.88	574	0.204	133
	2.364	1376	0.69	402	0.104	61
	0.915	1045	0.35	400	0.165	188
	1.495	1220	3.01	2456	0.056	46
	1.947	1641	1.70	1433	0.106	89
	0.661	366	1.34	741	0.096	53
	1.454	1076	0.84	622	0.099	73
	1.553±0.161	1061±109	1.05±0.26	760±218	0.132±0.022	92±15

Group No.	Plasma LH		Plasma Prolactin		Plasma LH ÷ Pituitary LH	Plasma Prolactin ÷ Pituitary Prolactin
	(ng/ml)	(μg)	(ng/ml)	(μg)	(μg/mg)	(μg/μg)
13[b]	3.9	37	77	728	16.71	5.56
	18.9 omit	227 omit	111	1333	83.43 omit	8.18
	4.3	40	21	193	20.77	1.45
	5.6	46	15	124	71.76	0.54
	1.3	11	191	1651	17.30	7.34
	2.1	18	26	221	9.37	1.04
	2.1	23	30	334	9.09	1.50
	48.9 omit	428 omit	29	254	566.89 omit	1.00
	4.7	39	26	217	18.13	1.48
	3.1	26	14	116	60.89	1.20
	3.4±0.5	30±4	54±18	517±173	28.00±8.55	2.93±0.92
14[c]	1.9	6	79	241	6.11	3.44
	3.6	11	45	140	13.70	0.93
	1.4	5	27	87	4.14	1.64
	1.8	6	47	168	6.71	1.26
	2.9	9	11	36	6.54	0.59
	1.7	7	87	381	6.70	2.03
	2.6	8	54	176	6.56	3.83
	4.5	17	56	210	10.36	2.36
	1.3	4	65	180	10.93	3.40
	1.6	5	113	340	4.65	4.66
	2.3±0.3	8±1	58±9	196±33	7.64±0.96	2.41±0.43

APPENDIX V.--Age at first breeding and conception, number of services, body size at parturition, dystocia rating, calf weight, sire, and sex, and first lactation milk yield for individual heifers.

Group No.	Heifer No.	Age At First Breeding (mo)	Age At Conception (mo)	No. Services	Weight Before Parturition (kg)	Weight After Parturition (kg)	Withers Height At Parturition (cm)
1[a]	206	15.2	15.2	1	575	530	131.0
	209	13.0	13.7	2	539	475	132.5
	212	12.6	12.6	1	520	472	130.0
	215	13.2	15.7	1	462	492	128.0
	232	13.5	16.3	5	545	457	128.0
	250	12.0	12.5	2	-	473	127.0
	273	15.0	15.0	1	478	419	131.0
	275	12.8	12.8	1	454	432	126.5
	284	12.3	18.3	7	455	391	124.0
Mean ± SE		13.3±0.4	14.7±0.7	2.3±0.7	504±17	460±14	128.7±0.9

[a]Heifer No. 286 slaughtered because infertile.

2[b]	203	10.5	16.8	6	577	499	129.0
	205	11.0	13.3	3	520	470	132.0
	207	12.2	13.0	2	534	475	127.0
	210	11.0	17.0	9	545	509	135.0
	213	12.0	13.6	3	-	-	--
	257	11.7	11.7	1	580	514	125.0
	268	11.8	12.5	2	545	511	130.0
	283	10.7	10.7	1	499	434	129.0
	288	11.0	11.7	2	520	391	131.5
Mean ± SE		11.3±0.2	13.4±0.7	3.2±0.9	540±10	475±15	129.8±1.1

[b]Heifer No. 213 stolen from Driggs Dairy, and heifer No. 270 slaughtered because infertile.

Group No.	Dystocia Rating	Calf Birth Weight (kg)	Sire Of Calf	Sex Of Calf	Estimated 305 Day Milk Yield (kg)	Actual First 60 Day Milk Yield (kg)
1[a]	4	36	Royal Pontiac	Male	4189	1066
	3	45	RP	Female	3433	806
	4	51	RP	M	4413	1036
	3	44	Wis Symbol	M	4610	1173
	4	32	RP	M	4138	1053
	1	25	WS	F	4060	953
	1	48	RP	F	3840	977
	1	27	WS	F	4870	1240
	1	39	WS	F	incomplete	incomplete
	2.4±0.5	39±3			4194±158	1038±47
2[b]	2	37	RP	M	3938	1002
	-	52	WS	M	3374	792
	1	39	WS	M	3745	879
	1	premature	WS	M	4281	1005
	-	-	RP	-	--	--
	1	33	RP	F	5159	1211
	1	36	WS	F	3887	989
	1	41	WS	M	4064	954
	4	45	RP	M	4256	999
	1.6±0.4	40±2			4088±184	979±42

Group No.	Heifer No.	Age at first Breeding (mo)	Age at Conception (mo)	No Services	Height Before Parturition (kg)	Height After Parturition (kg)	Withers Height at Parturition (cm)
3	208	10.7	15.2	6	598	509	133.5
	211	10.7	16.3	7	509	550	130.0
	220	14.0	14.6	2	473	443	124.0
	231	13.0	16.5	5	625	595	128.0
	236	12.8	15.0	3	488	446	126.5
	259	13.3	13.3	1	469	495	132.0
	261	12.0	12.0	1	493	411	126.0
	269	12.8	18.9	4	482	436	128.0
	272	12.0	12.4	2	516	479	125.5
	280	10.5	13.0	3	-	493	130.0
Mean ± SE		12.2±0.4	14.7±0.7	3.4±0.7	517±19	486±18	128.4±0.9
4[C]	225	10.7	14.0	6	614	560	131.5
	239	12.5	12.5	1	525	489	125.5
	249	12.0	19.0	6	500	464	132.0
	258	13.3	14.0	2	457	411	126.5
	274	13.0	14.8	2	460	416	128.0
	281	13.5	13.5	1	484	445	127.5
	287	13.0	14.7	3	495	436	129.0
Mean ± SE		12.6±0.4	14.6±0.8	3.0±0.8	505±20	460±20	128.6±0.9

[C]Heifers No. 218 and 235 slaughtered because infertile. Heifer No. 222 did not conceive until she was 22 months old, so she was excluded from the study.

Group No.	Dystocia Rating	Calf Birth Weight (kg)	Sire Of Calf	Sex Of Calf	Estimated 305 Day Milk Yield (kg)	Actual First 60 Day Milk Yield (kg)
3	3	45	RP	M	4185	1065
	1	26	WS	F	4197	1068
	1	37	WS	F	3804	893
	2	32	WS	M	4838	1231
	1	30	RP	M	2543	597
	2	34	RP	F	2248	572
	2	52	RP	M	3527	828
	1	41	WS	F	incomplete	incomplete
	1	34	RP	F	4637	1180
	1	34	WS	F	3733	950
	1.5±0.2	36±2			3746±292	931±78
4[c]	3	55	WS	F	4294	1008
	2	46	RP	M	4392	1031
	1	36	RP	F	incomplete	incomplete
	1	35	RP	F	4162	1059
	1	38	RP	F	3600	916
	1	-	WS	F	4645	1182
	4	56	RP	M	3242	825
	1.9±0.5	44±4			4056±216	1004±50