Manure and Nutrient Partitioning, Accretion, and Excretion in Holstein Heifers

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#### **ABSTRACT**

Considerable changes have occurred in environmental regulations in recent years, only one of which is the requirement of stand alone heifer operations and feedlots to carry environmental permits. While growth of heifers is a widely researched topic, publications concerning nutrient utilization, partitioning, and excretion are scarce and are becoming necessary. That combined with the fact that feeding programs for heifers are highly variable from region to region and even from farm to farm indicated the need to examine the effects of diet on nutrient utilization. Therefore, the objective of this work was to evaluate how differences in levels of dietary protein and energy will affect growth and nutrient utilization in heifers from birth to calving. Two projects were conducted, one in 20 month old bred heifers where forage level in the diet was altered to provide the required nutrients in less feed and one in young calves where energy and protein content of the milk replacer was altered. Three diets were fed to 18 (n=6), 20 month old heifers who were in late gestation; the first was the high forage (HF) ration which was 90.7% forage and 9.3% concentrate, the second was a by-product (BP) ration which was designed to have the same level of fiber as the HF ration, however soybean hulls and cottonseed hulls were added at the sacrifice of the grain mix which increased the fiber content; the last ration was the low forage (LF) which was 54.7% concentrate and 45.3% forage and was fed at ~89% of the ad libitum intake of heifers fed the HF ration. Heifers fed the HF ration had greater fecal excretion compared to those fed LF, however heifers fed the LF ration exceeded the heifers fed HF and BP by 4.5 and 2.5 times, respectively,

in urine volume excretion (40.2 vs. 8.9 and 16.9 kg/d, respectively). Although total N excretion (kg/d) was not different, heifers fed the LF ration tended to partition more N to

urine than to feces. Phosphorus excretion in the feces was not different, however heifers fed HF and BP tended to have greater fecal P (P < 0.06). Urinary P excretion was less in heifers fed HF and BP compared to LF, however these heifers were excreting as much urinary P as a lactating cow.

Calves were purchased from a commercial dairy at 3 d old ( $\pm$  1d) and transported to the VT Dairy Center. They remained on study until 63 d when they were harvested for body composition. Four treatment diets were fed; a control milk replacer (24/17; 24%) CP, 17% Fat), a high protein, low fat (32/17; 32% CP, 17% Fat), a high protein, high fat (31/24, 31% CP, 24% fat), and that same 31/24 milk replacer fed at 1460 g/d powder (31/24+). Calves were offered a 20% cottonseed hull starter at 1 d after arrival to the VT Dairy Center. Calves fed 24/17 consumed more starter than those fed the other milk replacers and therefore had a lower apparent digestibility and greater fecal excretion. Fecal N excretion was not different, although calves fed 24/17 tended to have greater fecal N excretion. Urinary N excretion was higher in calves fed 31/24+ compared to those fed 31/24. Total N excretion and N retention were not different. Empty body weight (EBW) gain was greater in calves fed 31/24+ compared to 31/24, however those same calves also had a higher percent of EBW as fat. Calves fed 32/17 had the most lean gain (in the form of N gain) compared to those fed extra energy (31/24) and also had a higher N as a percent of EBW.

Limit feeding Holstein heifers late in gestation did not reduce nutrient excretion, however, more digestible nutrients were available to the heifer and fetus. Heifers in late gestation are likely over fed P and therefore excrete nearly everything they consume which has negative implications for nutrient management planning. Calves fed a low

protein, low fat milk replacer did not grow as well as calves fed higher protein. Nitrogen retention and CP gain were higher when protein was at least 31% and fat was at least 17%. Feeding fat over 17% only increased fat gain and not CP gain. Overall, paying for extra nutrients in bred heifer diets seemed to be beneficial, however, feeding above 31% CP and 17% fat increased nutrient loss to the environment.

## **DEDICATION**

This dissertation is in dedicated to my parents, Tom, Peggy, and Kim Hill of Canton North Carolina.

My father didn't tell me how to live; he lived, and let me watch him do it.

-Clarence Budinton Kelland.

I have found the best way to give advice to your children is to find out what they want and then advise them to do it.

-Harry S Truman.

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# **TABLE OF CONTENTS**

ABSTRACT	II
DEDICATION	V
ACKNOWLEDGMENTS	VI
TABLE OF CONTENTS	VII
LIST OF TABLES	IX
CHAPTER 1 REVIEW OF LITERATURE	1
Environmental Regulations	2
FEEDING HEIFERS FROM BIRTH TO POST-WEANING	
FEEDING HEIFERS FROM BREEDING TO CALVING	
COSTS OF RAISING REPLACEMENT HEIFERS	
NITROGEN AND ENERGY METABOLISM IN PRE-WEANED CALVES	
SUMMARY	
References	
CHAPTER 2 NITROGEN AND PHOSPHORUS RETENTION, PARTITI	
AND EXCRETION IN LATE GESTATION DAIRY HEIFERS	
Interpretive Summary	
ABSTRACT	
Introduction	
MATERIALS AND METHODS	
Animals and Diets	
Sample Collection	
Lab Analysis	
Statistical Analysis	37
Comparison to ASAE Standards	37
RESULTS AND DISCUSSION	
Diet Composition, Intake, and Growth Measures	
Digestibility and Manure Excretion	
Nitrogen Intake, Excretion, and Partitioning	
Phosphorus Intake, Excretion and Partitioning	
Measure of Solids	
Comparison to ASAE prediction equations	
Conclusions	
REFERENCES	48
CHAPTER 3 MILK REPLACER COMPOSITION AND NUTRIENT UTILIZATION IN PRE-WEANED HOLSTEIN CALVES	59
Interpretive Summary	59
ABSTRACT	
Introduction	
MATERIALS AND METHODS	

Animals and diets	
Harvest Procedure	63
Sample collection and analysis	64
Calculation of Gain	64
Statistical Analysis	65
RESULTS AND DISCUSSION	65
Digestibility Trial	65
Body Composition	69
Chemical Composition	70
CONCLUSIONS AND IMPLICATIONS	73
References	76
CHAPTER 4 CONCLUSIONS	85

# LIST OF TABLES

TABLE 1-1. COMPOSITION OF WHOLE MILK, DRIED SKIM MILK, AND DRIED WHEY (% DM)	
TABLE 1-2 EMPTY BODY WEIGHT (EBW) GAIN AND COMPOSITION OF GAIN IN CALVES FE MILK REPLACERS VARYING IN PROTEIN AND ENERGY.	D
TABLE 1-3 NITROGEN INTAKE, DIGESTIBILITY, AND FECAL EXCRETION IN PRE-WEANED HEIFERS AND BULLS	
TABLE 1-4. NITROGEN DIGESTED, RETAINED AND URINARY EXCRETION IN PRE-WEANED HEIFERS AND BULLS	21
	23
Table 2-1. Ingredient composition of three treatment diets fed to Holstein Heifers.	51
TABLE 2-2 NUTRIENT COMPOSITION OF THREE TREATMENT DIETS FED TO HOLSTEIN HEIFERS	
TABLE 2-3. DRY MATTER INTAKE, DIGESTIBILITY AND MANURE PRODUCTION IN HOLSTEIN HEIFERS FED DIETS VARYING IN FORAGE CONTENT	
TABLE 2-4. BODY WEIGHT AND MEASURES IN GROWING HOLSTEIN HEIFERS FED DIETS VARYING IN FORAGE CONTENT	
*	
HEIFERS FED DIETS VARYING IN FORAGE CONTENT TABLE 2-7. MANURE CHARACTERISTICS FROM HOLSTEIN HEIFERS FED DIETS VARYING IN	
FORAGE CONTENT.  TABLE 2-8. COMPARISON OF OBSERVED AND PREDICTED VALUES OF MANURE AND	
NUTRIENT EXCRETION USING ASAE STANDARDS (2005)	58
PROTEIN AND FAT FED TO HOLSTEIN CALVES.  TABLE 3-2. NUTRIENT INTAKE AND DRY MATTER DIGESTIBILITY OF MILK REPLACERS	78
VARYING IN PROTEIN AND FAT FED TO HOLSTEIN CALVES	79
PROTEIN AND FATTABLE 3-4. MANURE EXCRETION AND NITROGEN METABOLISM IN HOLSTEIN CALVES FED	
MILK REPLACERS VARYING IN PROTEIN AND FAT	
VARYING IN PROTEIN AND FAT.  TABLE 3-6. COMPOSITION OF BODY COMPONENTS IN HOLSTEIN CALVES FED MILK	
REPLACERS VARYING IN PROTEIN AND FAT.	83

#### CHAPTER 1 REVIEW OF LITERATURE

## **Environmental Regulations**

The Clean Water Act was first enacted in 1948 with the goal of providing technical assistance and funding to state and local governments to improve and preserve water quality (Water Pollution Control Act, 1948). Frustrations over slow implementation of solutions lead to an overhaul of the law in 1972 (Clean Water Act, 1972). These amendments reshaped the statute and provided stricter regulations, more federal funding, and increased federal involvement in enforcing the law, however state governments can still restrict regulations as needed. Until 1987, this law focused primarily on point source pollution (waste from discrete sources such as municipal and industrial pipelines and outfalls). It is known that nearly 50% of the nation's water pollution is from non-point source pollution such as runoff from farm lands, construction sites, and urban areas (Meyer and Mullinax, 1999). Amendments in 1987 required states to develop and implement non-point pollution management programs and supplied grants for up to 60% of the costs (Copeland, 1999).

Several updates and amendments have been made to the Clean Water Act concerning non-point pollution sources, specifically large animal operations, since 1987. An animal feeding operation (AFO) is defined as "an operation that stables or confines and feeds or maintains animals for a total of 45 d or more in any 12-mo period and does not sustain crops, vegetation, forage growth, or post-harvest residues during the normal growing season over any portion of the lot or facility". Large AFO containing more than 1,000 animal units (AU) are defined as concentrated animal feeding operations (CAFO) mainly due to their size. Smaller units (301-1,000 AU) can be defined as CAFO if they

"directly discharge pollutants into water that originate outside of and pass over, across, or through the facility or otherwise come into direct contact with the confined animals or pollutants that are discharged through a man-made conveyance". Even smaller facilities (< 300 AU) can be considered CAFO if they are a "significant contributor of pollution to waters of the U. S." According to the Clean Water Act, CAFO are point source dischargers (EPA administered permit programs, 1974).

There is a trend across the U.S. and across all livestock species for greater numbers of animals per farm with fewer operations. This trend has led to an increased number of CAFO, especially in the dairy and beef cattle industry. These operations have been recognized as major contributors to low dissolved oxygen in surface waters due to nutrient contamination. The EPA has further propagated the Clean Water Act by publishing permit qualifications and regulations specific to different species of livestock. These publications detail the requirements necessary to obtain a permit and to keep it current (U.S. EPA 833-F-02-009).

### **Nutrient Management Planning on Dairy Farms**

With the changes in numbers and the definition (including small operations) of CAFO, nutrient management planning is quickly becoming a priority of livestock producers. A nutrient management plan (NMP) contains numerous components including aerial site photographs; soil and plant tissue test results; realistic yield goals, quantification of important nutrient sources; recommended rates, method and timing of applications; location of sensitive areas; and nutrient budgets for plant and animal production systems.

These plans are developed to meet the requirements for a resource management system nutrient application rate. When the operation is located within an impaired area, meaning that the soil or water is already loaded with nutrients, then the nutrient management plan should also include the potential risk of increasing the load of N (N) and P (P). Plans require adequate estimates of nutrient concentration of manures that are applied to crop acreage. Currently, many states are moving to base NMPs on P instead of N and this could have serious negative implications for many producers across all livestock species. Typically, when basing nutrient use across the whole farm on N needs of the soil or crop, P needs are exceeded. When basing nutrient use on P needs, N needs are not met and would have to be met with the purchase of commercial fertilizer which adds expense. Also, P based NMPs will make if difficult for areas with soils that are already saturated with P to handle manure because more P could not be added. The removal of P from manure before it leaves the farm or is land applied has thus recently become a heavily researched area.

Dou et al. (1996) developed a spreadsheet that quantifies the flow of N through a dairy farm. The program includes several factors including ration formulation, crop information, and manure application. This spreadsheet is based on diets formulated from production level requirements using the Cornell Net Carbohydrate and Protein System. The spreadsheet contains several different worksheets that focus on individual aspects of N utilization on the farm, including herd, manure, crops, and feed. These different worksheets allow the management to demonstrate overall N balance across the entire farm from N inputs (feed, fertilizer, bedding, cows, etc.) to outputs (milk, cows, exported manure, etc.). A system such as this could easily be used to also track P P and other

nutrients of interest on the farm and could be adapted by heifer growers when an accurate database of information concerning N and P accretion and excretion in heifers is established.

Meyer and Mullinax (1999) suggested that the dairy industry, scientists, and the EPA should work closely together to establish a factual database that can be used to write guidelines for CAFO. The National Dairy Heifer Evaluation Project (NDHEP) that was published by Heinrichs and others in 1994 is an excellent source of data concerning dairy producers throughout the U.S. This project included results from numerous surveys conducted across the nation to dairy producers, including heifer growers. Almost 2% of producers surveyed sold or removed all of their heifers within 24 hr of birth. Of that 2%, 10% contracted heifers out to growers. The average age of heifers contracted out was from birth to 4 months. Regardless of the age at which the heifer was contracted out, the majority remained in the contracted herd until close to calving. Since the NDHEP was completed, these numbers have likely increased. The need for increased body size at first calving, reduced age at first calving, and reduced mortality of heifers has made contract rearing of replacements an attractive option. These kinds of changes within the industry, along with the fact that stand-alone heifer operations can now be considered CAFO, call for reevaluation of manure management programs for both dairy producers and heifer growers.

Thus far, copious data has been published concerning N and P accretion, partitioning, and excretion in dairy cows at various stages of lactation (St-Pierre and Threan 1999; Knowlton et al. 2001; Kebreab et al. 2002; Haig et al. 2002; Dou et al. 2003; Davidson, et al. 2003). Less work has been done to produce quantitative and

qualitative manure excretion data in other groups of animals kept on dairy farms, specifically heifers and neonatal calves.

### Feeding Heifers from Birth to Post-Weaning

Practices for feeding young calves have undergone drastic changes in recent years. These new feeding techniques have been termed differently throughout the country and are commonly referred to as 'accelerated', 'intensified', 'rapid', or 'normal biological growth'. For the purpose of this paper, the terms 'accelerated' growth and 'intensified' milk replacers will be used as they exceed the current NRC (2001) recommendations for CP and fat content in young heifers. The desire for accelerated growth of heifers has lead to the formulation of advanced milk replacers and intensified starter rations. Approximately 10 million dairy calves are born in the U. S. each year (Davis and Drackely, 1998) and with the growth of the dairy heifer growers industry and the increasing animal density on farms, data are needed to determine the effects that these changes in calf diets might have on N N and P P excretion from the animal. New CAFO regulations will provide the demand for this sort of research, and the changes in protein and energy content of these milk replacers begs the question of changes in body composition and type of growth these calves are undergoing. It also leads to the need for data defining N and P accretion and partitioning in the young calf.

Table 1-1 shows the comparison of whole milk solids found in dried skim milk and dried whey, which are two common ingredients in milk replacers. The proportion of lactose to fat in milk replacer ingredients is much higher than that of whole milk. New formulas for milk replacers attempt to more closely match whole milk by increasing the fat content of milk replacer with the addition of feedstuffs like tallow, choice white

grease, coconut oil, and other oils. These ingredients have become default ingredients for milk replacers because they are highly digestible and less expensive than milk fat.

### Feeding Heifers from Breeding to Calving

Protein and energy (as well as other nutrients) requirements have long been established for lactating cows. Recent publications (NRC, 2001) have established more refined dietary guidelines and requirements for heifers, however these requirements are based on small data sets. Producers are often searching for the most economical way to meet these requirements, which often leads to the abundant use of forage and by-products in the rations. These types of feeds cause great variation in nutrient content in heifer rations. Also, dairy heifers are most often grouped by age or body weight and can be fed diets ranging from high energy and protein grain diets to complete pasture diets leading to a great difference in nutrient utilization. The variability of the diet that a heifer consumes contributes greatly to the variability of the nutrient content of the manure excreted.

### **Costs of Raising Replacement Heifers**

Tozer and Heinrichs (2001) reported that the cost of raising replacement heifers for a 100-cow dairy was \$32,344 per year. Reducing the culling rate by 20% and reducing the age at first calving by one month dropped the cost by \$7968 and \$1400, respectively. Of total expenses on a farm, raising replacements is second only to feed costs. Feed costs comprise more than 60% of the \$32, 344 estimated cost spent on replacement heifers and improving efficiency of nutrient metabolism and utilization will help producers achieve reduced age at first calving and increase their profit.

#### Nitrogen and Energy Metabolism in Pre-Weaned Calves

In 1976, Donnelly and Hutton completed a study where 46 Friesian bull calves were fed milk replacers and whole milk powder with 15.7, 18.1, 21.8, 25.4, 29.6, and 31.5% CP and 22.0, 24.2, 24.2, 23.1, 18.9, and 18.3% fat, respectively. These calves were fed for either 600 or 900 g/d of gain. The calves fed at the higher rate gained 36% more body weight than those fed at the lower rate. The authors concluded that at high protein intakes, the extra weight gain was due to the increased digestible energy intake, but at low CP intake, gain was due to energy and protein intake. Increasing dietary CP increased water and protein content (kg) and decreased fat. Increasing the level of feed offered increased fat content and decreased water content in body gain. These authors concluded that increasing dietary CP from 16 to 32% increased protein content by 2% and decreased fat content of the whole body by 4% indicating that there is a relationship between protein intake and energy utilization.

Donnelly and Hutton (1976 a, b; Table 1-2) were among the first researchers to publish data on body composition of calves under 100 kg of BW. Others (Reid et al. 1955; Simpfendorfer, 1974; Magia, 1975) published data soon after that contradicted the results of Donnelly and Hutton (1976 a, b) and indicated that diet, breed, or stage a maturity do not influence body composition. For over 25 years, this area of research was dormant and it was not until Murphy et al. (1991) and Waldo et al. (1997) observed that dairy cattle have changed greatly over the past 25 years and now have greater mature body size and possibly greater secretion of growth hormone was this area revisited.

Diaz et al. (2001; Table 1-2) was one of the first recent studies to evaluate the effects of diet on body composition of milk fed calves. In this study, calves were fed a

30% CP, 20% fat milk replacer at different rates to achieve gains of 500, 950, and 1400 g/d. The objective of their study was to determine whether or not the current NRC ( 1989) energy requirements for growth in young calves were accurate and if feeding those amounts of energy affected the protein requirement for lean tissue growth. Calves were harvested at 65, 85, or 105 kg of BW and the actual gains were 560, 973, and 1100 g/d. As the authors expected, with an increase in energy through increased intake, fat and protein deposition increased. As empty body weight gain increased the fat content of the gain increased and the protein content decreased (Table 1-2). These results are consistent with Donnelly and Hutton (1976 a, b) and agree with the hypothesis that protein requirements may be dependant on energy supply.

Blome et al. (2003; Table 1-2) were the next group to examine the effects of diet on body composition and they fed isocaloric milk replacers that ranged averaged 20% fat and ranged in CP from 16.1 to 25.8%. The objective of this study was to quantify the relationship between protein levels in milk replacers and the relationship with growth and composition. Results from this study indicated that calves had greater lean tissue and less fat when dietary CP increased. The authors concluded that dietary CP content and likely the CP:energy ratio influence body weight gain and protein deposition more than energy content alone. This study confirmed and built on earlier results noted by Donnelly and Hutton (1976 a, b) and Diaz et al. (2001).

Tikofsky et al. (2001; Table 1-2) examined how changing the source of energy in milk replacers changes the utilization of those calories. They found a greater utilization of calories from fat for fat deposition than for protein deposition. Thirty-two bull calves were fed either low fat (14.8%), medium fat (21.6%), or high fat (30.6%) milk replacers

and contained an average 25% CP. As the fat content of the milk replacer increased, the lactose content decreased. The treatment diets were isonitrogenous with isocaloric intake conditions. The design of these dietary treatments targeted the objective of determining the effect of the source of energy (fat or carbohydrate) on N utilization and retention.

Calves were not offered dry feed and had ad libitum access to water. Calves were fed until they reached 85 kg live body weight, then were harvested to evaluate carcass composition. The authors concluded that regardless of the source of energy (fat or lactose) the same proportion of consumed protein was retained (13.2%). It was also noted that while there were no differences in final body weights, calves that were fed the high fat diets partitioned their calories for more fat deposition than did calves fed the low fat diets.

A similar experimental approach in mice (Geiger and Canolty, 1978) yielded the same conclusion. To determine the energy deposition coefficient at varying levels of fat, carbohydrate, and protein, mice were fed purified diets at 158, 193, 220, 269, or 308 kcal/kg<sup>0.75</sup> each day and had unlimited access to water. The diets were formulated to have 14%, 29%, or 43% protein; 17%, 38%, or 53% fat; and ranged from 68% to 5% carbohydrate. The mice were not fed at rates of equal protein and energy intake as in Tikofsky et al. (2001), however Geiger and Canolty found that as the ratio of fat to carbohydrate increased, the energy deposition coefficient for fat increased. But at high protein levels (43%) there was no effect of fat to carbohydrate ratio on the deposition of fat most likely because fat was being used as a fuel source for protein deposition. These results indicate that the influence of the carbohydrate to fat ratio on energy deposition is protein dependent. The source of energy in the milk replacer does not appear to effect fat

deposition as significantly as does the level of energy or protein. Protein content in the Tikofsky et al. (2001) study averaged 25% CP which may not be high enough to meet requirements for lean growth at lower levels of fat thus producing not difference in retained protein.

Bartlett et al. (2006; Table 1-2) how the effects of altering dietary protein to energy ratios would change if energy intake varied. The basic design was to increase CP (14, 18, 22, and 26%) content while maintaining isocaloric intake. Calves were fed at either 1.25 or 1.75% of BW. As a percent of EBW, water and CP content were lower in calves fed at 1.75% of BW and increased with increasing CP in the diet. Fat (% of EBW) increased with increase in feeding rate and decreased with the increase in dietary CP. Empty body weight, CP, fat, ash and water gain increased with increasing feeding rate (1.75% of BW) and EBW, CP, and water increased as dietary CP increased while fat gain decreased. As a percent of EBW gain, fat was greater with an increase in feeding rate, however there were no effects of feeding rate on CP or water as a percent of EBW gain.

Theoretically, as CP and water gain increase, fat gain should decrease given that these three comprise the majority of body composition. That is evident in this data set as well as others (Diaz et al.,2001; Tikofsky et al. 2001; Blome et al., 2003). Bartlett et al. (2006) also noted that as feeding rate increased so did fat content of the body which agrees with results from Donnelly and Hutton (1976b) that is, that increasing the amount fed increased fat content of the body. Bartlett et al. (2006) concluded that when protein intake is lower than the requirement for lean growth, excess energy that would be used for protein deposition is stored as fat. Results from this study indicate that CP content greater than 22% of the DM was excessive when calves were only fed at 1.25% of their

BW. When calves were fed at 1.75%, and gain is higher, increasing CP content to 26% increased lean tissue gain.

Increasing nutrient supply in milk replacer can cause a decrease in dry feed intake which is important for rumen development. Jaster et al. (1992) noted a decrease in starter intake with high fat levels in milk replacer. The objective of this study was to test feeding additional energy as either fat or increased milk solids to calves raised outdoors during winter months. The treatments were control, control milk replacer plus 113 g/d of fat (MR 113), milk replacer at 15% DM fed at 10% of body weight (MR 15-10), control plus 226 g/d of fat (MR 226), and milk replacer at 15% DM fed at 14% of body weight (MR 15-14). The supplemental fat was choice white grease. Feeding increased milk solids and fat at 226 g/d decreased intake of starter during week 4 compared to control (con= 225 g/d, MR 15-14= 54 g/d, MR 226= 89 g/d). No body weight gain differences were detected after weaning, but calves fed increased solids or increased fat did have higher gains compared to control from d 7 to 28. The cost of additional solids or fat was only slightly offset by the additional gain before weaning. It was not recommended to feed the MR 15-14 diet or MR 226 because of observed decrease in starter intakes.

Increasing N intake while keeping energy intake constant did not increase N digestibility, but did increase N digested (g/d) and N retained (g/d) (Diaz et al.,2001; Blome et al., 2003; Table 1-3; Table 1-4). Changing the carbohydrate to fat ratio to alter caloric source but supply an isocaloric intake did not change N retention (Tikofsky et al., 2001). These data suggest that increasing protein and energy supply and finding the most appropriate energy to protein ratio in young heifer diets will increase growth efficiency of calves.

The results of the studies above indicate that increasing energy content of the milk replacer will improve lean growth only when CP content of the milk replacer is not limiting. The 'breakpoint' on milk replacer content appears to be around 22% when fed at a normal energy rate. Also, feeding milk replacer at a greater rate appears to affect lean growth as long as protein content is also increased.

Although these studies have improved the way we feed newborn calves, there is still very little data concerning N and P retention and excretion in heifer calves. Milk replacers are typically at least 90% digestible, which leaves small amounts for fecal excretion, but with the millions of calves born each year in the U.S., even small amounts of excretion can add up over time.

### **Nitrogen Metabolism in Post-Weaned Heifers**

Nitrogen utilization varies greatly with dietary N content. Increasing N in the diet will increase efficiency of N utilization until it is in excess of maintenance and growth requirements; additional N fed beyond that point is excreted. Diet digestibility can also affect N utilization. Marini and Van Amburgh (2003; Table 1-5) investigated N recycling and metabolism in dairy heifers by feeding isocaloric diets that were designed to be equal in NDF. These diets were 30% brome hay and 70% of a pelleted mixture that contained 1.44, 1.89, 2.50, 2.97, or 3.40% N. They used urea entry rates into the urine and into and out of the gut as indicators of N recycling. As the N content of the diet increased, the amount of urinary N increased, but fecal N did not (Table 1-5).

As a percent of N intake, the urea entry rate to the gastrointestinal tract (i.e. recycled urea) did not change among treatments, however the urine urea entry rate (i.e. urea production) was different in animals fed the low N diet (35 vs. 66%) versus the high

N diet. The heifers used in this trial were relatively young (BW of  $204 \pm 2$  kg) and still growing. The findings from this trial are similar to those found by Archibeque et al. (2001; Table 1-5) who measured urea flux in growing beef steers and reported that the urea entry rate was 60% of N intake. Huntington et al. (1996), however reported much higher urea entry rates with mature steers (380 kg). In the Marini and Van Amburgh (2003) trial, urea N accounted for 92% of the excess N fed in the 3.40% N diet compared to the 1.45% N diet. Huntington et al. (1996) noted a decrease in the ratio of urea produced to urea excreted in isocalorically and isonitrogenously fed beef steers, as the proportion of NDF increased in the diet.

Gabler and Heinrichs (2003; Table 1-5) fed four diets with different levels of CP (11.9, 16.7, 18.1, and 20.1) with a constant ME intake (2.6 Mcal/kg DM) to heifers at beginning at 146 d of age and 152.8 kg BW. They measured rumen NH<sub>3</sub>, N balance, total tract apparent digestibility, and urinary excretion of purine derivatives in 20 d periods. Heifers were fed once daily and then allowed access to an exercise lot post feeding. The authors noted that feed was consumed quickly and thus no refusals were analyzed. The results of this study were in agreement with Marini and Van Amburgh (2003) in that as CP intake increased, fecal N did not change, but urinary N excretion increased (Table 1-5). Urinary excretion of urea N increased with CP content, however as a percent of total urine N excreted, urea N plateaued at higher CP concentrations.

When dairy heifers were fed 9-21% CP (Gabler and Heinrichs, 2003; Marinini and Van Amburgh, 2003) delivered in two very different rations yet still typical of dairy heifer feeding standards, there were no changes in N retention as a percent of intake.

There were no changes in fecal N excretion (g/d), but urinary N excretion was lower in

heifers fed less than 16% CP compared to those fed greater than 18% CP (Gabler and Heinrichs, 2003; Marinini and Van Amburgh, 2003; Table 1-5).

When beef steers were fed diets of gamagrass or switchgrass (9 or 11% CP) with a corn/mineral supplement (typical of supplements fed to grazing animals), N intake increased and N retention decreased according to type of forage (gamagrass or switchgrass) and the amount of N applied to pasture. Urinary and fecal N values were higher with increased N intake and high N fertilization (Archibeque et al.,2001; Table 1-5). All of these diets had similar DM and N digestibilities, which generally did not differ according to treatments. Increasing the amount of N offered in the diet without increasing energy available (i.e. increased digestibility) does not increase the efficiency of N utilization.

While this research is well accepted, more work is needed to improve our understanding of nutrient metabolism in post-weaned dairy heifers. The animal units in the Huntington (1996) and Archibeque (2001) studies were beef steers and not dairy heifers. Also, the Marini and Van Amburgh (2003) study focuses on dairy heifers, but the dietary treatments were only designed to examine urea transport and N recycling. Gabler and Heinrichs (2003) focus on CP content and NDF levels in the diets were not different. Also, these studies do not include data on metabolism of P which has become a major nutrient of concern.

#### **Summary**

In the past, animal nutritionists and veterinarians have evaluated diets fed to cattle in terms of growth rates. While it is still important that rations fed now maintain support of growth rates, the focus of formulating rations is changing to include nutrient

management. Ruminant animals have been considered very efficient simply because they have the ability to make use of feed resources that monogastric animals cannot use. As we move forward, we have to find new ways to make the most of the abilities of these animals in order to reduce nutrient excretion.

Conclusions from the studies above led us to develop an experimental designs evaluating 1) the effects of fiber type and content on manure nutrient excretion and nutrient utilization in bred heifers, 2) the effect of limit feeding bred heifers on these parameters, and 3) the effect of increasing milk replacer fat content at constant CP and fed at elevated feeding rates on growth and composition of gain in pre-weaned calves.

Table 1-1. Composition of whole milk, dried skim milk, and dried whey (% DM).

Ingredient	Crude Protein	Crude Fat	Lactose	Ca	P
		% of dry n	natter <sup>1</sup>		
Whole Milk	25.6	29.6	39.2	0.95	0.76
Dried skim milk	34	0.1	54	1.25	1.06
Whey Protein Concentrate	34	3.5	52	0.54	0.67
Dried Whey	12	0.2	74	0.90	0.81

Adapted from Davis and Drackley, 1998.

Table 1-2 Empty Body Weight (EBW) gain and composition of gain in calves fed milk replacers varying in protein and energy.

	A 000	Weeks on		EBW gain,	C	P Gain	Fa	t Gain	A	sh Gain	
Animal	Age wk	Study	n	kg	kg	% EBW gain	kg	% EBW gain	kg	% EBW gain	Citation
Bull	<1	6	8- 9	11.3	2.25	20.3	1.85	15.4	0.70	6.01	1
Bull	<1	6	8- 9	12.7	2.74	21.6	1.59	12.7	0.81	6.32	1
Bull	<1	6	8- 9	15.6	3.32	20.7	1.39	8.70	0.78	4.84	1
Bull	<1	6	8- 9	18.6	4.03	22.0	1.41	6.74	0.86	5.12	1
Bull	<1	10	18	23.6	5.77	24.4	1.94	8.20	-	-	2
Bull	<1	14	18	37.5	9.60	25.6	2.89	7.70	-	-	2
Bull	<1	4	18	55.7	12.74	22.9	3.90	7.00	-	-	2
Bull	<1	6	18	24.9	5.00	20.1	3.60	14.5	-	-	2
Bull	<1	9	18	42.0	9.24	22.0	6.83	16.3	-	-	2
Bull	<1	3	18	52.2	11.1	19.4	9.67	15.2	-	-	2
Bull	<1	5	18	24.9	5.73	23.1	3.07	12.4	-	-	2
Bull	<1	7	18	40.8	8.90	21.8	5.73	14.0	-	-	2
Bull	<1	7	18	58.7	11.9	20.3	8.92	15.2	-	-	2

<sup>\*</sup>Citation: 1 = Blome et al. (2003), 2 = Diaz et al. (2001)

Table 1-2 (cont.) Empty Body Weight (EBW) gain and composition of gain in calves fed milk replacers varying in protein and energy.

					CP Gain		Fat	Gain	A	_	
	Age,	Weeks on		EBW gain,		% EBW		% EBW		% EBW	
Animal	wk	Study	n	kg	kg	gain	kg	gain	kg	gain	Citation*
Bull	2	6	6	8.92	1.51	17.3	1.93	21.1	0.39	4.10	1
Bull	2	6	6	10.5	1.96	17.9	1.51	14.4	0.42	4.20	1
Bull	2	6	6	13.4	2.76	20.2	1.58	11.3	0.39	2.90	1
Bull	2	6	6	12.0	2.56	21.1	1.16	9.50	0.35	2.90	1
Bull	2	6	6	17.2	2.77	16.0	3.78	22.1	0.63	3.60	1
Bull	2	6	6	18.7	3.36	17.7	3.40	17.8	0.32	1.70	1
Bull	2	6	6	23.4	4.34	18.3	3.05	12.9	0.81	3.50	1
Bull	2	6	6	24.3	4.76	19.3	2.84	11.5	0.98	3.90	1
Heifer	<1	10	32	33.2	5.77	17.3	3.88	11.7	1.22	3.67	2
Heifer	<1	10	32	34.4	5.70	16.6	5.11	14.9	1.09	3.20	2
Heifer	<1	10	32	35.9	6.08	16.9	6.12	12.0	1.07	2.98	2

<sup>\* 1=</sup> Bartlett (2006); 2 = Tikofsky et al. (2001)

Table 1-3 Nitrogen intake, digestibility, and fecal excretion in pre-weaned heifers and bulls

								Fec	al N excret	ion	
Animal	Age, wk	Weeks on Study	n	N intake, g/d	SE	N digestibility,%	SE	g/d	% N intake	SE	Citation*
Bull	<1	6	8-9	19.2 <sup>1</sup>		$96.3^2$		$2.3^{3}$	11.9		1
Bull	<1	6	8-9	23.2		92.9		3.3	14.2		1
Bull	<1	6	8-9	29.4		93.8		3.6	12.2		1
Bull	<1	6	8-9	33.9		95.5		3.3	9.7		1
Bull	<1	6	18	39.9	N/A	93.8	0.58	-	-		2
Bull	<1	10	18	44.4		92.5		-	-		2
Bull	<1	14	18	44.9		92.3		-	-		2
Bull	<1	4	18	56.6		92.5		-	-		2
Bull	<1	6	18	69.9		92.5		-	-		2
Bull	<1	9	18	72.2		93.9		-	-		2
Bull	<1	3	18	55.3		92.8		-	-		2
Bull	<1	5	18	72.5		94.2		-	-		2
Bull	<1	7	18	82.7		93.5		-	-		2

<sup>\*</sup>Citation: 1=Blome et al. (2003), 2= Diaz et al. (2001), 3= Tikofsky et al. (2001)  $^{1,2,3}$ Linear effect of intake N (P < 0.001), fecal N excretion (P < 0.04), and N digestibility (P < 0.05).

Table 1-3 (cont.) Nitrogen intake, digestibility, and fecal excretion in pre-weaned heifers and bulls

		Weeks						Fec			
Animal	Age, wk	on Study	n N intake, SE diagetibility %		digestibility %		SE	g/d	% N intake	SE	Citation*
Heifer	<1	10	32	23.1	0.01	93.0	N/A	-	-		3
Heifer	<1	10	32	23.3		93.0		-	-		3
Heifer	<1	10	32	22.4		93.0		-	-		3

<sup>\*</sup>Citation: 1=Blome et al. (2003), 2= Diaz et al. (2001), 3= Tikofsky et al. (2001) <sup>1</sup> N/A= SE not available because numbers were calculated from data presented in text.

Table 1-4. Nitrogen digested, retained and urinary excretion in pre-weaned heifers and bulls

					N retention		Urinary N			
Animal	Age, wk	Weeks on Study	n	N digested, g/d	g/d	% N intake	g/d	% N intake	N excretion, g/d	Citation*
Bull	<1	6	8-9	16.9 <sup>1</sup>	$7.6^{2}$	39.0	9.3 <sup>3</sup>	48.4	11.6	1
Bull	<1	6	8-9	20.0	9.0	38.4	11.0	47.2	14.3	1
Bull	<1	6	8-9	25.8	13.2	44.6	12.6	42.9	16.2	1
Bull	<1	6	8-9	30.6	15.6	45.9	15.0	44.2	18.3	1
Bull	<1	10	18	37.5	29.0 <sup>a</sup>	72.7	-	-	2.5	2
Bull	<1	14	18	41.1	25.9 <sup>b</sup>	58.4	-	-	3.3	2
Bull	<1	4	18	41.5	18.3 <sup>c</sup>	40.6	-	-	3.5	2
Bull	<1	6	18	52.4	49.4 <sup>a</sup>	87.1	-	-	4.3	2
Bull	<1	9	18	64.6	51.0°a	73.0	-	-	5.2	2
Bull	<1	3	18	67.8	43.3 <sup>b</sup>	59.9	-	-	4.4	2
Bull	<1	5	18	51.3	51.3 <sup>a</sup>	92.8	-	-	4.0	2
Bull	<1	7	18	68.3	61.4 <sup>b</sup>	84.8	-	-	4.2	2
Bull	<1	7	18	77.4	54.6 a	66.0	-	-	5.4	2

<sup>\*</sup>Citation: 1=Blome et al. (2003), 2= Diaz et al. (2001), 3= Tikofsky et al. (2001)  $^{1,2,3}$ Linear effect of N digested (P < 0.001), N retained (P < 0.001), urinary N excretion (P < 0.001).

Table 1-4 (cont.) Nitrogen digested, retained and urinary excretion in pre-weaned heifers and bulls

					N	N retention		inary N		
Animal	Age, wk	Weeks on Study	n	N digested, g/d	g/d	% N intake	g/d	% N intake	N excretion, g/d	Citation*
Heifer	<1	10	32	21.5	10.3	44.4	-	-	1.6	3
Heifer	<1	10	32	21.7	10.1	43.5	-	-	1.6	3
Heifer	<1	10	32	20.8	10.8	48.3	-	-	1.5	3

<sup>\*</sup>Citation: 1=Blome et al. (2003), 2= Diaz et al. (2001), 3= Tikofsky et al. (2001)

Table 1-5. Nitrogen intake, digestibility, and fecal excretion in post-weaned heifers and steers

							Fecal N excretion			
Animal	BW, kg	n	N intake, g/d	SE	N digestibility,%	SE	g/d	% N intake	SE	Citation*
Beef steer	$217 \pm 15$	8	$64.0^{a}$	1.73	51.8 <sup>a</sup>	1.52	30.8 a	48.1	0.78	1
Beef steer	$217 \pm 15$	8	80.5 <sup>b</sup>		62.4 <sup>b</sup>		30.4 <sup>a</sup>	37.7		1
Beef steer	$217 \pm 15$	8	62.2 <sup>a</sup>		55.2 <sup>a</sup>		27.7 <sup>b</sup>	44.5		1
Beef steer	$217 \pm 15$	8	72.3 <sup>b</sup>		59.1 <sup>b</sup>		29.0 <sup>b</sup>	40.1		1
Dairy heifer	$153 \pm 8.6$	4	62.1 <sup>a</sup>	6.04	69.6 <sup>a</sup>	1.80	18.2	29.3	1.21	2
Dairy heifer	$153 \pm 8.6$	4	88.0 <sup>b</sup>		77.3 <sup>b</sup>		20.0	22.7		2
Dairy heifer	$153 \pm 8.6$	4	96.0°		79.7 <sup>b</sup>		19.3	20.1		2
Dairy heifer	$153 \pm 8.6$	4	105°		80.7 <sup>b</sup>		20.3	19.2		2
Dairy heifer	$267 \pm 3.6$	4	87.6	$5.10^{1}$	47.1	$N/A^2$	46.3	52.9	1.22	3
Dairy heifer	$267 \pm 3.6$	4	110		55.1		49.6	44.9		3
Dairy heifer	$267 \pm 3.6$	4	148		66.6		49.2	33.4		3
Dairy heifer	$267 \pm 3.6$	4	179		70.9		52.0	29.1		3
Dairy heifer	$267 \pm 3.6$	4	266		75.3		50.3	24.7		3

<sup>\*</sup>Citation: 1= Archibeque et al. (2001), 2= Gabler and Heinrichs (2003), 3= Marini and Van Amburgh (2003)  $^{1}$  Means from same study within a column with different superscripts are different, P < 0.05.  $^{2}$  N/A= SE not available because numbers were calculated from data presented in text.

Table 1-5 (cont.) Nitrogen digested, retained and urinary excretion in post-weaned heifers and steers

•	,				N retention				Urin	ary N		
Animal	BW, kg	n	N digested, g/d <sup>1</sup>	SE	g/d	SE	% N intake	SE	g/d	SE	% total N excretion	Citation*
Beef steer	217 ± 15	8	33.2 <sup>a</sup>	1.75	19.5	0.198	30.5	N/A <sup>2</sup>	13.7 <sup>a</sup>	0.635	30.8	1
Beef steer	217 ± 15	8	50.2 <sup>b</sup>		23.5		29.2		26.7 <sup>b</sup>		46.8	1
Beef steer	217 ± 15	8	34.5 <sup>a</sup>		20.5		33.0		14.0 <sup>a</sup>		33.6	1
Beef steer	217 ± 15	8	43.3 <sup>b</sup>		18.1		25.0		25.2 <sup>b</sup>		46.5	1
Dairy heifer	153 ± 8.6	4	43.2	N/A	21.2	N/A	30.2	6.67	22.7 <sup>a</sup>	3.50	55.5	2
Dairy heifer	$153 \pm 8.6$	4	68.1		34.0		37.7		34.0°		63.0	2
Dairy heifer	$153 \pm 8.6$	4	76.5		30.0		32.3		46.7 <sup>b</sup>		70.8	2
Dairy heifer	153 ± 8.6	4	85.3		23.2		22.2		62.2 °		75.4	2
Dairy heifer	$267 \pm 3.6$	4	41.3	N/A	19.7	3.99	22.5	N/A	21.7	5.88	31.9	3
Dairy heifer	$267 \pm 3.6$	4	60.8		24.8		22.5		36.1		42.1	3
Dairy heifer	$267 \pm 3.6$	4	98.3		29.6		20.0		68.7		58.3	3
Dairy heifer	$267 \pm 3.6$	4	126		32.3		18.1		94.3		64.5	3
Dairy heifer	267 ± 3.6	4	153		32.4		15.9		120		70.6	3

<sup>\*</sup>Citation: 1= Archibeque et al. (2001), 2= Gabler and Heinrichs (2003), 3= Marini and Van Amburgh (2003)  $^{1}$  Means from same study within a column with different superscripts are different, P < 0.05.  $^{2}$  N/A= SE not available because numbers were calculated from data presented in text.

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# CHAPTER 2 NITROGEN AND PHOSPHORUS RETENTION, PARTITIONING, AND EXCRETION IN LATE GESTATION DAIRY HEIFERS

# **Interpretive Summary**

New environmental regulations are requiring heifer operations to carry nutrient management permits. Very little published data exists concerning nutrient excretion in heifers so this study examined manure and nutrient excretion in heifers fed diets with different levels of forage and by-products. Limit feeding a low forage ration increased urine output by 5-fold over heifers fed high forage. Total nutrient retention was not different according to diet, however heifers fed low forage partitioned more N to urine which could have negative implications for air quality.

#### Abstract

The objectives of this study were to evaluate the effect of varying feed intake and proportions of forage, grain, and byproducts on growth and excretion of urine, feces, nitrogen, and P in growing heifers. Eighteen Holstein heifers confirmed pregnant were grouped by due date and fed one of three diets (n = 6 per treatment) for the last 14 weeks of pregnancy. Diets were high forage, fed ad libitum (HF); byproduct-based (BP); or low forage (LF), fed at 75% of the HF diet. Diets were designed to supply equal quantities of phosphorus, nitrogen, and metabolizable energy. Total collection of feces and urine was conducted in weeks 14, 10, 6 and 2 week pre-partum. The HF ration was 90.7% forage, 13.7% CP, and contained orchardgrass hay, corn silage, corn grain, soybean meal 44%, and a vitamin-mineral pre-mix. The LF ration was 45.3% forage and 17.8% CP, and fed at 89% of ad libitum. The BP diet was 46.2% forage and 14.0% CP, with 70% of the grain mix space replaced with soybean hulls and cottonseed hulls in a 1:1 ratio. The

effect of diet was analyzed with repeated measures, using preplanned contrasts to compare HF with LF and HF with BP. As expected, heifers fed HF and BP had greater DMI than the heifers limit-fed LF and there was no effect of diet on average daily gain or BW. Intake and digestibility of N was lower, and fecal N excretion higher, in heifers fed HF and BP than heifers fed LF. Mean feces excretion on both wet and dry basis was highest in heifers fed HF, but heifers fed LF excreted more urine than those fed HF or BP. Despite differences in urine output, diet had no effect on urea N excretion but there was a trend for heifers fed the high forage ration to excrete more urinary N compared to those fed LF. Measured manure and urine excretion from heifers fed LF was greater than current ASAE values, while heifers fed HF excreted less manure and urine than predicted. Heifers achieving similar rates of gain from diets differing in forage, grain and byproduct content excreted widely varying quantities of manure.

**Key Words**: Manure excretion, dairy heifer growth, nutrient excretion

#### Introduction

Concentrated animal feeding operations (CAFO) have been recognized as major contributors to nutrient contamination of surface waters. The EPA has recently published permit qualifications and regulations specific to different species of livestock. These publications detail the requirements necessary to obtain and maintain a permit (EPA, 2002).

With the changes in the definition of CAFO and the inclusion of smaller farms, nutrient management planning is a priority for livestock producers. These revised regulations do not just affect dairy producers with lactating cows; they also affect heifer growers, calf ranches, and any facility where water may come in contact with animal

waste as it exits the farm. There is an abundance of information on manure nutrient excretion from lactating cows (St-Pierre and Threan 1999; Knowlton et al. 2001; Haig et al. 2002; Kebreab et al. 2002; Davidson, et al. 2003; Dou et al. 2003), but data about heifer nutrient excretion are scarce.

Diets fed to dairy heifers vary widely, and fiber and energy concentration and source can affect nutrient digestibility and nutrient excretion, both nitrogen (N) and phosphorus (P), and growth. Raising dairy heifers is the second largest expense on a dairy farm (Heinrichs, 1993) and enabling heifers to reach puberty and pregnancy faster to become a productive member of the heard is economically beneficial to the producer. Altering the forage to concentrate ratio to increase concentrate intake can improve prepubertal ADG and help producers grow heifers to ideal breeding weights faster (Zanton, 2005). Our objectives were to evaluate the effect of varying forage type and content on growth, efficiency of gain, and excretion of feces, urine, N, and P in growing, gestating dairy heifers.

#### **Materials and Methods**

#### Animals and Diets

Eighteen Holstein heifers  $(494 \pm 34 \text{ kg} \text{ and } 21.3 \pm 1.4 \text{ months of age at start})$  were fed treatment diets for the last 14 projected weeks of pregnancy. Heifers were grouped according to due date and were assigned randomly to one of three treatment diets formulated to provide equal intake of ME, P and N. Heifers were housed in three pens in a partially open barn. Each pen had 6 heifer-sized Calan gates and was bedded with wood shavings. The high forage (HF), by-product (BP), and low forage (LF) diets were formulated to contain 90.7, 46.2 and 45.3% forage, respectively. All diets contained a

mixture of chopped orchardgrass hay and corn silage as the forage source. All ration ingredients were mixed in a Calan Data Ranger for approximately 15 min and were delivered to heifers as a total mixed ration (TMR) once daily at 1300h. Water was added to all diets to decrease DM and increase palatability. The heifers fed the HF diet were allowed ad libitum intake. The concentrate portion of the HF diet was comprised of soybean meal, mineral mix and calcium carbonate (Table 2-1). The BP diet was equal in forage content to the LF diet, and the grain mix contained a 1:1 mixture of soybean hulls (SH) and cottonseed hulls (CSH) that was mixed with corn, soybean meal and mineral mix. Feed offered to the heifers fed the BP diet was matched to the intake of the HF heifers. The concentrate portion of the LF diet was composed of corn, soybean meal and mineral mix; feed offered was restricted to 89% of the intake of the HF heifers. Heifers fed HF were allowed ad libitum intake and feed offered was increased daily if there were less than 10% refusals. This experiment was conducted under approval of the Virginia Tech Animal Care and Use Committee.

# Sample Collection

Total collection of feces, urine and feed refusals was conducted in 14, 10, 6 and 2 wks pre-partum. Heifers were housed in groups, but were fed individually via Calan doors throughout the study, and moved to individual stalls for the 5-d collection period. On day one of each collection period, a sterile Foley catheter (22 French, 75cc, C. R. Bard, Inc., Covington, GA) was placed in the urethra for total collection of urine. All excreted urine was weighed and a subsample (130 ml urine) collected every 6 h. Remaining urine was acidified (7.7 ml of 2N H<sub>2</sub>SO<sub>4</sub>/kg of urine) every 6 h, pooled, and sampled after 24 h. Acidified urine was frozen and stored for later analysis (described

below). Unacidified subsamples were analyzed immediately as required for specific assays. All excreted feces were collected, stored in a sealed container, and weighed, mixed and subsampled after 24 h. One subsample per heifer was frozen for later analysis and one sample per heifer was analyzed immediately (described below). Feed refusals were weighed daily and feed and refusal samples were collected on day 5 of each collection period. Body weight (BW), wither height, and hip height were recorded for two consecutive days before the beginning of each collection period. Initial BW and stature measures were taken at the end of the 2-week adaptation period when each heifer started the study. Each time, heifers were measured from the front left hoof to the top of the withers for wither height, from rear left hoof to the top of the hip for hip height, and from center of left hip laterally across to center of right hip for hip width using a cloth measuring tape.

## Lab Analysis

Feed, refusals, and fecal samples were dried at  $60^{\circ}$  C to a constant weight then ground through a 1-mm screen in a Wiley Mill (Arthur H. Thomas, Philadelphia, PA). These samples were analyzed in duplicate for Ca, P, and total Kjeldahl N (TKN; AOAC, 1984). Feed and refusals were analyzed sequentially for NDF and ADF according to Van Soest et al. (1991) with the addition of  $\alpha$ -amylase in the initial wash. Feces samples were analyzed using the same method without  $\alpha$ -amylase in the initial wash. Urine samples were analyzed for Ca and P according to standard methods (AOAC, 1984). Urine samples were analyzed in duplicate for TKN, urea N, total suspended solids (TSS) and volatile suspended solids (VSS; AOAC, 1984; APHA, 1998). Feces samples were also analyzed for TSS and VSS.

### Statistical Analysis

Diet composition was analyzed using PROC GLM of SAS. Growth and nutrient utilization data were analyzed using the RANDOM and REPEATED statements of the PROC MIXED procedure of SAS. Effects of treatment, heifer, group, period or day, and the interaction of treatment and group were represented by the model:

$$Y = \mu + T_i + G_i + H_{k(ij)} + P_l + TG_{(ij)} + E_{ijkl}$$

Where:

 $\mu$  = overall population mean

 $T_i$  = effect of  $i^{th}$  diet (i = 1, 2, 3; fixed);

 $G_J$  = effect of  $j^{th}$  group (j = 1, 2, 3; fixed);

 $H_K = \text{effect of } k^{\text{th}} \text{ heifer } (k = 1, 2, 3, ...6; \text{ random});$ 

 $P_1$ = effect of  $l^{th}$  period (l = 1, 2, 3, 4; fixed);

 $TG_{(ij)}$  = interaction of diet and group; and

 $E_{ijkl}$ = residual error term (random)

Preplanned contrasts were used to compare HF and BP with LF and HF with BP. Data are reported as LS means and differences were declared significant at P < 0.05.

# Comparison to ASAE Standards

Equations from the new ASAE Manure Production and Characteristics Standards (2005) were used to compare the data from this study to the predicted values. Individual heifer observations from each period from the current data set were used and the difference between actual and predicted values was calculated. Regression analysis was done on observed and predicted values for both dairy and beef equations for manure, N, and P excretion. The following equations from Section 4.0 Equations for As-Excreted

Manure Characteristics Estimates for Beef Cattle and Section 5.0 Equations for As-Excreted Manure Characteristics for Dairy Cattle were evaluated.

**Beef Standard Equations** 

[4.3.1-1] 
$$DM_E = [DMI*(1-DMD/100)] + 20.3*(0.06*BW)$$

[4.3.2-3] 
$$OM_E = [DMI*(1-ASH/100)*(1-OMD/100) + 17*(0.06*BW)]$$

$$[4.3.3-5] \qquad N_{E-T} = \sum_{x=1}^{n} (DMI_x *C_{cp-x}*DOF_x/6.25) - [41.2*(BW_f - BW_i)] + \\ [0.243*DOF_{T_t}*[(BW_f + BW_i)/2]^{0.75}*(SRW/(BW_f*0.96))^{0.75}*[(BW_f - BW_i)/DOF_T]^{1.097}]$$

$$[4.3.4-6] \qquad P_{E-T} = \sum_{x=1}^{n} (DMI_x *C_{p-x}*DOF_x) - [10.0*(BW_f - BW_i)] + \{5.92*10^{-2}*DOF_T **[(BW_f + BW_i)/2]^{0.75}*(SRW/(BW_f *0.96))^{0.75}*[(BW_f - BW_i)/DOF_T]^{1.097}]$$

Where  $DM_E = Dry$  matter excretion per animal per day

 $N_{E-T}$  = Total N excretion per finished animal

 $P_{E-T}$  = Total P excretion per finished animal

DOF = Day on feed

BW<sub>f</sub>= Final body weight

 $BW_i = Initial body weight$ 

SRW = Standard reference weight for expected final body fat (478kg)

 $C_{cp}$  = Concentration of CP in the ration DM

 $C_p$ = Concentration of P in the ration DM

**Dairy Standard Equations** 

[5.3.3-5] 
$$M_E = (DMI \times 3.886) - (BW \times 0.029) + 5.641$$

[5.3.0-19] 
$$N_E = (DMI \times Ccp \times 78.390) + 51.350$$

[5.3.12-24]  $P_E = (DMI \times 1000) \times Cp$ 

Where  $M_E$  = Total wet manure excretion per animal per day

 $N_E$  = Total N excretion per animal per day

 $P_E$  = Total P excretion per animal per day

 $C_{cp}$  = Concentration of CP in the ration DM

 $C_p$  = Concentration of P in the ration DM

#### **Results and Discussion**

Diet Composition, Intake, and Growth Measures

The Effects of Limit-Feeding Low Forage. Ingredient composition of each treatment diet is listed in Table 1. The LF ration was higher in CP than HF and BP, and was much lower in NDF and ADF concentration. Calcium concentration did not change when forage content was decreased, however P concentration was higher in the LF ration compared to HF and BP (0.41 vs. 0.29 Table 2-2).

Dry matter intake (kg/d) of the LF ration was designed to be 75% of the intake of heifers fed HF and BP. Actual DMI of heifers fed LF was 85% of that of heifers fed HF most likely due to variation associated with different feeders. Organic matter intake was lower in heifers fed LF compared to HF and BP (Table 2-3). There were no significant differences in growth measures across the treatments (Table 2-4)

The Effects of Adding By-Products. Adding cottonseed hulls (CSH) to the BP ration decreased the NDF concentration but increased ADF (36.3 vs. 39.5). The addition of cottonseed hulls to the BP ration was likely responsible for the increase in ADF concentration, as they are 65% ADF (NRC, 2001). Calcium concentration also increased

39

with the addition of by-products, however P concentration was not different between diets (Table 2-2).

Heifers fed the BP ration were fed at rates that matched the previous week's intake of heifers fed HF. Because of this lag, DMI was lower in heifers fed BP than in heifers fed HF (Table 2-3). Organic matter intake followed the same trends as DMI and was higher in heifers fed HF compared to BP (Table 2-3).

## Digestibility and Manure Excretion

The Effect of Limit Feeding Low Forage. Apparent DMD was higher in heifers fed the LF ration compared to HF and BP. Organic matter digestibility (OMD) was higher in heifers fed LF compared to HF and BP. Heifers fed the LF ration excreted about a kilogram less feces (DM) per day compared to those fed HF and BP.

As DMD increased, fecal output decreased. An 11% reduction in DMI between high forage rations (HF and BP) and a high concentrate ration (LF) lead to a 25% decrease in fecal output. Driedger and Loerch (1999) noticed a similar trend when dry cows were limit-fed a high corn diet. These authors restricted intake in the cows fed the high corn ration by 29% compared to other cows fed high forage ration and noted a 40% reduction in fecal excretion.

Urinary output was much higher in heifers fed LF compared to those fed HF and BP. Heifers fed the LF ration increased urine output by almost 5-fold compared to heifers fed HF and excreted more than twice the amount of urine compared heifers fed BP. Because of the increase in urinary output in heifers fed LF, total weight of manure excreted was also higher. The increase in urine output may be explained by changes in energy or salt content, may have been a side effect of the reduced gut fill of the LF diet,

or may have been due to behavioral impacts of limit feeding. Johnson and Combs (1991) noted that feeding a high energy diet to multiparous cows during the pre-partum period increased water intake from 104 kg/d to 127 kg/d. The LF ration from the current study is comparable to the high energy ration fed by Johnson and Combs (1991) and if the assumption of increased water intake is made in the heifers fed LF, then the increase in urinary output would be expected (Murphy, 1992). Increased water intake is also commonly an affect of increased salt intake. The rations were not analyzed for salt content, but it was calculated from NRC (2001) values for each feed ingredient. Salt intake was not higher in heifers fed LF and actually tended to be higher in heifers fed HF (0.02 vs. 0.05 kg/d, respectively; Table 2-3) due to the higher content of chlorine in grass hays.

Alternatively, heifers fed the LF ration may have consumed water to increase rumen fill. Those heifers cleaned up their total daily ration by the time rations were mixed and delivered for heifers fed HF and BP (about 45-60 min) and may have consumed more water to replace the typical bulk of the HF ration. In addition, rapid consumption of the day's DM, as a side effect of limit feeding, can lead to behavioral problems. Essentially, increased water intake and therefore increased urine output, may have been driven by boredom. This could also lead to oral behavior problems. Kopp et al. (1986) noted that more curious animals spend more time involved in non-nutritive behaviors, such as sniffing, licking, and biting inanimate objects. Heifers are by nature curious animals and without access to feed could also develop these non-nutritive oral behaviors which could lead to poor oral health.

The Effect of Adding By-Products. Dry matter digestibility was not different between heifers fed HF and BP. Heifers fed the BP ration excreted less feces (kg DM/d) than those fed the HF ration. There were no differences in urinary output or total manure excretion with the addition of BP.

# Nitrogen Intake, Excretion, and Partitioning

The Effects of Limit Feeding Low Forage. Nitrogen intake (g/d) was greater in heifers fed the LF ration compared to those fed HF and BP. Apparent N digestibility was higher in heifers fed LF compared to BP and HF. Because of improved N digestibility in heifers fed LF, fecal excretion of N was greater in heifers fed HF and BP compared to LF and there tended to be an increase in urinary N excretion (87.2 vs. 111, P < 0.07). Heifers fed the LF ration did have a greater overall N retention compared to those fed the HF and BP rations. These results are similar to those of Zanton and Heinrichs (2005), however where they noted no difference in urinary N excretion when heifers were fed rations with either 25% or 75% forage.

Nitrogen retention as a percent of N intake was not different. This indicates that increasing N intake does not increase N retention, proportionally, even with a higher N digestibility. In diets as digestible as the LF ration, this N is more likely to be excreted in a volatile form which can have implications for air and odor quality. Both Marini and Van Amburgh (2001) and Gabler and Heinrichs (2003) reported increasing N excretion with increased N intake due entirely to increased urinary N (no differences in fecal N excretion). Zanton and Heinrichs (2005) did note that fecal N excretion tended to increase when heifers (666 d of age) were fed a 25% forage ration instead of a 75% forage ration.

Urinary N as a percent of N intake, urinary urea N, total N excretion (g/d and percent of N intake) were not affected by forage content in this study.

The Effects of Adding By-Products. Heifers consuming the HF ration had higher N intake than those fed BP (Table 2-5). By design, heifers consuming the HF and BP rations should have similar N intakes and CP percent of the diet was not different, however DMI was lower in heifers fed BP compared to HF and this resulted in a lower N intake. Heifers fed HF also had higher N intake compared to those fed BP, but there was no difference in N digestibility. Fecal and urinary excretion of N was not affected by addition of by-products (Table 2-5).

# Phosphorus Intake, Excretion and Partitioning

The Effects of Limit Feeding Low Forage. Phosphorus intake averaged 33 g/d and did not change with limit feeding. The LF ration was higher in P (% DM) compared to HF and BP, however DMI of heifers fed LF was less than that of those fed HF and BP leaving no significant difference in P intake (g/d). Phosphorus digestibility was higher in heifers fed LF compared to HF and BP. P intake and fecal P excretion varied greatly, dietary P was much greater than NRC (2001) requirements, and digestibility values were not different from zero. Fecal excretion of P was not affected by forage amount, however urinary P excretion was higher in heifers fed LF compared to those fed HF and BP.

Urinary P excretion is typically minimal in lactating dairy cows (< 1 g/d; Morse et al. 1991; Knowlton and Herbein, 2001), but the heifers fed the LF ration excreted almost five times as much urinary P as expected for lactating cows. Total P excretion (g/d) was not different on an absolute basis, however as a percent of P intake heifers fed the LF ration had lower total P excretion compared to HF and BP. Driedger and Loerch (1999)

noticed a similar pattern in non-lactating cows where P excretion was lower as a percentage of intake in cows fed a high energy diet.

The Effects of Adding By-Products. Phosphorus intake did not differ with the inclusion of by-products in the ration. Phosphorus digestibility in heifers fed HF was less than that of BP and both diets resulted in negative P digestibility. A 95% confidence interval for both HF and BP rations included zero, indicating that P digestibility of these two diets were not different from zero. These heifers were excreting essentially all the P they were consuming. It seems contradictory that a gestating animal would not be retaining P, however P requirements in animals of this age and status are very low. There are no data on the pregnancy requirements of late-gestation heifers, but requirements for mature cows of comparable stage range from less than 1 g/d to less than 6 g/d just before calving (NRC, 2001).

Also, P requirements for growth are very low. Erickson et al. (2002) evaluated the P requirement of finishing feedlot steers Diets ranged from 0.16 to 0.40% of DM and even at the low level of 0.16%, no differences in ADG or bone breaking strength were noted. The diets fed in Erickson et al. (2002) contained by-products such as brewers grits and cottonseed hulls. Although these are considered non-traditional diets, most heifer rations also could be classified as non-traditional.

Driedger and Loerch (1999) fed gravid dry cows varying levels of fiber and P in the diet. These cows were fed at nearly 5 times the suggested level of P (0.95-1.1% of DM and 0.25-0.30% of DM, respectively; NRC, 2001). Although these cows were over fed P they still retained about 30% of the intake P. The major difference between a dry cow with calf and a heifer with calf is that cows have been through one or more lactation

cycles and would need to replace bone mineral that had been depleted by the demands of lactation. The heifers in this study did not have the demands of a previous lactation and with very fetal requirements for P likely being minimal in late gestation, they were significantly over-fed P at 0.3-0.4% of DM. The compiled results of these studies suggest that the P requirement of late-gestation large frame dairy heifers needs to be revisited.

Fecal excretion of P tended to be higher in heifers fed HF compared to BP.

Urinary excretion of P was not different between heifers fed HF or BP, but these heifers were excreting as much urinary P as lactating cows.

Very little published data exists concerning P metabolism in heifers. Gabler and Heinrichs (2003) measured P intake and fecal, urinary, and total P excretion in heifers of 146 days of age and found no differences with increasing concentration of protein.

Because of the negative correlation between ration fiber (NDF and ADF) and daily energy intake (Quigely et al.,1986), it is difficult to separate effects of fiber and energy concentration in a ration. In this study, both the ration fiber and energy concentration change with diet. The independent effect of each on P metabolism cannot be determined.

Increased P excretion, as observed in heifers fed HF compared to those fed BP, is important because of environmental concerns across the livestock industries. We observed that heifers in late gestation are excreting 60 to 70% as much P as a lactating cow (Nennich et al., 2005) and are excreting nearly all the P they consumed. This is important considering changes in CAFO regulations to explicitly include stand-alone heifer operations. Reducing P intake in lactating cows is a heavily researched area and this same type of research is needed in heifers.

### Measure of Solids

Total solids (TS) are a measure of the amount of matter in water, which is directly related to turbidity, and consists of both suspended and dissolved material. Volatile solids (VS) are a measure of the organic component of TS. Total and volatile suspended solids (TSS, VSS) are the solids in water that can be trapped by a filter (particulates). Chemical oxygen demand (COD) is a measure of the amount of oxygen required to completely oxidize all the organics. All of these are useful in evaluating water quality, but since both VS and VSS are determined by calculating weight lost after ignition some minerals could volatilize during combustion, making COD a better measure of organic compounds in water (APHA, 1998). These types of measures are not typically done on manure waste, but are common in municipal waste samples. Measure of solids is important to know when designing new manure treatment technologies such as anaerobic digesters.

We observed no differences in fecal or urinary TS, VS, TSS, or VSS concentrations with limit-fed heifers compared to heifers fed high forage diets. There was no difference in either TS or VS concentration when by-products were added to the diet compared to the HF ration, but urinary solids concentration were higher in heifers fed the HF ration compared to those fed BP. Chemical oxygen demand in feces and urine was not affected by the addition of by-products.

## Comparison to ASAE prediction equations

The dairy standard equations were not good predictors of total wet manure excretion ( $R^2 = 0.114$ ). Because heifers fed the LF ration excreted much more than normal urine, the predicted values did not fit well with the data. However the beef standard equations predict total manure excretion on a dry basis and the valuespredicted

with these beef equations did fit the data well ( $R^2 = 0.556$ ). Neither the beef nor the dairy equations predict values that fit well to the N excretion data ( $R^2 = 0.003$ ,  $R^2 = 0.0001$ , respectively) or to the P excretion data ( $R^2 = 0.038$ ,  $R^2 = 0.187$ ; Table 2-8). Overall and within treatment, the beef prediction equations for DM excretion predicted manure excretion well. Equations that are used for predicting manure DM output are likely inherently more precise than those predicting wet amounts or nutrient content.

## **Conclusions**

Growth was similar in heifers fed high and low forage diets and did not change with the addition of by-products. Total nutrient excretion did not differ for heifers fed ad libitum high forage diets compared with limit-fed heifers, but the increase in urine output in heifers fed low forage diets would make manure management more difficult. Limit feeding heifers a low forage ration could be a concern especially in environmentally sensitive areas because additional manure storage capacity would be required.

Low to zero P digestibility and retention in heifers fed LF or HF and BP, respectively, indicates that heifers in late gestation are not retaining significant P; heifers excreted nearly all the P consumed. The current ASAE beef equations are better predictors of DM and P excretion when heifers are fed a low forage ration. Overall, heifers achieving similar rates of gain from diets differing in forage, grain and byproduct content excreted widely varying quantities of manure.

47

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  Annual Symposium Nutritional Biotechnology in the Feed and Food Industries.

Table 2-1. Ingredient composition of three treatment diets fed to Holstein heifers.

		Dietary Treatments	
_	$\mathrm{HF}^1$	$\mathrm{BP}^2$	LF <sup>3</sup>
		% of diet DM	
Orchardgrass hay	71.3	36.3	35.7
Corn silage	19.4	9.90	9.64
Cottonseed hulls	0	16.0	0
Soybean hulls	0	16.0	0
Corn	0	6.50	35.0
Soybean meal, 44%	8.70	14.3	18.6
Mineral	0.60	0.90	0.50
Calcium carbonate	0.09	0.00	0.50

<sup>&</sup>lt;sup>1</sup> HF= high forage
<sup>2</sup> BP= by-product
<sup>3</sup> LF= low forage
<sup>4</sup> Mineral = 4.5% CP, 0.2% Fat, 14.6% Ca, 7.1% P, 20.3% Na, 22.4% Cl, Vit A 138 IU/kg, Vit D 13.2 IU/kg, Vit E 68.1mg/kg.

Table 2-2 Nutrient composition of three treatment diets fed to Holstein heifers.

	Die	etary Treatme	nts			P <	
	$HF^2$	$BP^3$	$\mathrm{LF}^4$	SEm <sup>5</sup>	Treatment	HF vs. LF <sup>6</sup>	HF vs. BP <sup>7</sup>
		% of diet DM					
DM	51.9	51.0	51.7	0.13	0.01	0.29	0.01
$\mathbb{CP}^1$	13.7	14.0	17.8	0.21	0.01	0.01	0.31
NDF	65.9	62.9	43.9	0.25	0.01	0.01	0.01
ADF	36.3	39.5	21.0	0.34	0.01	0.01	0.01
Ca	0.28	0.54	0.51	0.04	0.01	0.08	0.01
P	0.28	0.31	0.41	0.02	0.01	0.01	0.34
Salt <sup>8</sup>	0.62	0.23	0.34	0.03	-	-	-
Ash	7.91	7.79	7.30	0.0003	0.31	0.14	0.75

<sup>&</sup>lt;sup>1</sup>Values calculated from individual ingredient chemical analysis.

<sup>&</sup>lt;sup>2</sup> HF= high forage

<sup>&</sup>lt;sup>3</sup>BP= by-product

<sup>&</sup>lt;sup>4</sup>LF= low forage

 $<sup>^{5}</sup>$ n= 18

<sup>&</sup>lt;sup>6</sup>high forage and by-product diet vs. low forage diet

<sup>&</sup>lt;sup>7</sup>high forage vs. by-product diet <sup>8</sup> Salt values calculated from Na (%) and Cl (%) values for each feed ingredient from NRC (2001) times the DM kg of each ingredient. Values were not statistically compared because there was no variation in the proportions of each ingredient fed.

Table 2-3. Dry matter intake, digestibility and manure production in Holstein heifers fed diets varying in forage content.

	Die	etary Treatme	ents	P <			
	$\mathrm{HF}^1$	$BP^2$	$LF^3$	SEm <sup>4</sup>	Treatment	HF vs. LF <sup>5</sup>	HF vs.BP <sup>6</sup>
DM intake, kg/d	8.93	8.28	7.65	0.06	0.01	0.01	0.01
Apparent DMD,%	58.7	61.8	67.2	1.10	0.01	0.01	0.11
OM intake, kg/d	8.29	7.60	7.08	0.18	0.01	0.01	0.03
OMD,%	60.3	62.8	68.2	0.01	0.01	0.01	0.16
Salt Intake, kg/d <sup>7</sup>	0.05	0.02	0.02	0.001	0.01	0.01	0.01
Feces							
kg DM/d	3.61	3.16	2.54	0.08	0.01	0.01	0.01
kg wet/d	22.4	17.9	15.3	0.73	0.01	0.01	0.01
Urine							
kg/d	8.92	16.9	40.2	5.50	0.02	0.01	0.35
Total Manure kg wet/d	31.3	34.8	55.5	6.2	0.07	0.03	0.71

<sup>&</sup>lt;sup>1</sup> HF= high forage <sup>2</sup> BP= by-product <sup>3</sup> LF= low forage <sup>4</sup> n= 18

<sup>5</sup> high forage and by-product diet vs. low forage diet
6 high forage vs. by-product diet
7 Salt values calculated from addition of Na (%) and Cl (%) values for each feed ingredient from NRC (2001) times the weekly DMI (kg/d) of each treatment diet.

Table 2-4. Body weight and measures in growing Holstein heifers fed diets varying in forage content.

	D	ietary Treati	ments			P<	
	HF <sup>1</sup>	$BP^2$	$LF^3$	SEm <sup>4</sup>	Treatment	HF vs. LF <sup>5</sup>	HF vs. BP <sup>6</sup>
Body Weight gain, kg <sup>7</sup>	58.8	54.1	57.7	7.59	0.90	0.90	0.69
Average Daily Gain, kg/d	0.57	0.52	0.56	0.07	0.89	0.91	0.66
Feed: Gain	21.8	17.1	15.1	1.66	0.08	0.09	0.09
Wither Height, cm	136	136	136	0.81	0.52	0.29	0.73
$\Delta$ Wither Height, cm <sup>7</sup>	1.85	1.52	1.60	0.97	0.98	0.94	0.82
Hip Height, cm	141	140	138	0.99	0.31	0.16	0.61
$\Delta$ Hip Height, cm <sup>7</sup>	-0.68	-0.38	0.56	0.99	0.68	0.41	0.84
Body Condition Score	3.31	3.29	3.26	0.35	0.36	0.27	0.38

HF= high forage
BP= by-product
LF= low forage
n= 18

high forage and by-product diet vs. low forage diet
 high forage vs. by-product diet
 Means calculated by final – initial over entire study.

Table 2-5. Nitrogen digestibility, excretion and retention in Holstein heifers fed diets with varying forage content.

	Di	etary Treatme	ents			P<	
	$\mathrm{HF}^{1}$	$BP^2$	$LF^3$	SEm <sup>4</sup>	Treatment	HF vs. LF <sup>5</sup>	HF vs. BP <sup>6</sup>
N intake, g/d	194	185	218	1.43	0.01	0.01	0.01
N digestibility,%	60.8	59.4	73.2	1.10	0.01	0.01	0.41
Fecal N g/d	74.2	74.8	57.8	1.69	0.01	0.01	0.80
Urine N g/d % of N intake	87.5 46.4	87.7 49.0	111 53.8	9.76 5.10	0.18 0.69	0.07 0.35	0.98 0.72
Urinary Urea N g/d	67.4	50.6	56.0	15.7	0.75	0.89	0.46
Total N excretion g/d % of N intake	161 85.6	163 89.6	168 80.6	8.53 4.30	0.85 0.37	0.57 0.21	0.94 0.52
N retention g/d % of N intake	32.6 14.4	22.7 10.3	49.8 19.3	8.23 4.30	0.08 0.37	0.04 0.21	0.40 0.52

<sup>&</sup>lt;sup>1</sup> HF= high forage <sup>2</sup> BP= by-product <sup>3</sup> LF= low forage <sup>4</sup> n=18

<sup>&</sup>lt;sup>5</sup>high forage and by-product diet vs. low forage diet <sup>6</sup>high forage vs. by-product diet

Table 2-6. Phosphorus digestibility, excretion, and retention in Holstein heifers fed diets varying in forage content.

	Di	etary Treatme	ents		P<		
	$-$ HF $^1$	$BP^2$	LF <sup>3</sup>	SEm	Treatment	HF vs. LF <sup>4</sup>	HF vs.BP <sup>5</sup>
P intake, g/d	30.0	30.9	33.1	1.6	0.35	0.17	0.68
r ilitake, g/u	30.0	30.9	33.1	1.0	0.55	0.17	0.08
P digestibility,%	-34.1	-8.4	12.6	9.2	0.01	0.01	0.05
Fecal P							
g/d	39.4	32.5	29.4	2.9	0.06	0.08	0.11
Urine P							
g/d	0.78	1.67	4.80	0.83	0.01	0.01	0.46
% intake	2.79	5.64	14.9	2.7	0.01	0.01	0.47
Total P excretion							
g/d	40.1	33.9	36.2	2.47	0.22	0.80	0.09
% intake	134	117	105	2.78	0.02	0.02	0.09
P retention							
g/d	-10.4	-3.58	-0.55	2.5	0.02	0.04	0.06
% intake	-36.3	-13.8	-2.32	9.0	0.03	0.04	0.09

<sup>&</sup>lt;sup>1</sup> HF= high forage <sup>2</sup> BP= by-product <sup>3</sup> LF= low forage <sup>4</sup> n=18

<sup>&</sup>lt;sup>5</sup>high forage and by-product diet vs. low forage diet <sup>6</sup>high forage vs. by-product diet

Table 2-7. Manure characteristics from Holstein heifers fed diets varying in forage content.

	Die	etary Treatme	ents			P<	
	$\mathrm{HF}^1$	$BP^2$	$LF^3$	SEm <sup>4</sup>	Treatment	HF vs. LF <sup>5</sup>	HF vs. BP <sup>6</sup>
Feces							
$\mathrm{TS}^7$ , $\mathrm{mg/L}$	$1.7 \times 10^5$	$1.8 \times 10^5$	$1.7 \times 10^5$	$4.5 \times 10^3$	0.15	0.85	0.06
$VS^8$ , mg/L	$1.6 \times 10^4$	$1.8 \times 10^4$	$1.6 \times 10^4$	932	0.50	0.43	0.39
COD <sup>9</sup> , mg/L	$4.5 \times 10^6$	$6.6 \times 10^6$	$3.8 \times 10^6$	$1.5 \times 10^6$	0.47	0.39	0.39
Urine							
TSS <sup>10</sup> , mg/L	429.9	276.5	230.5	45.4	0.05	0.078	0.05
VSS <sup>11</sup> , mg/L	139.6	95.11	91.31	27.96	0.42	0.47	0.28
COD, mg/L	$3.1 \times 10^4$	$3.7 \times 10^4$	$3.4 \times 10^4$	$5 \times 10^4$	0.68	0.97	0.40

<sup>&</sup>lt;sup>1</sup> HF= high forage <sup>2</sup> BP= by-product <sup>3</sup> LF= low forage

 $<sup>^{4}</sup>$  n= 18

<sup>&</sup>lt;sup>5</sup> high forage and by-product diet vs. low forage diet <sup>6</sup> high forage vs. by-product diet <sup>7</sup> Total Solids

Volatile Solids
 Chemical Oxygen Demand
 Total Suspended Solids
 Volatile Suspended Solid

Table 2-8. Comparison of observed and predicted values of manure and nutrient excretion using ASAE Standards (2005).

egression Equation	Item	Standard
-0.0894x + 25.616	Manure	Dairy
= 0.6733x + 1.342	Manure	Beef
-0.0049x + 148.85	N	Dairy
-0.0326x + 117.95	N	Beef
= 0.1017x + 23.308	P	Dairy
= -0.8929x + 155.2	P	Beef
= 0.6733x + 1.342 $-0.0049x + 148.85$ $-0.0326x + 117.95$ $= 0.1017x + 23.308$	Manure N N P	Beef Dairy Beef Dairy

# CHAPTER 3 MILK REPLACER COMPOSITION AND NUTRIENT UTILIZATION IN PRE-WEANED HOLSTEIN CALVES

# **Interpretive Summary**

Calves fed a standard milk replacer (24% CP/ 17% fat) ate more grain than calves fed higher protein and fat milk replacers and feces excretion was highest and urine output lowest. Adding fat to the ration increased fat, but not protein gain. Adding both protein and fat to the 24/17 milk replacer led to an increase in both fat and protein gain. Increasing amount of milk replacer fed from 2 to 3% of BW increased overall EBW and CP gain. The addition of nutrients to a calf ration is costly and results here suggest that it may not be beneficial past 28% CP and 20% fat.

#### **Abstract**

Twenty-four newborn Holstein heifer calves (n=6) were fed one of 4 diets: 24/17 (fed at 350 g/d, 24% CP, 0.53% P); 32/17 (fed at 764 g/d, 32% CP, 0.55% P); 31/24 (fed at 782 g/d, 32% CP, 0.46% P); and 31/24+ (fed at 1177 g/d, 32% CP, 0.46% P). Calves were grouped by age and treatments were assigned randomly within group. Calves were fed 3.4 L of colostrum twice within 16 h of birth. Upon arrival at the research farm, calves were fed a 24/17 milk replacer for the first two feedings. On d 3, treatments were imposed and calf starter (20% CP, 0.48%P) comprised of corn (44.4%), soybean meal (44.4%), cottonseed hulls (11.1%), and molasses (1.0%) was offered free choice. Calves were on study for ~63 d. Total collection of feed refusals, feces and urine was initiated on d 59 ± 2d. Body weight and body size measures were taken weekly. Feces, urine, milk replacers, and starter samples were pooled (25% of each daily sample) by calf or diet, across collection period and analyzed for total Kjeldhal N and total P. All calves were harvested at 63 d to evaluate body composition and mammary development

(reported elsewhere). Preplanned contrasts were used to compare 24/17 to all, 32/17 to 31/24, and 24/17 to 31/24+. Total DMI was not different as calves fed 24/17 consumed more starter than those fed greater amounts of milk replacer. Apparent DMD was lower for calves fed 24/17. Fecal output (kg DM/d) and fecal N excretion were highest in calves fed 24/17 while urine output (kg/d) and urine N excretion were lowest. Nitrogen intake and urine N excretion were highest for calves fed 31/24+ but were not affected by fat content (31/24 vs. 32/17). Nitrogen retention was not improved by increasing energy intake (mean = 34.9%), nor was it improved by increasing the amount fed. Phosphorus digestibility, total excretion, partitioning, and retention were not impacted by treatment. The addition of fat to the milk replacer reduced protein gain (kg and % of EBW), increased fat gain (kg and % of EBW), and decreased ash gain (kg and % of EBW). Increasing the volume fed did increase protein gain, fat gain (kg and % of EBW) and ash gain. These results indicate that 20% fat may not be enough energy to support protein gain when CP is greater than 28% of the diet DM. However, frame growth appeared to increase when calves were fed the 32/17 compared to 31/24, indicated by increased ash gain and increased body measurements.

**Keywords:** calf, milk replacer, nutrient excretion

#### Introduction

Practices for feeding young calves have changed significantly in recent years. There is a trend for producers to grow heifers faster in order to reach puberty at a younger age and become productive sooner. Tozer and Heinrichs (2004) estimated that reducing the age at first calving by only one month can decrease the cost of raising a dairy heifer by up to \$1400. The economics alone are incentive to implement these types of feeding programs, but Danish researchers have shown a negative affect on mammary tissue growth in the early weeks of life with increased

growth due to intensified feeding (Sejrsen et al., 2000). Preliminary data (Akers, unpublished data, 2004) show a dramatic increase in mammary parenchymal mass during calfhood; a nearly 60-fold increase in parenchymal mass was measured from birth to 3 mo of age. This observation tells us that the mammary glands of the heifer calf are anything but quiescent. Brown et al. (2005) demonstrated that increased energy and protein intake associated with accelerated calf growth programs increased growth of mammary parenchyma in calves from 2 to 8 wk of age. These differences did not hold up when heifers were evaluated at 14 wk of age (after weaning), which suggests that the calf is more sensitive to nutrient intake prior to weaning and that the enhancement of mammary development cannot be recovered once the animal is weaned. Smith et al. (2002) demonstrate that in well-managed milk-fed calves, the somatotropic (GH/IGF-I) axis is functionally coordinated and sensitive to nutrient intake and GH.

Much research has been published evaluating the effects of varying protein and energy content of milk replacer on weight gain and N retention in pre-weaned calves. As dietary CP increased lean gain increased when energy was not limiting and fat gain increased when protein was limiting (Jaster et al. 1992; Diaz et al., 2001; Tikofsky et al. 2001; Blome et al. 2003, and Bartlett et al. 2006). Blome et al. (2003) fed Holstein calves milk replacers with 16.1, 18.5, 22.9, 25.8% CP from whey protein. As the CP increased in the diet, body weight gain, gain:feed ratio, absorbed N, and retained N all increased linearly. More information on nutrient partitioning and excretion and effects on mammary tissue proliferation is needed. Approximately 10 million dairy calves are born in the U. S. each year (Davis and Drackely, 1998) and with the growth of the dairy heifer growers industry, increasing animal density on farms, and new CAFO regulations, data is needed to determine the effects of these changes in calf diets on N and P excretion from

the animal. Also how changes in protein and energy content of these milk replacers influence body composition and type of growth these calves are undergoing needs to be evaluated.

The data from these studies indicate the potential for altering the protein and energy content of the milk replacer to reduce N excretion, improve lean growth, an influence mammary development. Our objective was to examine the impact on nutrient utilization and excretion when both protein and fat were fed at high rates and when milk replacer was fed at 2 and 3% of BW.

#### **Materials and Methods**

#### Animals and diets

Twenty-four newborn Holstein heifer calves (n=6) were fed one of four treatment diets (Table 3-1). Calves were purchased from a commercial dairy at  $2 \pm 2d$  of age and transported to the research center. Research calves were isolated from other calves at birth and were fed 3.4 L of colostrum once a day for two days after birth. Upon arrival at the research center, calves were fed a 24/17 milk replacer for the first two feedings. On d 3, they began receiving their treatment diets (Table 3-1). Heifers were grouped according to age, and treatments were assigned randomly within each group. Each group of heifers remained on study for approximately 63 d. Total collection of feed refusals, feces and urine were conducted for 4 d beginning on d  $59 \pm 2d$ .

Treatment milk replacers were designed to compare two isonitrogenous diets fed with either 17 or 24% fat, and to provide a standard diet as control and an extreme diet to evaluate the effects of aggressive growth promotion. The treatment diets were as follows: the control (24/17; fed at 350 g/d, 24% CP, 0.53% P); a high protein/low fat (32/17 fed at 764 g/d, 32% CP, 0.55% P); a high protein/high fat (31/24 fed at 782 g/d, 32% CP, 0.46% P); and the same milk replacer fed at a higher rate (31/24+ fed at 1177 g/d, 32% CP, 0.46% P). The 31/24+ milk replacer was

mixed at an increased percent solids to increase milk replacer intake. Calf starter was comprised of corn (44.4%), soybean meal (44.4%), cottonseed hulls (11.1%), and molasses (1.0%) and fed free choice to all calves, to prevent behavioral problems and allow normal gut development.

### Harvest Procedure

All calves were harvested at 63 d to evaluate body composition, gut development, and mammary development. Calves were fasted for 12 h before harvest and were weighed and transported to the necropsy lab at the Virginia Maryland Regional College of Veterinary Medicine for processing the morning before harvest. Calves were euthanized by phenobarbitol injection (Euthasol, 10 mg/kg BW) and immediately exsanguinated. Three components were collected: blood and organs (BO); head, hide, feet and tail (HHFT); and half of the carcass (HC). After exsanguination, blood was collected and the HHFT were removed and contained separately. All internal organs were removed and combined with the blood. Separate weights were collected for whole gut (full and empty); rumen (separate from other compartments; full and empty); liver; ovary; kidney; mammary tissue; heart/lungs; and spleen. A total weight was recorded for all blood and organs combined. The carcass was split and the weight of both sides was recorded and the left side was retained for analysis. All the components were refrigerated immediately and then frozen for later analysis. The components were transported to the abattoir at the USDA Meats Lab, (Beltsville, MD) and were ground through Autio Gear Head Grinder (Model 801GH, Astoria, OR). After assuming that the HHFT fraction would be difficult to grind alone, the frozen samples were cut into smaller pieces using a band saw and combined with the BO fraction. This also avoided excess loss of BO through thawing during the grinding process. These two fractions were later analyzed as one component (HHFT/BO). Components were ground three times and samples were taken and frozen. Further subsamples (100 g) were taken

of each component (HC and HHFT/BO) and were then freeze dried (FreeZone Plus, Freeze Dry Systems, Labconco Corp, Kansas City, MO) and ground through a 2-mm screen in a Wiley Mill (Arthur H. Thomas, Philadelphia, PA).

# Sample collection and analysis

Feces and urine samples were pooled (25% of each daily sample) by calf across the collection period, and analyzed for total Kjeldahl N (TKN) and total P (AOAC, 1984). Samples of milk replacers, calf starter, and body tissue (HHFT/BO and HC) were collected and analyzed for DM, TKN, total P (AOAC, 1984), total fat by supercritical fluid extraction (TFE2000 LECO Fat Extractor, St. Joseph, MI) and gross energy by bomb calorimetry (Parr 1271 Automatic Bomb Calorimeter, Parr Instrument Company, Moline, IL).

### Calculation of Gain

For economic reasons, the experimental design did not include a group of calves harvested at birth for baseline composition. Others (Diaz et al. 2001; Tikofsky et al. 2001, Blome et al. 2003; and Bartlett et al. 2006) have published the nutrient composition of their newborn calves. As our baseline, we used the baseline body composition data from Bartlett et al. (2006) due to similarity of study design and recentness of publication. The assumption made is that Holstein bulls and heifers at that young age (< 10 d) do not vary greatly in body composition. Therefore, body and component gains in this study were calculated as the (final percents of water, protein, fat and ash times the final empty body weight (EBW) from calves harvested in this study) minus (initial percent water, protein, fat, and ash from Bartlett et al. (2006) times the initial EBW of calves on this study (0.95 x initial live body weight)).

No published data on the P content of newborn calves was found, so P gain was calculated as final P content times total amount of EBW gain.

## Statistical Analysis

With the exception of growth and intake data, all data were analyzed using PROC GLM of SAS. Growth and intake data were analyzed using the RANDOM and REPEATED statements of the PROC MIXED procedure of SAS. Effects of treatment, group, heifer and the interaction of treatment and group were represented by the model:

$$Y=\mu+T_i+G_j+H_{k(ij)}+TG_{(ij)}+E_{ijk}$$

Where:

 $\mu$  = overall population mean;

 $T_i$  = effect of the i<sup>th</sup> diet (i = 1, 2, 3, 4);

 $G_i$  = effect of  $j^{th}$  group (j = 1, 2, 3);

 $H_k = \text{effect of } k^{\text{th}} \text{ heifer } (k = 1, 2, 3...6) \text{ (random)};$ 

 $TG_{(ij)}$  = interaction of diet and group; and

 $E_{ijk}$  = residual error term (random)

Preplanned contrasts were used to compare 24/17 to all other treatments, 32/17 to 31/24, and 31/24 to 31/24+. Data are reported as least squares means and differences were declared significant at P < 0.05.

#### **RESULTS AND DISCUSSION**

## Digestibility Trial

Nutrient Composition, Dry Matter Intake, and Manure Excretion. Milk replacer and starter composition are listed in Table 1. Calves fed the 24/17 (control) milk replacer consumed the most starter compared to the average of all other treatments (Table 3-2). However total DMI was not different across treatments because control calves were offered less milk replacer than others. Calves fed the 24/17 milk replacer had the lowest DMD compared to the other

treatments. This is likely due to the higher starter consumption in calves fed that treatment ration. While fecal excretion was the highest (kg DM/d) in calves fed 24/17, urinary output was the lowest (Table 3-4). Given the lower digestibility due to increased starter intake greater fecal excretion and lower urinary excretion would be expected.

Starter DM and nutrient intake did not differ with the addition of fat to the high protein milk replacer (32/17 vs. 31/24; Table 3-2) which is contradictory to results of Jaster et al. (1992) when calves were fed either an addition 226 g/d of fat or increased milk solids (15% DM) at 14% of BW. In the current study, the amount of milk replacer fed each day was fixed for each diet and calves consumed all milk replacer within 30 minutes of feeding. Therefore no variation could be detected in milk replacer intake and no probability of error calculated. Dry matter digestibility was not different between calves fed 32/17 and 31/24. There was a trend (P < 0.06) for increased fecal excretion (kg DM/d) when fat content of the milk replacer was increased. Urine excretion (kg/d) was higher in calves fed 31/24 compared to those fed 32/17 (2.70 vs. 1.48)

Starter intake did not change with the increase in volume of 31/24 milk replacer fed to calves (31/24 vs. 31/24+; Table 3-2). Dry matter digestibility did not change with the increase in volume of milk replacer fed (Table 3-2). Increasing the volume of milk fed did not change fecal DM output, but urine excretion was higher in calves fed the 31/24+ milk replacer compared to those fed the lower volume (Table 3-4).

*Growth and Health Measures.* Growth measures and scour scores can be found in Table 3-3. Overall body weight (BW), wither height (WH), body length (BL), hip width (HW), and hip height (HH) were lowest in calves fed 24/17 and highest in calves fed 31/24+. Adding fat (32/17 vs. 31/24) to the milk replacer did not change BW, WH, HW, or HH. Body length was greater in

calves fed 32/17. Feeding increased CP in the milk replacer increased the change in BW, BL, and HH except when fat was added to the diet and then body measures decreased.

Average daily gain increased with the addition of CP and fat, and with increased feeding rate. Calculated feed to gain ratio for calves fed the control diet was 4.5, which is much higher than those of calves fed the other treatments.

Respiratory and scour scores were measured daily in calves before feeding by the feeder. Temperatures were taken for 7 d after arrival to the research center and were not analyzed because of so few observations. Respiratory scores were not different among treatments and are not reported. Scour scores were not different among treatments as well (Table 3-3). One calf fed 31/24+ died at 6 wks of age (data removed from analysis) from abomasal ulcer caused by *C. perfringes*, which may have been proliferated by the extra nutrient supply. Just before harvest, two other calves on the same treatment showed similar symptoms, were treated with intravenous fluids and antibiotics, and survived until harvest.

*Nitrogen Intake and Partitioning.* As designed, milk replacer N intake was lowest in calves fed 24/17, averaged 39.3 g/d in calves fed 32/17 and 31/24, and was highest (150%) in calves fed 31/24+. Starter supplied 29 g/d of N to calves fed the 24/17 milk replacer and that was significantly higher than all the other treatments. Total N intake, however was lowest in calves fed 24/17 compared to the other treatments, because those calves were fed the milk replacer lowest in N content. Nitrogen digestibility was lowest in calves fed 24/17. There was a trend for higher fecal N excretion compared to the other treatments (15.8 vs. 12.1, P < 0.07) and urinary N excretion was lower than all the other treatments ( Table 3-4).

As a percent of N intake, calves fed 24/17 milk replacer had the highest fecal and urinary N excretion. Total N excretion (g/d) was not different, but these calves retained the least amount

of N (g/d; Table 3-4). Increasing N supplied from milk replacer increased digestibility, however increasing N supplied from dry feed decreased digestibility and increased N excretion. Blome et al. (2003) reported a significant quadratic effect of dietary CP on fecal N excretion. Fecal N excretion increased with N intake until milk replacer CP reached 25%; then fecal N declined (Blome et al., 2003). Our results contradict Blome et al. (2003) as this data set shows that fecal N was higher when less CP was fed in the milk replacer. The contradiction is due to our ad libitum feeding of calf starter. It is difficult to separate the effects of starter intake from those of milk replacer intake. Given that calves consuming the lowest amount of CP in the milk replacer were consuming the greatest amount of starter, independent effects of protein content of the milk replacer may not be evident. Our results reflect the integrated responses of claves under likely commercial conditions.

Increasing fat content of the high protein milk replacer did not have any effects on N intake, digestibility, or fecal and urinary excretion. Contrasting results were reported by Bascom (2002) in a comparison of 21/21, 27/31, 29/16 (% CP/ % fat) milk replacers and whole milk in Jersey calves. In that study, adding fat to the milk replacer increased N retention (%) with increasing fat; we did not observe this result.

More N was supplied to calves fed the 31/24+ through milk replacer than those fed 31/24, which was the goal of the treatment design (Table 3-4). Nitrogen digestibility was higher in calves fed 31/24+, which led to no differences in fecal N excretion between calves fed 31/24 and 31/24+. Urinary N was higher in calves fed a higher volume of milk replacer on a g/d basis, but not as a percent of intake. Calves fed the 31/24+ treatment were consuming almost 15 g/d more N than those fed 31/24+, but they were only retaining about 3 g/d more N with excess N being partitioned to urine (Table 3-4). Nitrogen excreted in urine is typically more volatile than that

excreted in feces (James et al., 1999) so having a greater amount of N being excreted in urine rather than being retained or excreted in feces could have negative implications for air and odor quality.

**Phosphorus intake and partitioning.** Phosphorus intake, digestibility, and excretion were not different in calves fed the control diet compared to calves fed more nutrient dense diets (Table 3-5). These measures were similarly unaffected by adding fat to the high protein diet. Phosphorus intake, digestibility, and fecal excretion were not different when the volume of 31/24 milk replacer was increased, but there was a trend for higher urinary P excretion with increasing volume (Table 3-5). It is important to note that these young calves were excreting nearly as much urinary P as do lactating cows. It is possible that urinary P was elevated due to contamination of urine with blood and other cells as an immune response to catheterization. Phosphorus digestibility varied greatly among treatments (SEm = 13.9%) and it is possible that intake P was underestimated. These calves were tethered individually in hutches, but still had contact with each other and were bedded on gravel to prevent intake of fibrous material such as straw. At harvesting, several hairballs and rocks were noted in the rumens of these calves indicating that intake of physical matter such as rocks, soil, and scurf may have led to an underestimation of P intake and highly variable P digestibilities. There are no other published data on P digestibility, excretion or retention in pre-weaned calves.

# **Body Composition**

Component and Empty Body Weights. Empty body weight (EBW) and weights of the body components are in Table 3-6. Components (kg and % EBW) do not add back to the whole of the EBW and the authors assume some loss of sample during processing. Calves that were fed the 24/17 milk replacer had lower final EBW (kg) compared to those fed the other milk replacers

(55.0 vs. 73.9) There was no difference in final EBW when more fat was added to the milk replacer, but calves fed the increased volume of 31/24 had greater EBW compared to those fed lower amounts of 31/24. Other authors (Blome et al. 2003; Bartlett et al. 2006) have reported lower EBW compared to the current data but calves in those studies were harvested at an earlier age. Diaz et al. (2001) and Tikofsky et al. (2001) both report similar EBW in calves of similar age to calves in this study. The viscera-free carcass comprised more than half of the EBW which was also noted by Bartlett et al. (2006). Empty body weight gain in this study was higher than typically noted by others (Diaz et al. 2001; Blome et al. 2003; Bartlett et al. 2006) and increased as CP percent of the milk replacer increased, but decreased with the addition of fat (Table 3-6). Water content decreased as calves grew which is similar to results of Bartlett et al. (2006).

### **Chemical Composition**

*Nitrogen.* Calves fed 32/17 had greater total body protein compared to those fed 31/24, although total N retention (from digestibility study) was not different between the two treatments indicating that the addition of fat to the diet did not improve N utilization. Differences in body protein and N retention are likely due to differences in the measurement period. Body protein changes reflect diet effects accrued across the 63 day study, while N retention results reflect changes in the final 4 days of the study. Diaz et al. (2001) noted that when N retention was calculated as a result from a digestibility trail versus protein content at harvest, N retained was overestimated by 27.5% using digestibility data. The current digestibility study only overestimated N retention by 8% when compared with data from the harvested trial. Possible losses of N in a digestibility trial include N loss in scurf, N loss due to splashing or evaporation, and due to loss of volatile N compounds in a drying oven (Blome et al. 2003). In the current

study, urinary catheters were placed in the heifers which likely prevented loss of N due to splashing or evaporation and decreased the error in N retention values.

Calves fed the 31/24+ had consistently higher body protein content, however given in the difference in N intake (14 g/d) and the increased cost of intensified milk replacer, the increase in body N (1.2 g/d) is probably not economically meaningful. Total body protein (kg) is similar to the results of Bartlett et al. (2006) and the protein content of components (HHFT/BO and HC) are similar to results found by Blome et al. (2003). In both studies, whole body protein content was slightly lower than in the current study, but calves in the current study were larger and the milk replacers were higher in CP.

Protein gain was lowest in calves fed 24/17 and greatest in calves fed 32/17 (Table 3-6). Calves fed 31/24 gained only half the protein of those calves fed 32/17 and 31/24+. Tikofsky et al. (2001) noted that when calves were fed a high fat diet compared to a low fat (30.6 vs. 23.5%), they used ME from fat calories for protein deposition as opposed to using calories from carbohydrates (lactose) for protein deposition. The latter is more energetically expensive. In the current study, the addition of fat to the diet (31/24) did not improve lean tissue accretion and in fact calves fed the 32/17 milk replacer had more than two times the protein gain of those fed 31/24.

*Phosphorus and Ash.* There were no treatment effects on body P content, but P gain was lowest in calves fed 24/17 compared to other treatments and higher in calves fed 31/24+ simply because those calves gained the least and the most BW, respectively (Table 3-6). There are no other reported values of P content of the total calf. Differences noted here are likely due to changes in frame sizes and more bone growth.

Total body ash content was lower in calves fed 24/17 compared to the average of the other treatments (Table 3-6). Calves fed 32/17 had the highest total body ash followed by those fed the 31/24+. Calves fed the 31/24 had the lowest total body ash. Ash gain (kg) was higher in calves fed 32/17 but as a % of EBW, ash gain was highest in calves fed 24/17. Increases in ash gain indicate an increase in bone (frame) growth and calves fed 24/17 had the least amount of EBW gain. The gain of these calves was mostly frame growth and not protein or fat gain which increased ash content. Ash gain was also higher in calves fed 32/17, but so was protein gain implying that calves fed 32/17 had greater tissue accretion as well as frame growth.

Fat and Energy. Fat content was lowest in the calves fed 24/17. Calves fed 32/17 and 31/24 did not differ in total fat content (kg), but because calves fed 31/24 gained less EBW fat, as a percent of EBW, was higher than in calves fed 32/17 (Table 3-6). Calves fed 32/17 had approximately 5% more total fat than those fed 24/17 indicating that increasing the protein in the milk replacer and keeping fat constant and increasing the amount fed increased the amount of fat deposited in the body. Both Bartlett et al. (2006) and Blome et al. (2003) reported a decrease in fat as protein (% of EBW) increased when calves were fed a low fat diet. In that study, there was no noted effect of dietary CP on fat content when calves were fed a high fat diet. Tikofsky et al. (2001) held CP percent constant (~13% CP) and increased fat in the milk replacer. They reported results of increased fat content with increasing fat in the milk replacer, which is similar to the current study. Also, total fat gain (kg and % of EBW gain) was lowest in calves fed 24/17 and was not different in calves fed 32/17 compared to 31/24 (kg) (Table 3-6). Calves fed 31/24+ had the greatest amount of fat gain. Again, the results of Blome et al. (2003) contradict these data where they report a decrease in fat gain as dietary CP increased from 16.1 to 25.8%, we report an increase in fat gain when milk replacer CP was greater than 23%. One likely reason for

this is that Blome et al. (2003) held milk replacer intake constant while it was varied across treatments in the current study.

Energy content was lowest in calves fed 24/17 milk replacer (Table 3-6). There was no difference in body energy content (Mcal) between calves fed 32/17 and 31/24. This could be expected given the similarities in fat content (kg) between the two diets. Some energy can be released from protein metabolism, but 210 g of protein are required to obtain 700 g of gain (Davis and Drackley, 1998) so protein is a very small source of fuel.

Energy gain was lowest in calves fed 24/17 compared to those fed the other treatments and highest in those fed 31/24+ (Table 3-6). Adding fat to the milk replacer did not change energy content (Mcal; 32/17 vs. 31/24). Energy per kg of EBW was lowest in calves fed 24/17 and highest in calves fed 31/24+. Calves fed 32/17 had less energy per kg of EBW, but their fat content (% of EBW) was lower compared to calves fed 31/24. Energy gain per kg of EBW gain is similar to energy per unit of EBW, implying that composition of gain was not different from composition of final empty body weight. Blome et al. (2003) and Bartlett et al. (2006) noted a decrease in energy gain (Mcal/kg) as CP (% DM) increased. In the current study, as dietary CP and fat increased so did energy content of gain. Increasing the amount of milk replacer offered increased energy gain (Table 3-6). Similarly, Bartlett et al. (2006) also reported an increase in energy gain with a higher feeding rate. Tikofsky et al. (2001) reported an increase in fat (Mcal per kg EBW gain) with an increased fat (% DM) in the milk replacer.

### **Conclusions and Implications**

Due in changes in CAFO regulations, it is necessary to begin to collect more data on P excretion in heifers. Our data suggests that it is time to reevaluate P requirements in heifers

since these small calves were excreting nearly all the P they were consuming. As Nennich et al. (2005) observed there is a lack of data concerning nutrient excretion in this age group of calves.

Feeding fat above 17% lead to higher fat deposition which is important in growing heifers, but too much accretion of fat can lead to reproductive and health problems. Feeding protein above 25% increased protein gain. Others have noted that increasing fat only increased fat gain when protein intake was limiting (Tikofsky et al. 2001; Blome et al. 2003). In the diets described above, when protein was fed past 25% of the DM and fat content was increased, only fat gain increased, but when both protein and fat were increased protein gain increased indicating that protein intake may drive protein gain more so than fat intake. Feeding a high protein, low fat milk replacer, like 32/17 fed in this study, increased tissue gain and frame growth (as indicated by increased ash content, kg) and may be an economically sound feed choice to achieve accelerated growth in replacement heifers. Perhaps feeding this milk replacer at a greater than 2% of BW would increase tissue gain and/or frame growth even more.

Feeding the 31/24 did not produce any beneficial results except for increased fat gain, which could be important in preventing health problems when calves are raised outside their thermoneutral zone. Feeding 31/24 at the higher rate (1177 g/d powder) increased protein and fat gain, but not beyond that of calves fed the 32/17 and extra nutrients were lost to the environment. The increased cost of a milk replacer similar to 31/24+ would most likely not be recovered in increased gain. Moreover, the death of one calf and poor health of two others were caused by abomasal ulcers, triggered by over-growth of *C. perfringes*. The added nutrients supplied to these common environmental microbes from the 31/24+ milk replacer may have aggravated the condition. Therefore, implications from this study are that a milk replacer that is

25-32% CP and not above 20% fat could produce ideal gains in young Holstein calves while still helping meet CAFO regulations.

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Table 3-1. Ingredient and nutrient composition of milk replacers varying in protein and fat fed to Holstein calves.

	Dietary Treatments									
	Milk Replacer									
	24/17 <sup>1</sup>	$32/17^2$	31/24 <sup>3</sup>	31/24+4						
CP, % DM	23.6	32.3	31.2	31.2						
Fat, % DM	17.1	16.5	23.9	23.9						
P, % DM	0.53	0.55	0.46	0.46						
Powder, g/d	350	764	782	1177						
		Starter Ingred	dients,% diet							
Corn grain, ground	44.4									
Soybean Meal	44.4									
Cottonseed Hulls	11.1									
Dried Molasses	1.0									
		Starter Compo	osition,% DM							
CP,%	20.3	•								
Fat,%	NA									
P,%	0.48									

<sup>&</sup>lt;sup>1</sup> 24/17= milk replacer with 24% CP and 17% fat
<sup>2</sup> 32/17= milk replacer with 32% CP and 17% fat
<sup>3</sup> 31/24= milk replacer with 31% CP and 24% fat
<sup>4</sup> 31/24+= milk replacer with 31% CP and 24% fat fed at 1177g/d

Table 3-2. Nutrient intake and dry matter digestibility of milk replacers varying in protein and fat fed to Holstein calves.

	]	Dietary T	P <						
	24/17 <sup>1</sup>	$32/17^2$	31/24 <sup>3</sup>	$31/24+^4$	SEm <sup>5</sup>	Treatment	24/17 vs. all	32/17 vs. 31/24	31/24 vs. 31/24+
Starter									
DM, kg/d	0.89	0.59	0.40	0.32	0.15	0.08	0.02	0.40	0.71
N, g/d	29.0	17.9	16.0	10.7	4.40	0.08	0.02	0.79	0.41
P, g/d	4.30	3.09	2.37	1.58	0.59	0.04	0.02	0.44	0.37
Energy, Mcal	3.15	2.10	1.41	1.14	0.59	0.08	0.02	0.41	0.72
Milk Replacer <sup>6</sup>									
DM, g/d	350	764	782	1177					
N, g/d	13.2	39.5	39.0	58.7					
P, g/d	1.88	4.20	3.60	5.41					
Energy, Mcal	1.64	3.74	4.10	6.17					
Total DMI, kg/d	1.25	1.31	1.18	1.50	1.6	0.56	0.65	0.60	0.78
DMD,%	53.4	70.3	73.5	81.3	5.3	0.02	0.01	0.70	0.32
Total Energy, Mcal	4.79	5.84	5.51	7.31	0.59	0.03	0.04	0.69	0.03

<sup>&</sup>lt;sup>1</sup> 24/17= milk replacer with 24% CP and 17% fat <sup>2</sup> 32/17= milk replacer with 32% CP and 17% fat <sup>3</sup> 31/24= milk replacer with 31% CP and 24% fat <sup>4</sup> 31/24+= milk replacer with 31% CP and 24% fat fed at 1177g/d

 $<sup>^{5}</sup>$  n = 23

<sup>&</sup>lt;sup>6</sup>No statistical analysis was completed because within treatment, equal amounts were fed daily and probability of difference could not be calculated.

Table 3-3. Growth measures in Holstein calves fed milk replacers varying in protein and fat.

	Die	etary Trea	P <						
	$24/17^{1}$	$32/17^2$	$31/24^3$	$31/24+^4$	SEm <sup>5</sup>	Treatment	24/17 vs. all	32/17 vs. 31/24	31/24 vs. 31/24+
Body weight, kg <sup>6</sup>	51.3	64.6	58.9	67.5	2.80	0.01	0.01	0.21	0.03
Wither height, cm <sup>6</sup>	81.0	83.3	84.1	84.1	1.00	0.09	0.02	0.42	0.98
Body length, cm <sup>6</sup>	77.7	83.2	78.9	81.5	1.20	0.04	0.02	0.06	0.16
Hip width, cm <sup>6</sup>	22.4	24.5	23.4	24.5	0.61	0.12	0.04	0.21	0.24
Hip Height, cm <sup>6</sup>	85.6	86.3	86.9	87.2	2.10	0.94	0.65	0.84	0.91
$\Delta$ Body weight, kg <sup>7</sup>	26.3	47.1	36.2	51.2	3.60	0.01	0.01	0.06	0.01
$\Delta$ Wither height, cm <sup>7</sup>	10.4	17.2	14.9	15.8	1.60	0.01	0.01	0.27	0.61
$\Delta$ Body length, cm <sup>7</sup>	15.5	20.5	23.6	23.2	2.53	0.08	0.02	0.41	0.89
$\Delta$ Hip width, cm <sup>7</sup>	5.23	9.07	7.24	8.29	1.38	0.19	0.04	0.35	0.52
ΔHip height, cm <sup>7</sup>	9.90	15.3	13.3	11.8	2.65	0.47	0.20	0.60	0.64
Average daily gain, kg/d	0.41	0.68	0.63	0.82	0.05	0.01	0.01	0.57	0.03
Feed to gain	4.54	2.56	2.16	2.10	0.97	0.18	0.03	0.94	0.96
Scour score <sup>8</sup>	1.70	1.60	1.80	2.10	0.14	0.09	0.50	0.18	0.18

<sup>&</sup>lt;sup>1</sup> 24/17= milk replacer with 24% CP and 17% fat
<sup>2</sup> 32/17= milk replacer with 32% CP and 17% fat
<sup>3</sup> 31/24= milk replacer with 31% CP and 24% fat
<sup>4</sup> 31/24+= milk replacer with 31% CP and 24% fat fed at 1177g/d

 $<sup>^{5}</sup>$  n = 23

<sup>&</sup>lt;sup>6</sup>Averages across entire 63 d trial analyzed with initial values as covariate.

<sup>&</sup>lt;sup>7</sup>Change in growth calculated by final-initial.

<sup>&</sup>lt;sup>8</sup>Fecal Score Scale: 1= Normal (soft, solid, no fluid), 2= Soft (semi-solid), 3= Runny (semi-solid, mostly fluid), 4= Water (fluid), 5= Bloody

Table 3-4. Manure excretion and nitrogen metabolism in Holstein calves fed milk replacers varying in protein and fat.

			Dietary	Treatmen	ts		P <		
	24/17 <sup>1</sup>	$32/17^2$	$31/24^3$	31/24+4	SEm <sup>5</sup>	Treatment	24/17 vs. all	32/17 vs. 31/24	31/24 vs. 31/24+
Feces, kg DM/d	0.52	0.42	0.30	0.28	0.34	0.01	0.01	0.06	0.79
Urine, kg/d	1.34	1.48	2.70	4.38	0.35	0.01	0.01	0.04	0.01
N intake, g/d	42.3	57.5	55.0	69.4	4.4	0.01	0.01	0.74	0.04
N digestibility,%	63.0	79.8	78.4	80.2	4.5	0.04	0.01	0.83	0.02
Fecal N									
g/d	15.8	11.0	11.7	13.7	1.56	0.20	0.07	0.74	0.37
% of N intake	36.9	18.4	21.6	19.8	2.27	0.01	0.01	0.38	0.59
Urine N									
g/d	6.04	8.22	11.7	21.2	2.2	0.01	0.01	0.33	0.01
% of N intake	19.7	14.9	20.4	31.0	6.5	0.44	0.76	0.60	0.27
Total N excretion									
g/d	21.8	19.3	23.4	39.9	2.7	0.01	0.24	0.33	0.01
% of N intake	56.7	33.6	41.9	50.8	7.9	0.28	0.14	0.49	0.44
N retention									
g/d	20.5	38.2	31.6	34.5	4.8	0.12	0.02	0.37	0.64
% intake	43.3	66.4	58.0	49.2	7.9	0.28	0.14	0.49	0.44

<sup>1 24/17=</sup> milk replacer with 24% CP and 17% fat
2 32/17= milk replacer with 32% CP and 17% fat
3 31/24= milk replacer with 31% CP and 24% fat
4 31/24+= milk replacer with 31% CP and 24% fat fed at 1177g/d

 $<sup>^{5}</sup>$  n = 23

Table 3-5. Phosphorus metabolism in Holstein calves fed milk replacers varying in protein and fat.

		Dieta	ary Treat	ments			P <				
	24/17 <sup>1</sup>	$32/17^2$	31/24 <sup>3</sup>	31/24+4	SEm <sup>5</sup>	Treatment	24/17 vs. all	32/17 vs. 31/24	31/24 vs. 31/24+		
P intake, g/d	6.16	7.29	5.97	7.00	0.59	0.41	0.41	0.17	0.24		
P digestibility,%	32.7	71.8	59.1	46.6	13.9	0.33	0.13	0.56	0.54		
Fecal P											
g/d	3.99	2.13	2.29	3.56	7.72	0.32	0.17	0.89	0.27		
% intake	67.3	28.1	40.9	53.4	13.9	0.33	0.13	0.56	0.54		
Urine P											
g/d	1.31	1.12	1.05	1.98	0.31	0.19	0.85	0.89	0.06		
% intake	28.6	16.1	18.3	29.3	8.63	0.64	0.48	0.87	0.38		
Total P excretion, g/d											
g/d	5.31	3.25	3.35	5.54	0.90	0.22	0.26	0.95	0.11		
% intake	95.9	44.2	59.2	82.8	18.6	0.30	0.15	0.61	0.39		
P retention											
g/d	0.84	4.06	2.62	1.46	1.27	0.41	0.24	0.48	0.53		
% intake <sup>6</sup>	13.6	55.7	43.9	20.9							
Composition											
Total P, kg	1.01	1.06	0.83	1.23	0.14	0.14	0.82	0.22	0.03		
P gain, kg	0.55	0.60	0.44	0.72	0.08	0.02	0.65	0.13	0.01		
P, % of EBW	1.86	1.44	1.23	1.55	0.19	0.12	0.04	0.43	0.21		

<sup>1 24/17=</sup> milk replacer with 24% CP and 17% fat
2 32/17= milk replacer with 32% CP and 17% fat
3 31/24= milk replacer with 31% CP and 24% fat
4 31/24+= milk replacer with 31% CP and 24% fat fed at 1177g/d

 $<sup>^{5}</sup>$  n = 23

<sup>&</sup>lt;sup>6</sup> Phosphorus retained as a percent of intake was calculated as a ratio of least squares means (retained/intake)\*100. Statistical analysis was not done because of negative retention values.

Table 3-6. Composition of body components in Holstein calves fed milk replacers varying in protein and fat.

	]	Dietary T	reatment			P <				
	$24/17^{1}$	$32/17^2$	$31/24^3$	$31/24^4$	SEm <sup>5</sup>	Trt	24/17 vs. all	32/17 vs. 31/24	31/24 vs. 31/24	
SBW, kg wet	79.1	85.1	77.4	79.1	7.38	0.88	0.85	0.45	0.85	
EBW, kg DM	41.1	53.5	46.7	58.6	2.56	0.01	0.01	0.07	0.01	
CP, kg DM	25.3	32.4	26.2	31.7	1.45	0.01	0.01	0.01	0.01	
Fat, kg DM	6.89	10.3	11.2	16.0	0.85	0.01	0.01	0.48	0.01	
Ash, kg DM	6.95	8.32	6.51	7.45	0.44	0.05	0.32	0.01	0.11	
Component, % of EBW										
CP, %	61.8	60.6	56.0	54.2	0.81	0.01	0.01	0.01	0.10	
Fat, %	16.7	19.3	23.8	27.2	0.90	0.01	0.01	0.01	0.01	
Ash, %	16.8	15.6	13.9	12.8	0.56	0.01	0.01	0.06	0.11	
EBW gain, kg DM	31.1	42.3	37.9	48.9	2.56	0.01	0.01	0.22	0.01	
CP gain, kg DM	18.2	24.5	19.8	24.7	1.44	0.01	0.01	0.03	0.02	
Fat gain, kg DM	5.44	8.74	9.88	14.6	0.85	0.01	0.01	0.33	0.01	
Ash gain, kg DM	5.51	6.72	5.22	6.04	0.44	0.08	0.27	0.02	0.14	
Component, % of EBW										
CP, %	58.6	57.8	52.4	50.8	0.99	0.01	0.01	0.01	0.22	
Fat, %	17.5	20.6	26.0	29.6	1.12	0.01	0.01	0.01	0.02	
Ash, %	17.7	15.9	13.9	12.4	0.70	0.01	0.01	0.06	0.13	

<sup>1 24/17=</sup> milk replacer with 24% CP and 17% fat
2 32/17= milk replacer with 32% CP and 17% fat
3 31/24= milk replacer with 31% CP and 24% fat
4 31/24+= milk replacer with 31% CP and 24% fat fed at 1177g/d
5 n = 23

Table 3.6 (cont.) Composition of body components in Holstein calves fed milk replacers varying in protein and fat.

	I	Dietary T	reatment	S	·		<i>P</i> <			
	$24/17^{1}$	$32/17^2$	$31/24^3$	31/244	SEm <sup>5</sup>	Trt	24/17 vs. all	32/17 vs. 31/24	31/24 vs. 31/24	
Energy										
Mcal	200	271	250	329	14.6	0.01	0.01	0.32	0.01	
Mcal/ kg EBW	4.86	5.05	5.36	5.59	0.10	0.01	0.01	0.04	0.08	
Mcal gain/ Mcal EBW gain	4.80	5.05	5.47	5.73	0.13	0.01	0.01	0.04	0.08	

<sup>&</sup>lt;sup>1</sup> 24/17= milk replacer with 24% CP and 17% fat
<sup>2</sup> 32/17= milk replacer with 32% CP and 17% fat
<sup>3</sup> 31/24= milk replacer with 31% CP and 24% fat
<sup>4</sup> 31/24+= milk replacer with 31% CP and 24% fat fed at 1177g/d
<sup>5</sup> n = 23

### **CHAPTER 4 CONCLUSIONS**

Due to in changes in CAFO regulations, it is necessary to begin to collect more data on N and P excretion in heifers. What little published data there is on P excretion, suggests that it is time to re-evaluate P requirements in heifers since these small calves and bred heifers were excreting nearly all the P they were consuming. As Nennich et al. (2005) observed there is a lack of data concerning nutrient excretion in this age group of animals.

Growth was similar in bred heifers fed high and low forage diets and did not change with the addition of by-products. Total nutrient excretion did not differ for heifers fed ad libitum high forage diets compared with limit-fed heifers, but the increase in urine output in heifers fed low forage diets would make manure management more difficult. Limit feeding heifers a low forage ration could be a concern because additional manure storage capacity would be required, especially in areas where nutrient and waste management are an immediate concern. There are three possible causes for increased urine output in the LF heifers. 1) increased salt or energy intake (Johnson and Combs, 1991) which could lead to increased water intake and increased water output (Murphy et al. 1992). In this study, heifers fed the HF ration actually consumed more salt than those fed the LF ration, however, increased energy intake could still be an issue 2) if heifers are eating to satiety triggered by gut fill, without the bulkiness that is typical in a HF ration, they may consume more water to achieve gut fill. And 3) heifers fed the LF ration consumed all their feed within an hour, typically, and given that they are curious animals may have consumed more water due to boredom. In the latter situation, limit feeding

heifers could possible lead to other non-nutritive oral behaviors such as pica and tongue rolling, etc.

Given the higher urinary volume and tendency to increase N excretion in urine, there is a greater potential for ammonia volatilization in heifers fed the LF ration. The LF ration mimics a typical feedlot ration although those animals may often be fed an even higher concentrate ration and be more restricted in intake indicating an even greater potential for N volatilization and contribution to poor odor and air quality.

Low to zero P digestibility and retention in heifers fed LF or HF and BP, respectively, indicates that heifers in late gestation are not retaining any P when over fed. Heifers excreted nearly all the P consumed and 95% confidence intervals revealed that these negative numbers are really not different from zero. Given that P requirements during pregnancy are about 5 g/d (for late pregnant, mature cow), and P requirements for growth are minimal for heifer of this age, over feeding heifers results in excretion of all or nearly all consumed P. The P content of these heifer rations was similar to the requirement of lactating cow and without the demand of milk production 0.3-0.4% P is excessive in a heifer ration.

The current ASAE standard equations for dairy heifers and beef cattle are poor predictors of nutrient and manure excretion in bred heifers. The one exception is the beef cattle manure prediction equation. Given the great variation in urine (water) excretion, the dairy equation was inadequate. Overall, heifers achieving similar rates of gain from diets differing in forage, grain and byproduct content excreted widely varying quantities of manure and these quantities were not well predicted by the ASAE Standard Equation (2005).

Feed is the most expensive cost on a dairy operation and costs of regular or accelerated milk replacers contribute to that cost. Paying for extra nutrients is redundant if they are not being used by the animal. In this study, calves responded positively to increasing protein content while maintaining a low energy content. Feeding fat above 17% lead to higher fat deposition which is important in growing heifers, but too much accretion of fat can lead to reproductive and health problems. Feeding protein above 25% increased protein gain. Others have noted that increasing fat only increased fat gain when protein intake was limiting (Tikofsky et al. 2001; Blome et al. 2003). In the diets described above, when protein was fed past 25% of the DM and fat content was increased, only fat gain increased, but when both protein and fat were increased protein gain increased indicating that protein intake may drive protein gain more so than fat intake. Feeding a high protein, low fat milk replacer, like 32/17 fed in this study, increased tissue gain and frame growth (as indicated by increased ash content, kg) and is an economically sound feed choice to achieve accelerated growth in replacement heifers. Perhaps feeding this milk replacer at a greater than 2% of BW would increase tissue gain and/or frame growth even more.

Feeding the 31/24 did not produce any beneficial results except for increased fat gain, which could be important in preventing health problems when calves are raised outside their thermoneutral zone. Feeding 31/24 at 1177 g/d powder increased protein and fat gain, but not beyond that of calves fed the 32/17 and extra nutrients were lost to the environment. The increased cost of a milk replacer similar to 31/24+ would most likely not be recovered in increased gain. Moreover, the death of one calf and poor health

of two others were caused by abomasal ulcers, which were proliferated by the overgrowth of C. *perfrenges*.

Calves fed 32/17 had approximately 5% more total fat than those fed 24/17 indicating that increasing the protein in the milk replacer and keeping fat constant increased the amount of fat but increasing the amount of fat and maintaining protein did not increase fat content. That said, fat (% of EBW) was higher in calves fed 31/24 compared to those fed 32/17 so increase in fat content between calves fed 24/17 and calves fed 32/17 is likely due to an overall increase in total gain and not necessarily to the increase in CP content of the milk replacer. The calves fed the 32/17 milk replacer retained more N and energy with less fat gain. Increased rate of gain in heifers is encouraged as long as the gain is not predominantly fat and, in this study, the 32/17 milk replacer encouraged gain without fattening. Therefore, implications from this study are that a milk replacer that is 25-32% CP and below 20% fat could produce ideal gains in young Holstein calves while still helping meet CAFO regulations.

Overall, the need to revisit nutrient (specifically N and P) requirements in heifers is evident. In both of these studies, heifers were overfed P and as a result, essentially all of the P consumed was excreted which has poor implications for nutrient management.

The concept of feeding required nutrients in less feed, i.e. a high concentrate diet, is typical in the feedlot industry and is becoming more popular with dairy heifer producers. Results of the bred heifer study indicate that these low forage rations may increase the potential for nutrient loss and could have negative implications for rumen and oral health.

Growing quality replacement heifers is integral to the dairy industry and that starts as young as one week of age. Increasing growth through feeding intensified milk replacers has positive benefits, however pushing nutrient intake too high will increase nutrient excretion and result in nutrient loss. Further research in the area of heifer growth will lead to more accurate nutrient requirements and better nutrient utilization.