

# DAIRY RESEARCH 2009

REPORT OF PROGRESS 1021



KANSAS STATE UNIVERSITY  
AGRICULTURAL EXPERIMENT  
STATION AND COOPERATIVE  
EXTENSION SERVICE



## Dairy Days Program

**February 4, 2010**

Reno County

**February 5, 2010**

Nemaha County

### ***Graduate Student Presentations***

Milk production and composition of cows fed wet brewers grains

*Christa Mahnken*

Determining the water needs of dairy farms

*Justin Potts*

Effect of acidulant addition on yogurt fermentation

*Tori Boomgaarden*

Effects of acidified fermentation by-products and prepartum DCAD on feed intake, performance, and health of transition dairy cows

*DJ Rezac*

Dietary molasses increases ruminal pH and enhances ruminal biohydrogenation during milk fat depression

*Cynthia Martel*

### ***Faculty Presentations***

Milk components and other processing issues

*Karen Schmidt*

Estrumate vs. Lutalyse?

*Jeff Stevenson*

Relationships between feed cost and milk production

*Mike Brouk*

Inflammation in transition cows

*Barry Bradford*

K-State Dairy Facility update

*John F. Smith*

# DAIRY RESEARCH 2009

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## Foreword

Members of the Dairy Team at Kansas State University are pleased to present the 2009 Dairy Research Report of Progress. Dairying continues to contribute significantly to the agricultural economy of Kansas. In 2007, dairy farms accounted for 2.9%, or \$296 million, of all Kansas farm receipts, ranking seventh overall among all Kansas farm commodities. In the United States, Kansas had the greatest percentage increase in milk produced between 1999 and 2004 (+57.7%). At the end of 2008, Kansas ranked 10th nationally in milk yield per cow at 20,641 lb, 18th in the number of dairy cows (117,000), and 17th in total milk production (2.425 billion lb). Wide variation exists in the productivity per cow as indicated by the Heart of America Dairy Herd Improvement Association (DHIA) production testing program. At the end of 2008, nearly 108,000 cows were enrolled in the DHI program from Kansas, Nebraska, Oklahoma, Arkansas, North Dakota, and South Dakota, including herds from Colorado (3), Missouri (10), Montana (10), and Texas (1). A comparison of Kansas DHIA cows with all those in the Heart of America DHIA program for 2008 is shown in the following table.

### **Comparison of Heart of America Cows with Kansas Cows - 2008**

| Item                       | Heart of America | Kansas |
|----------------------------|------------------|--------|
| Herds, no.                 | 576              | 194    |
| Cows/herd, no.             | 173              | 114    |
| Milk, lb                   | 20,717           | 20,744 |
| Fat, lb                    | 767              | 785    |
| Protein, lb                | 642              | 655    |
| Somatic cell count × 1,000 | 306              | 341    |
| Calving interval, mo       | 14.0             | 14.4   |

Most of this success occurs because dairy producers better manage what is measured in monthly DHI records. Continued emphasis should be placed on furthering the DHI program and encouraging use of its records in making management decisions. In addition, continued use of superior, proven sires and emphasis on use of superior genetics in artificial insemination programs is essential.

The excellent functioning of the Dairy Teaching and Research Center (DTRC) is due to the special dedication of our staff. We acknowledge our current DTRC staff for their dedication: Michael V. Scheffel (manager), John P. Dwyer, Daniel J. Umsheid, Alan J. Hubbard, and Kris Frey. Special thanks are given to Jamie Gardner, Colleen Hill, Cheryl K. Armendariz, and a host of graduate and undergraduate students for their technical assistance in our laboratories and at the DTRC.

Milk production from 253 cows at the DTRC continues to improve according to our last test day in September 2009. Our rolling herd average for milk surpassed 30,000 lb for the first time in August 2006. Now without bST use, but milking 3 times daily, our 365-day rolling herd averages for the September 2009 test day were 31,215 lb of milk, 1,051 lb of fat, and 964 lb of protein.

Thorough, quality research is not only time intensive and meticulous, but also expensive. Each dollar spent for research yields a 30 to 50% return in practical application. Those interested in

supporting dairy research are encouraged to consider participation in the Livestock and Meat Industry Council (LMIC), a philanthropic organization dedicated to furthering academic and research pursuits by the Department of Animal Sciences and Industry. Additional details about the LMIC are found at the end of this report.

J. S. Stevenson, Editor  
2009 Dairy Research Report of Progress

## Biological Variability and Chances of Error

Variability among individual animals in an experiment leads to problems in interpreting the results. Although the cattle on treatment X may have produced more milk than those on treatment Y, variability within treatments may indicate that the differences in production between X and Y were not the direct result of the treatment alone. Statistical analysis allows us to calculate the probability that such differences occur because of the treatment applied rather than from chance.

In some of the articles herein, you will see the notation  $P < 0.05$ . That means the probability of treatment differences resulting from chance is less than 5%. If two averages are reported to be significantly different, the probability is less than 5% that the difference is from chance, or the probability exceeds 95% that the difference resulted from the treatment applied.

Some papers report correlations or measures of the relationship between traits. The relationship may be positive (both traits tend to get larger or smaller together) or negative (as one trait gets larger, the other gets smaller). A perfect correlation is one (+1 or -1). If there is no relationship, the correlation is zero.

In other papers, you may see an average given as  $2.5 \pm 0.1$ . The 2.5 is the average; 0.1 is the standard error. The standard error is calculated to be 68% certain that the real average (with an unlimited number of animals) would fall within one standard error from the average, in this case between 2.4 and 2.6.

Using many animals per treatment, replicating treatments several times, and using uniform animals increases the probability of finding real differences when they exist. Statistical analysis allows more valid interpretation of the results, regardless of the number of animals in the experiment. In all the research reported herein, statistical analyses are included to increase the confidence you can place in the results.

# Differences Among High, Medium, and Low Profit Dairy Operations: An Analysis of 2004-2008 Kansas Farm Management Association Dairy Enterprises

*K. M. Schulte and K. C. Dhuyvetter*

## Summary

The financial bottom line, or net income, is a key factor in determining how successful a dairy has been historically as well as an indicator of the financial ease or struggles the dairy might have in the future. What causes net income to vary from one operation to another is a key question for dairy farmers. For example, does milk price received, feed cost, total cost, or milk production have the greatest impact on net return variability? In this study, we evaluated Kansas Farm Management Dairy Enterprise data from the past 5 years to determine correlation of revenue, production, and cost factors among groups of high, medium, and low profit dairy operations. High-profit producers had larger operations, had slightly greater total costs (\$62.63 per cow), and received slightly lower milk prices (\$0.56/100 lb of milk) compared with low-profit producers. In contrast, the high profit group produced significantly more milk per cow. Milk price received and cost per cow did not affect profit nearly as much as total milk produced per cow. This study was conducted with data reported by small to midsize dairy herds. Further research should examine whether these results hold true for large herds.

## Introduction

Profitability within the dairy industry has been in the spotlight during the past 5 years as a result of extreme volatility in the commodity markets that greatly affects not only income from milk sales but also feed costs, which represent a large percentage of total expenses. Recently, many producers have focused on marketing milk and feed to create set prices received and paid. In addition, because of the decline in the milk price during 2009, some producers cut feed costs to make up for loss in milk income and others focused on improving production to generate more revenue. Small to midsize producers have felt a major crunch in their cash flow and net income in the past few years. Net income largely determines how long a farm can survive. A dairy farmer who made little to no profit during 1 year with no capital saved will continue to struggle in future years. In contrast, producers who have capital saved from previous profitable years can survive longer during times of low milk prices and increased costs.

Dairy farmers exercise different management approaches to increase profit including: (1) minimizing costs to increase profit, (2) minimizing assets per production unit to reduce fixed costs, (3) marketing milk to receive the best milk price possible to increase revenue, and (4) increasing production to maximize revenue and profit. In theory, a skilled manager should be able to balance asset utilization, reduce cost, and maximize revenue to increase net income. Many managers, however, are unable to optimize each of these factors. The objective of this study was to determine which factors (receiving a higher milk price, reducing cost, or increasing milk production) a small to midsize dairy enterprise should focus on to increase net income.

## Experimental Procedures

Forty dairy farms from the Kansas Farm Management Association database were selected on the basis of their participation in reporting data during the past 5 years (2004 to 2008). To be

included in the analysis, a farm must have reported data a minimum of 3 of the 5 years. The average number of years farms reported data was 4.5 years (6 farms had 3 years, 11 had 4 years, and 23 had 5 years). The farms were sorted from high to low on average returns over total costs (i.e., profit) and then separated into the top, middle, and bottom third (13, 14, and 13 farms, respectively). The multiyear average number of cows per farm was 115 (range: 35 to 257 cows). The various cost categories were aggregated into 9 groups, and revenue was grouped into milk sales, net cattle sales, and other income. Other factors evaluated included pounds of milk per cow, culling rate, milk price per 100 lb of milk, and income over feed cost.

To normalize data, results are reported on a per-cow basis. There is debate within the dairy industry regarding whether data should be analyzed on a per-cow or per-hundredweight basis. The 40 farms were ranked from 1 to 40 (1 = highest profit, 40 = lowest profit) on the basis of both profit per cow and profit per 100 lb of milk. Figure 1 plots per-cow rankings on the vertical axis against per-hundredweight rankings on the horizontal axis. Most of the farms fall very close to the 45-degree line, indicating the rankings are quite similar (points falling on the line represent farms with the same ranking for each measure). On the basis of the output for these 40 farms, the ranking of net returns over total cost is closely correlated for both outcomes (profit per cow vs. profit per 100 lb of milk). Thus, analyzing either outcome is sufficient.

The farms were analyzed by calculating an average of each revenue, cost, or other factor category from its years of reporting data. On the basis of this average, the farm was placed in the top, middle, or low profitability group. Once profitability groups were formed, group data were evaluated at an average for each category. Then, the various farm characteristics (e.g., farm size, milk production, milk price, income, and costs) were compared for all 3 profitability groups. Differences between the high and low groups were reported as both absolute and percentage differences. As an additional analysis, regression models were estimated to quantify relationships that existed among farm characteristics (independent variables) and profit and cost (dependent variables). Independent variables included were herd size, milk production (pounds per cow), milk price (profit regression only), cull rate, feed as a percentage of total cost, and percentage of farm labor allocated to livestock.

## Results and Discussion

Table 1 reports farm information, income, and cost data for the 3 profitability groups (high, medium, and low). Farms in the high profit group had larger herds than the medium and low profit farms. Culling rate, defined as cows purchased divided by herd size, was similar for high and low profit farms but considerably less for the medium profit farms. Pounds of milk produced per cow was the most significant independent factor affecting profit, with a total difference between the top and bottom profitability groups of 30% (5,268 lb per cow). Milk price was similar for all 3 groups and was actually slightly greater (\$0.56/100 lb of milk) for the low profit farms compared with the high profit farms. Given the greater milk production and comparable price, the high profit farms generated nearly \$800 per cow more income from milk sales. As profitability increased, net cattle sales increased because of greater culling rates or because cows sold had greater value as a result of increased production or genetics. Given that culling rates were similar, greater net cattle sales for the high profit farms were likely the result of cows in the high profit group being sold for greater value than those in the low profit group. Other income was a fairly minor category, and no differences existed among the 3 profitability groups. When all categories were included, the high profit farms averaged \$1,065 more income per cow than the low profit farms (slightly more than \$500 per cow compared with medium profit farms).

## ECONOMICS

A comparison of cost categories among the different profitability groups showed that high profit farms had greater costs in some categories and lower costs in others, and they had a slightly higher total cost per cow (Table 1). High profit farms spent about \$90 per cow more on feed costs, which, given the large difference in milk production, clearly indicates a considerably smaller feed cost per 100 lb of milk. Labor costs per cow were slightly lower for the high profit farms, which is likely due to high profit farms being larger and therefore relying on more hired labor (i.e., operator labor makes up a smaller percentage of total labor). When aggregated, veterinary and dairy supplies costs were about \$140 per cow greater for the high profit farms compared with the low profit farms. Machinery and utilities/fuel were lower for the high profit farms (\$49.71 and \$30.64, respectively), which likely is a result of spreading costs over more cows (i.e., economy of size). The high profit farms averaged \$62.63 per cow more cost, but when coupled with the significantly greater income (\$1,065), net returns were greater. High profit farms averaged \$1,000 more profit than low profit farms and had a \$450 per cow advantage over medium profit farms. These differences were very significant and almost entirely the result of high profit farms producing more milk while holding costs constant.

To independently evaluate the effects of various farm characteristics on economic variables, 2 regression models were estimated: 1 focused on profit and 1 on total costs. In the profit analysis, pounds of milk produced per cow accounted for the most variation ( $P < 0.001$ ) in profitability, revealing that production is the driving factor behind profitability differences between operations (Table 2). Milk price in relation to profit, with all other variables held constant, was also a key factor ( $P < 0.05$ ). Of the cost factors evaluated, feed as a percentage of total cost and percentage of farm labor devoted to livestock were not significantly related to profit. In addition, variability in culling rates between operations was not related to profitability differences.

When costs per cow were evaluated with farm characteristics variables, pounds of milk produced per cow again was the most economically significant factor (Table 2). Percentage of labor allocated to livestock was positively associated with costs per cow and tended ( $P < 0.10$ ) to account for a significant proportion of the variation associated with cost per cow. This positive relationship may indicate that farms that are more diversified (i.e., lower percentage of labor devoted to livestock) have lower costs per cow than farms that are more specialized. In contrast, feed cost as a percentage of total costs was not related to costs per cow.

Two other factors evaluated in the regression were culling rate and mean year of reported data. Culling rate, which was not statistically related to profitability, was positive and affected ( $P < 0.05$ ) total cost. In other words, selling cull cows did not affect profit, but greater culling rates were associated with higher total costs per cow. In addition, the mean year of reported data tended ( $P < 0.10$ ) to be significant in explaining profit and was related ( $P < 0.05$ ) to explaining costs per cow. These trends in year differences may slightly skew the results reported in Table 1 because the year effect is not accounted for. However, because the regression results account for year differences and because data used in the analyses represent 4.5 of the 5 years for all reporting farms, one can be confident the results are accurate and can conclude that increasing milk production is key to increasing profit for small to midsize dairy enterprises in Kansas.

ECONOMICS

**Table 1. Dairy enterprise measures among high, medium, and low profit groups<sup>1</sup>**

| Item <sup>2</sup>              | All farms | Profit category |         |           | High minus low |     |
|--------------------------------|-----------|-----------------|---------|-----------|----------------|-----|
|                                |           | High            | Middle  | Low       | Absolute       | %   |
| Number of farms                | 40        | 13              | 14      | 13        |                |     |
| Number of cows per herd        | 115       | 135             | 130     | 79        | 57             | 72  |
| Culling rate, %                | 25.6      | 28.1            | 21.0    | 28.0      | 0.13           | 0   |
| Pounds of milk                 | 20,610    | 22,966          | 21,129  | 17,697    | 5,268          | 30  |
| Milk price, per 100 lb of milk | \$16.48   | \$16.32         | \$16.25 | \$16.88   | -\$0.56        | -3  |
| Milk sales                     | \$3,369   | \$3,731         | \$3,420 | \$2,951   | \$780          | 26  |
| Net cattle sales               | \$267     | \$440           | \$209   | \$156     | \$284          | 183 |
| Other income                   | \$59      | \$59            | \$60    | \$58      | \$0.89         | 2   |
| Gross income                   | \$3,695   | \$4,230         | \$3,689 | \$3,165   | \$1,065        | 34  |
| Feed                           | \$1,749   | \$1,763         | \$1,807 | \$1,672   | \$90.91        | 5   |
| Labor                          | \$596     | \$594           | \$528   | \$672     | -\$78.19       | -12 |
| Vet                            | \$115     | \$113           | \$128   | \$102     | \$10.47        | 10  |
| Dairy supplies                 | \$300     | \$354           | \$321   | \$224     | \$130.32       | 58  |
| Marketing/breeding             | \$93      | \$86            | \$111   | \$82      | \$4.05         | 5   |
| Machinery                      | \$323     | \$308           | \$304   | \$358     | -\$49.71       | -14 |
| Utilities/fuel                 | \$158     | \$151           | \$143   | \$182     | -\$30.64       | -17 |
| Interest                       | \$319     | \$335           | \$291   | \$332     | \$3.00         | 1   |
| Other                          | \$76      | \$76            | \$60    | \$93      | -\$17.59       | -19 |
| Total cost                     | \$3,729   | \$3,779         | \$3,694 | \$3,716   | \$62.63        | 2   |
| Net return to management       | -\$34.34  | \$451.06        | -\$5.04 | -\$551.31 | \$1,002        |     |

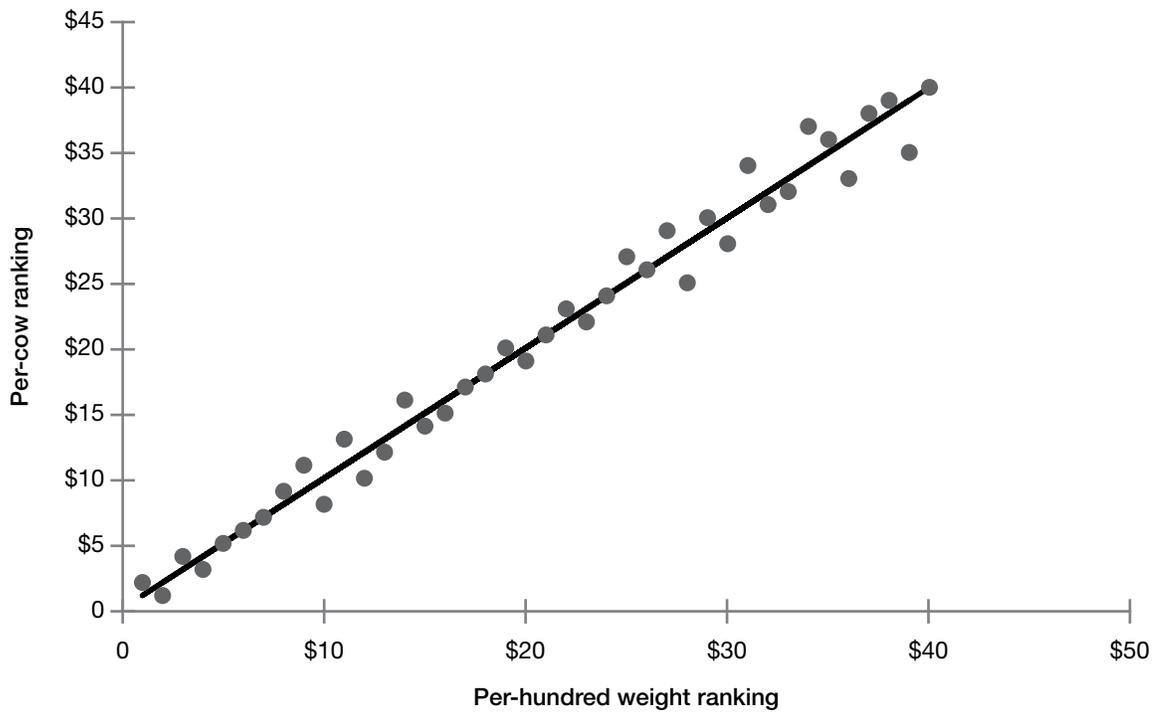
<sup>1</sup> Sorted by net return to management (returns over total costs) per cow.

<sup>2</sup> All items are on a per-cow basis unless indicated otherwise.

**Table 2. Regression analysis for profit and cost models**

| Variable                      | Profit (\$/cow) |         | Cost (\$/cow) |         |
|-------------------------------|-----------------|---------|---------------|---------|
|                               | Coefficient     | P value | Coefficient   | P value |
| Intercept                     | 36,901          | 0.113   | -42,958       | 0.025   |
| Cows, number of head          | 1.00            | 0.385   | -0.26         | 0.798   |
| Milk production, lb/cow       | 0.08            | 0.002   | 0.10          | 0.001   |
| Milk price, \$/100 lb of milk | 176.82          | 0.024   |               |         |
| Culling rate, %               | 1.14            | 0.793   | 9.64          | 0.038   |
| Feed percent of total cost    | 17.07           | 0.175   | -15.27        | 0.157   |
| Livestock labor percentage    | -3.64           | 0.464   | 7.68          | 0.079   |
| Years                         | -398.91         | 0.080   | 421.27        | 0.021   |
| R-square                      | 0.4547          |         | 0.6217        |         |

### ECONOMICS



**Figure 1. Gross return less total cost (i.e., profit) is correlated positively ( $r = 0.99$ ) when analyzed per farm on a per-hundredweight (100 lb of milk) basis versus a per-cow basis.**

# Temperature Changes in a Low-Profile, Cross-Ventilated Building in the High Plains

*J. P. Harner<sup>1</sup> and J. F. Smith*

## Summary

Performance of an evaporative cooling system was evaluated in the High Plains in a low-profile, cross-ventilated dairy facility housing 4,200 lactating cows. The temperature decrease across the 6-inch cellulose evaporative pad during the afternoon hours from July 15 to August 14, 2008, was 12.6°F. The temperature-humidity index was below 72 for 14 and 19 hours/day in pens near the outlet (exhaust fans) and inlet (near evaporative cooling pad), respectively, compared with 12 hours under ambient conditions. Throughout the study period, the evaporative cooling system decreased the number of hours that cows were housed in a heat stress environment irrespective of pen location in the building.

## Introduction

Optimizing the environment where cows are housed increases milk production, improves feed efficiency, raises income over feed cost, strengthens reproductive performance, allows for controlled lighting, reduces lameness, and lessens fly-control costs. Dairy cows housed in an environment outside their thermoneutral zone alter their behavior and physiology to adapt. Adaptations help maintain a stable core body temperature, but nutrient utilization and profitability are negatively affected. Previous studies of the performance of LPCV buildings were based on data collected in the north central part of the United States. The objective of this study was to evaluate the performance of an LPCV building located in the High Plains, a more arid climate.

## Experimental Procedures

A 4,200-cow LPCV building located in the Texas Panhandle was monitored in summer 2008 to evaluate the performance of an evaporative cooling system in the High Plains. The building was oriented north to south with a center transfer lane to the milk center. The building was 320 ft wide by 1,400 ft long. The building had a 10-ft evaporative cellulose pad along the west side. The pad was 6 inches thick. Three temperature-humidity recording devices were placed on 6 baffles located in the northern half of the building. Each half of the building had 6 groups of lactating dairy cows with a baffle located between the head-to-head stalls in each pen. Devices recorded temperature and humidity every 15 minutes throughout the study period. Data were analyzed from July 15 to August 14, 2008. Devices were hung approximately 7 ft above the concrete alleys. Two temperature-humidity devices were placed in sun shields and installed outside the building to record ambient conditions. Three hundred sixty data points were averaged to obtain the average hourly temperature and humidity value at each baffle during the study period. Temperature-humidity data were used to calculate the ambient and interior temperature-humidity indices (THI).

## Results and Discussion

Figures 1 and 2 show the average hourly temperature and relative humidity, respectively, during the 30-day study period.

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<sup>1</sup> Department of Biological and Agricultural Engineering, Kansas State University.

## FACILITIES AND ENVIRONMENT

Baffle 1 was located approximately 23 ft from the evaporative pad at the inlet side of the building. Baffle 6 was located approximately 23 ft from the exhaust fans at the outlet side of the building. The average temperature drop observed between noon and 2000 hours was 12.6°F. During these afternoon hours, the relative humidity was less than 40% (Figure 2). The relative humidity decreased 5 to 10% as the air moved across the building and was warmed by the heat being transferred from the lactating cows to the air. The average hourly air temperature increase across the building was 0.0094°F/ft of building width.

Figure 3 shows the hourly ambient and interior THI. From 1000 to 2000 hours, the ambient THI was above 76. The THI remained at or below 72 during the 24-hour period in pen 1 near the inlet. The THI remained below 75 in the pen near the exhaust side of the building.

Figure 4 compares ambient temperature with the temperature at baffle 1 for the entire data collection period. The temperature difference between these 2 points reflects the ability of the evaporative pad to cool the incoming air. The lack of accuracy, as indicated by the low  $R^2$  value, in predicting the temperature at baffle 1 on the basis of ambient temperature is due to the variability of the ambient relative humidity. Relative humidity in the High Plains is variable at a given temperature. Figure 2 shows that humidity varied from 40% during the afternoon hours to nearly 80% during the early morning hours.

Figure 5 compares the ambient THI to the THI at baffle 1. When humidity is included, the correlation increases and the ambient THI value could be used to predict the THI at baffle 1, indicating the potential for heat stress. Figure 6 shows the average hours per day during the study period when the THI was less than 72. The ambient THI was less than 72 for only 12 hours/day. At baffle 1 near the inlet, the THI was less than 72 for nearly 19 hours/day. This resulted in 58% less time each day when the cows were experiencing heat stress. The THI was less than 72 for only 14 hours/day at baffle 6 near the exhaust fan, which provided 16% fewer heat stress hours per day. Figure 6 shows the impact of the air absorbing heat generated by the lactating cows as the air moves from baffle 1 (inlet) to baffle 6 (exhaust).

Temperature drop across the 6-inch evaporative pad during the afternoon hours from July 15 to August 14, 2008, was 12.6°F. The THI was below 72 for 14 and 19 hours/day in pens near the outlet and inlet, respectively, compared with 12 hours under ambient conditions. Throughout the study period, the evaporative cooling system decreased the number of hours that cows were housed in a heat stress environment irrespective of pen location in the building.

### FACILITIES AND ENVIRONMENT

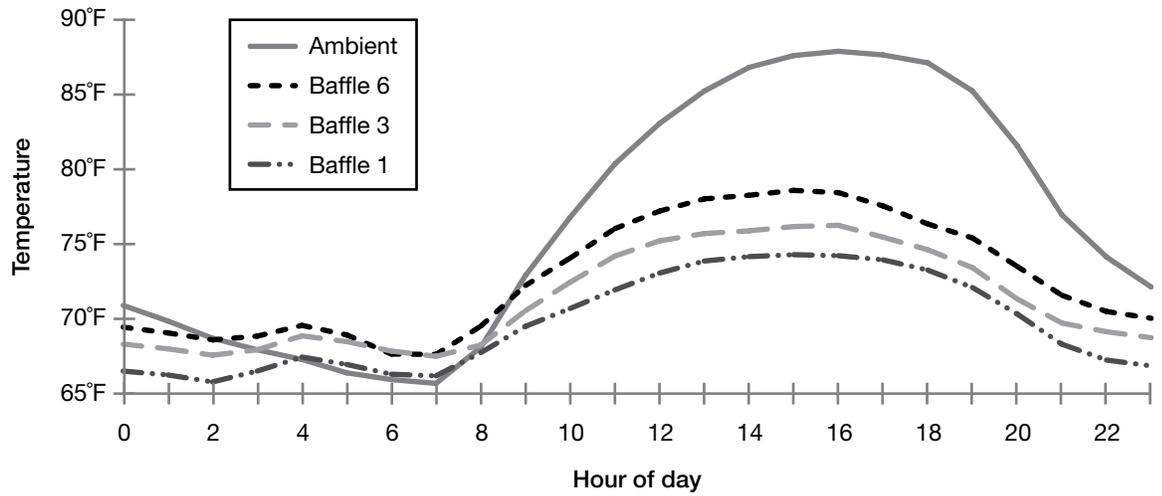


Figure 1. Hourly average ambient temperature and temperature at different locations inside a low-profile, cross-ventilated dairy facility in the High Plains from July 15 to August 14, 2008.

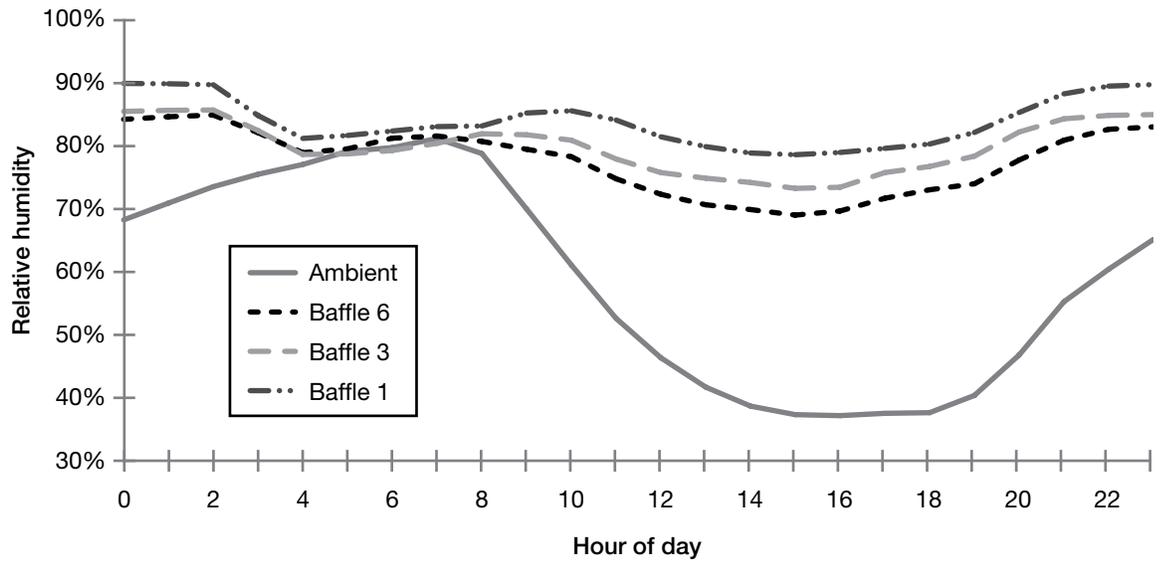


Figure 2. Hourly average ambient relative humidity and relative humidity at different locations inside a low-profile, cross-ventilated dairy facility in the High Plains from July 15 to August 14, 2008.

### FACILITIES AND ENVIRONMENT

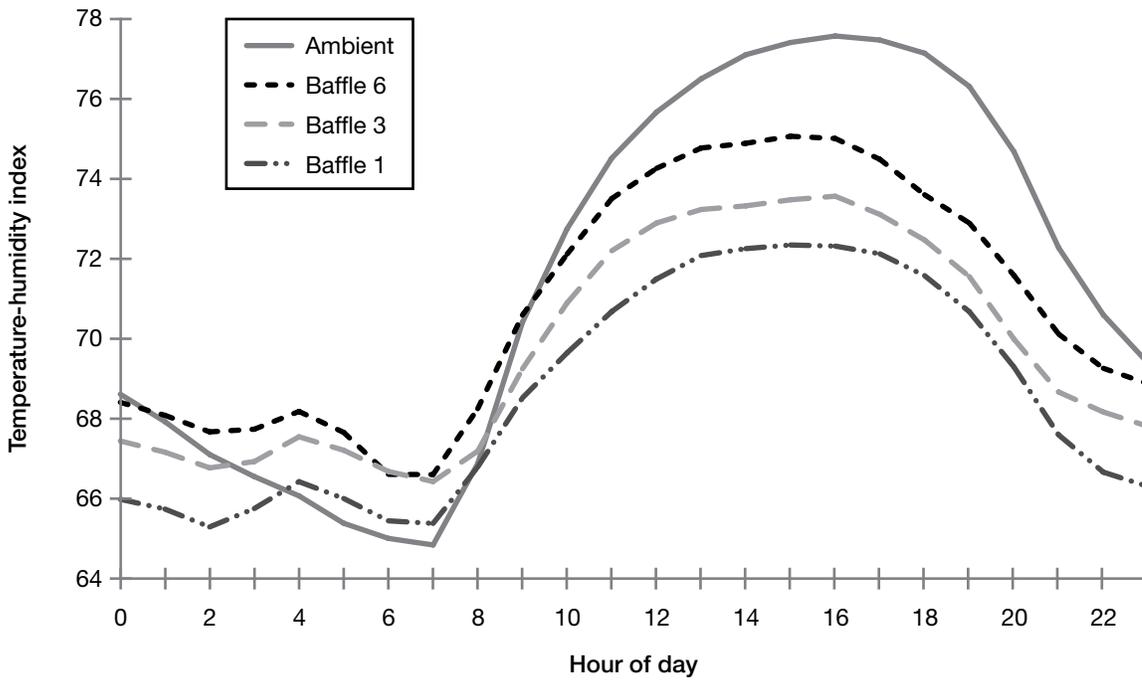


Figure 3. Hourly average ambient temperature-humidity index and temperature-humidity index at different locations inside a low-profile, cross-ventilated dairy facility in the High Plains from July 15 to August 14, 2008.

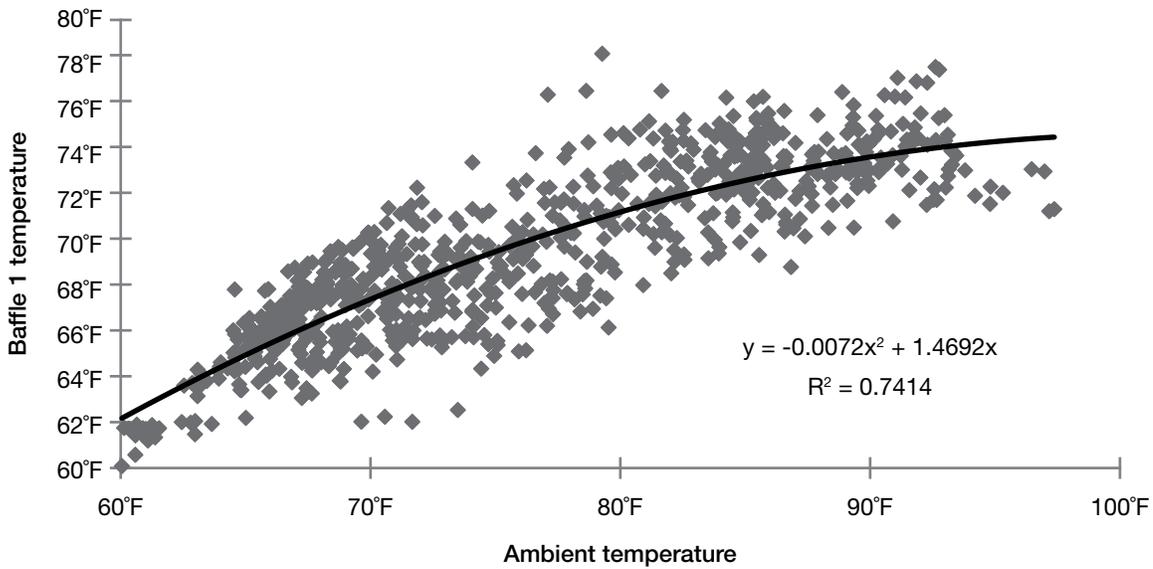
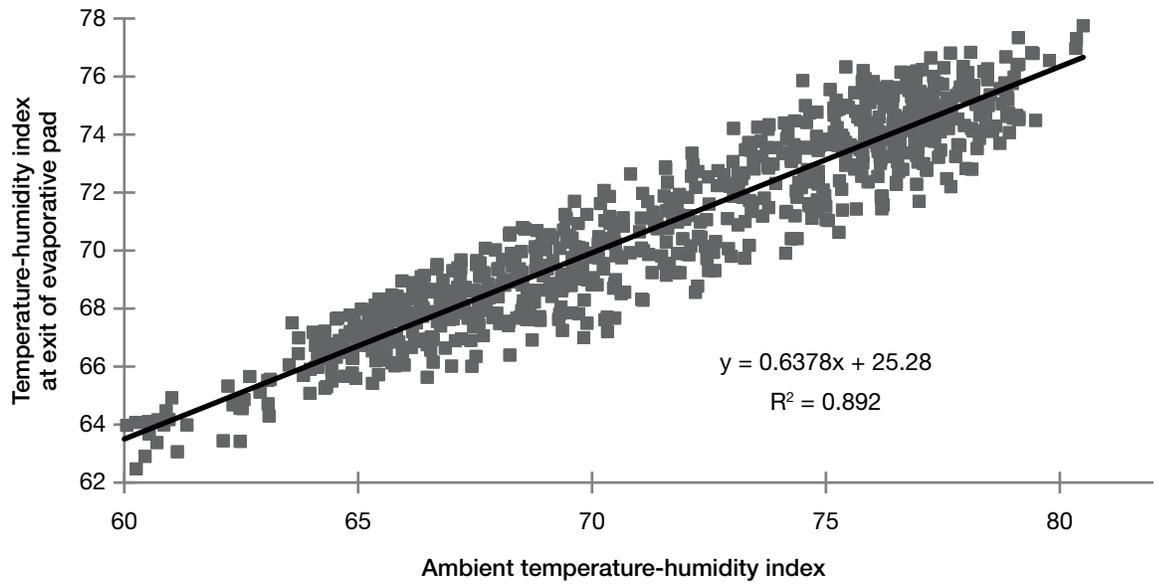
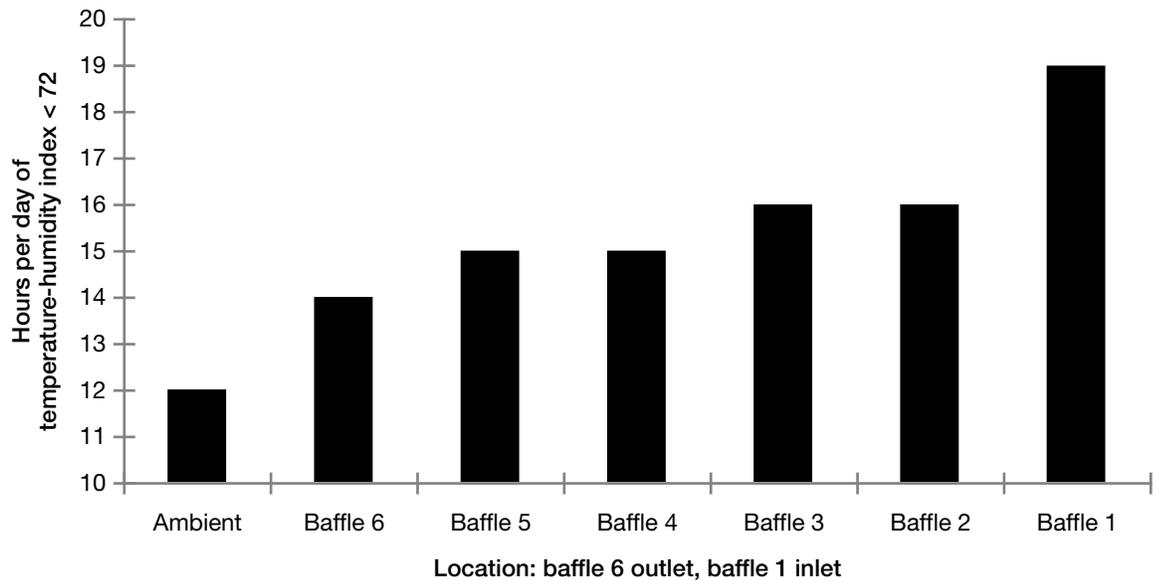


Figure 4. Comparison of ambient and baffle 1 temperature in a low-profile, cross-ventilated dairy facility in the High Plains from July 15 to August 14, 2008.

FACILITIES AND ENVIRONMENT



**Figure 5. Comparison of ambient and baffle 1 temperature-humidity indices in a low-profile, cross-ventilated dairy facility in the High Plains from July 15 to August 14, 2008.**



**Figure 6. Comparison of hours per day at different locations when the temperature-humidity index was less than 72 in a low-profile, cross-ventilated dairy facility in the High Plains from July 15 to August 14, 2008.**

# Land Requirements for Freestall Dairy Facilities

*J. P. Harner<sup>1</sup> and J. F. Smith*

## Summary

Existing blueprints were used to estimate land requirements for new dairy facilities. The average land requirement for constructing a new dairy complex with freestall housing and a new parlor is 915 ft<sup>2</sup> per lactating cow. Approximately 52% of the overall land space is used for dairy operations including a milk center, housing, transfer lanes, vehicle roads, a feed center, and a manure processing center. The remaining 48% is green space, areas between buildings or along driveways, and separation distance from main roads and neighboring property.

## Introduction

New dairies ask engineering firms to develop an overall site layout of proposed facilities. These plans are submitted as part of the water, air, environmental, and zoning permitting requirements to local and state agencies. Permitting may require air modeling to estimate potential gaseous or particulate emissions. Currently, information is not available to estimate land requirements necessary for the overall physical footprint of a dairy before actual design of a dairy. Allocated areas may affect air quality differently. For example, nonpaved feed roads may be the main source of particulate emissions, whereas manure storage structures may be the main source of odorous compounds. The objective of this paper was to establish baseline data to estimate the land area required for the physical structures located on a dairy.

## Experimental Procedures

Thirty sets of site plans were obtained from 8 different North American engineering firms, and useable data were obtained from 28 of the 30 site plans. Each plan was a proposed or recently constructed dairy with freestall housing and a new milk center. Some plans had exercise lots located between the freestall buildings. Exercise lots commonly are found on freestall facilities in western states. Not all of the plans had space allocated for a feed center. Scaled measurements taken from the plans formed a database that was used evaluate space requirements for housing, feed, parlor, and waste management systems.

## Results and Discussion

The dairies represented in the site plans averaged 2,589 lactating cows (range: 246 to 10,000 lactating cows). The average land requirement was 915 ft<sup>2</sup> per lactating cow. The maximum and minimum land area allocated per lactating cow was 1,566 and 358 ft<sup>2</sup>, respectively. The freestall housing area averaged 102 ft<sup>2</sup> per lactating cow, and the special needs space required 29 ft<sup>2</sup> per lactating cow. Roads providing access to the milk center, housing area, and feed center occupied 51 ft<sup>2</sup> per lactating cow. Average space for exercise lots on freestall dairies was 167 ft<sup>2</sup> per lactating cow. Table 1 summarizes the overall requirements for land and housing areas.

Space requirements for the milk center are shown in Table 2. Average space in the milk center was 7 ft<sup>2</sup> per lactating cow. The parlor equipment room averaged 2 ft<sup>2</sup> per lactating cow, and travel lanes to the parlor averaged 7 ft<sup>2</sup> per lactating cow. Significant variation existed between the average, maximum, and minimum space requirements for the travel lanes. Dairies with exer-

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cise lots tended to have greater travel lane space requirements than dairies in northern climates, where facilities tend to be more compact.

Feed center space requirements averaged 86 ft<sup>2</sup> per lactating cow. The feed centers included space for hay storage, commodity buildings, and silage structures. On average, about half of this space was used for silage storage structures.

Nearly all plans showed at least 1 solids storage basin or pad if a mechanical separator was shown along with liquid storage for handling manure, wash water, and runoff. The maximum number of solids storage basins was 4, which is common on flush dairies that use long, narrow trenches for solids separation. The average surface area of the space allowed for solid and liquid storage was 41 and 140 ft<sup>2</sup>, respectively, per lactating cow.

Figure 1 shows facility space and overall space for the different dairies by number of lactating cows. Approximately 52% of the overall land space used for dairy operations consists of dairy operations including the milk center, housing, transfer lanes, vehicle roads, the feed center, and the manure processing center. The remaining 48% is green space, areas between buildings or along driveways, and separation distance from main roads and neighboring property.

**Table 1. Overall requirements for land and housing areas**

| Item                                  | Average | Maximum | Minimum | Deviation | Count <sup>1</sup> |
|---------------------------------------|---------|---------|---------|-----------|--------------------|
| Number of cows                        | 2,589   | 10,000  | 246     | 2,041     | 28                 |
| Overall land, ft <sup>2</sup>         | 915     | 1,566   | 371     | 345       | 23                 |
| Freestall/Housing, ft <sup>2</sup>    | 102     | 134     | 79      | 15        | 28                 |
| Special needs, ft <sup>2</sup>        | 29      | 191     | 1       | 44        | 28                 |
| Exercise lots, ft <sup>2</sup>        | 167     | 293     | 68      | 92        | 5                  |
| Feed lanes and roads, ft <sup>2</sup> | 51      | 156     | 18      | 35        | 18                 |

<sup>1</sup>Number of useable site plans.

**Table 2. Space requirements for the milk center (ft<sup>2</sup>)**

| Item                  | Average | Maximum | Minimum | Deviation | Count <sup>1</sup> |
|-----------------------|---------|---------|---------|-----------|--------------------|
| Milk center           | 7       | 20      | 3       | 3         | 28                 |
| Parlor equipment room | 2       | 9       | 1       | 2         | 28                 |
| Lanes to parlor       | 7       | 23      | 1       | 5         | 26                 |

<sup>1</sup>Number of useable plans.

### FACILITIES AND ENVIRONMENT

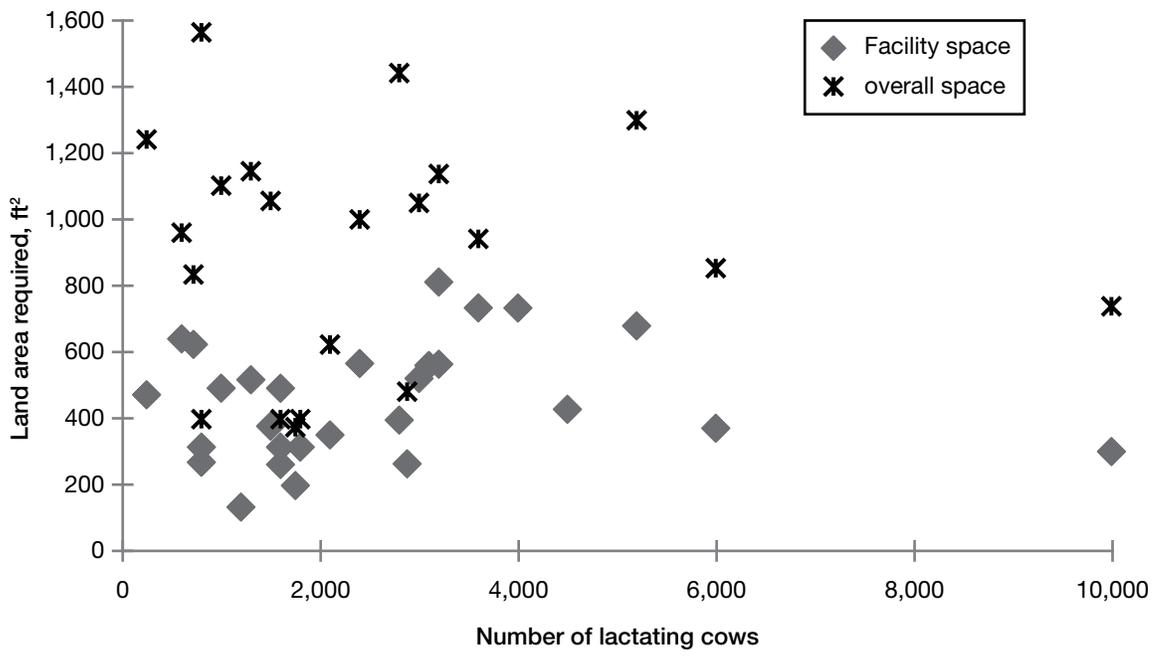


Figure 1. Comparison of land area requirements for facility space and overall space based on number of lactating cows in a dairy.

# Water Consumption of an Evaporative Cooling System in the Midwest

*J. P. Harner<sup>1</sup>, J. F. Smith, R. Zimmerman<sup>2</sup>, and M. VanBaale<sup>3</sup>*

## Summary

Water meters were installed on the evaporative cooling system of a long, low-profile, cross-ventilated dairy in the upper Midwest. The evaporative pad along the west side measured 10 by 350 ft. The water usage per unit surface area of the evaporative pad was 0.29 gallons/hour per square foot of evaporative pad surface area. The total daily water usage per stall averaged 13 gallons with a maximum of 22.7 gallons. Results from this study indicate that peak hourly water usage may be as much as 3 times the average values. The evaporative pad efficiency was 65% between noon and 0800 hours and 79% between midnight and 0400 hours.

## Introduction

Consumptive water use on a dairy for heat abatement increases the daily water requirements during summer. Water usage depends on weather conditions, the heat abatement system, and operational characteristics. Evaporative cooling systems cool the air around a cow's body to help minimize heat stress and maintain an animal environment in the thermoneutral zone. Distributors of evaporative pads assume a pad efficiency of 75% regardless of the air properties or air speed. Little information exist describing water demand and water usage of evaporative pads, making it difficult to properly design the water distribution system. The objective of this study was to determine consumptive water usage of evaporative cooling systems on a low-profile, cross-ventilated dairy facility.

## Experimental Procedures

Water meters were installed on the evaporative cooling system of a low-profile, cross-ventilated dairy in the upper Midwest. The building was 372 ft wide and 390 ft long with an evaporative pad along the west side that measured 10 by 350 ft. The facility housed 1,200 dairy cows assuming a 1:1 stocking ratio of the freestalls. The cellulose pad was 6 inches thick. Water was delivered to the supply line of the pad by 5 sump pumps. Each pump supplied water to a pad section measuring 10 ft tall and approximately 70 ft long. Automatic data loggers were installed at the inlet to each sump and programmed to record consumptive water use every 15 minutes. Ambient temperature and humidity were obtained at the nearest town from a weather service provider. Water use data were evaluated during 8-hour periods (noon to 2000 hours) from July 1 to 31, 2008. The evaporative pad area was 2.9 ft<sup>2</sup> per freestall. A psychometric spreadsheet model was used to estimate the maximum potential water used assuming 100% efficiency of the evaporative cooling system. Pad efficiency was calculated as the actual water used divided by the maximum potential water that could have been absorbed by the air.

## Results and Discussion

During the 31-day period, ambient relative humidity was  $63.8 \pm 12.2\%$ , ambient temperature was  $81.2 \pm 4.4^\circ\text{F}$ , and the ambient temperature-humidity index was  $76.5 \pm 3.5$ . Total water consumption during the 31-day period was 482,350 gallons. The maximum water used during

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## FACILITIES AND ENVIRONMENT

a 24-hour period was 27,231 gallons, and the minimum usage was 5,493 gallons. Average total daily water usage per stall was 13 gallons with a maximum of 22.7 gallons per stall. Water usage exceeded 16.7 gallons/day per stall during 23% of the days. Pad efficiency was  $65.0 \pm 16.2\%$  during the 31-day period between noon and 2000 hours. Average hourly water usage was  $1,028 \pm 405$  gallons/hour, and average water usage per unit pad area was 0.29 gallons/hour per square foot. Peak hourly usage rate during the 31-day period was 2,655 gallons/hour, resulting in a peak water usage rate of 0.76 gallons/hour per square foot of pad area. Defining peak water usage is critical because the water system must meet this demand. During 4% of the study period, water usage exceeded 1,500 gallons/hour. Average water usage was 358 gallons/hour during the night, when the humidity averaged 86%. The pad efficiency also increased to 79% during the night. It is essential that evaporative pad systems be designed to meet peak demand.

# Impact of Evaporative Pads and Cross Ventilation on Core Body Temperature and Resting Time of Lactating Cows

*J. F. Smith, B. J. Bradford, J. P. Harner<sup>1</sup>, K. Ito<sup>2</sup>, M. von Keyserlingk<sup>2</sup>, C. R. Mullins, and J. Potts*

## Summary

A trial was conducted to determine the impact of evaporative cooling pads on core body temperature (CBT), time spent lying, and number of lying bouts of Holstein cows housed in cross-ventilated freestall facilities. Despite cool ambient conditions during the trial, cows without evaporative pads tended to have elevated CBT above 102°F for 2.3 more hours per day and elevated CBT above 102.5°F for 0.95 more hours per day than cows with evaporative pads. These trends were evident even though the stocking density of the freestalls was greater in the facility with evaporative pads than in the facility without pads (123 vs. 113%). Lying times and lying bouts did not differ between treatments. Results of this study indicate that CBT tended to be reduced when evaporative pads were used, even under relatively mild ambient conditions.

## Introduction

With the adoption of low-profile, cross-ventilated freestall facilities, dairy producers are asking questions concerning the impact of evaporative pads on dairy cattle performance. Cooling systems that reduce core body temperature (CBT) improve milk production and reproductive performance of dairy cattle. Increasing resting time also has a positive impact on milk production. During the summer of 2009, a trial was conducted at Morris, MN, to evaluate the impact of evaporative pads on CBT, time spent lying, and number of lying bouts of Holstein cows housed in cross-ventilated freestall facilities.

## Experimental Procedures

Two cross-ventilated facilities — 1 with and 1 without evaporative pads — were used in this study. Each facility had 4 pens of cows and a nominal width of 400 ft. Both facilities had 1 baffle per pen of cows. One hundred forty-three cows were fit with activity data loggers (HOBOS) to determine time spent lying and number of lying bouts per day, and 87 cows were fit with blank intravaginal HOBOS to determine CBT every 5 minutes. Environmental data were collected with activity loggers every 15 minutes at both sites to determine temperature, relative humidity, and temperature-humidity index. Individual cow CBT and activity data (9 days per cow) were analyzed to determine the amount of time when CBT exceeded 102°F or 102.5°F, time spent lying, and number of lying bouts per day. These variables were analyzed statistically using pen as the experimental unit and including cow and day as additional random effects. Parity, reproductive status, and days in milk were tested as covariates in each model but were removed if they did not contribute significantly to the prediction equation.

## Results and Discussion

Environmental data for the 2 locations are presented in Figures 1, 2, and 3. Environmental data are illustrated for ambient conditions and at the baffle closest to the intake and exhaust for each

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freestall facility. Average ambient conditions were mild during the trial, with afternoon highs of 77°F. Lying times and lying bouts did differ between treatments. Cows housed in the facility with evaporative pads had lying times of 660 minutes/day and 12 lying bouts per day, whereas cows without evaporative pads had lying times of 654 minutes/day and 12.9 lying bouts per day. Time that CBT exceeded 102°F or 102.5°F tended ( $P = 0.06$ ) to be greater for cows without evaporative pads. Core body temperature was above 102°F for 566 and 704 minutes/day and above 102.5°F for 321 and 378 minutes/day for cows with and without evaporative pads, respectively. Despite the cool ambient conditions, cows without evaporative pads tended to have elevated CBT above 102°F for 2.3 more hours per day and elevated CBT above 102.5°F for 0.95 more hours per day than cows with evaporative pads. These trends were evident even though the stocking density of the freestalls was greater in the facility with evaporative pads than in the facility without pads (123.4 vs. 113.1%). Results of this study indicate that CBT tended to be reduced when a cross-ventilated barn was equipped with cooling evaporative pads, even under relatively mild ambient conditions.

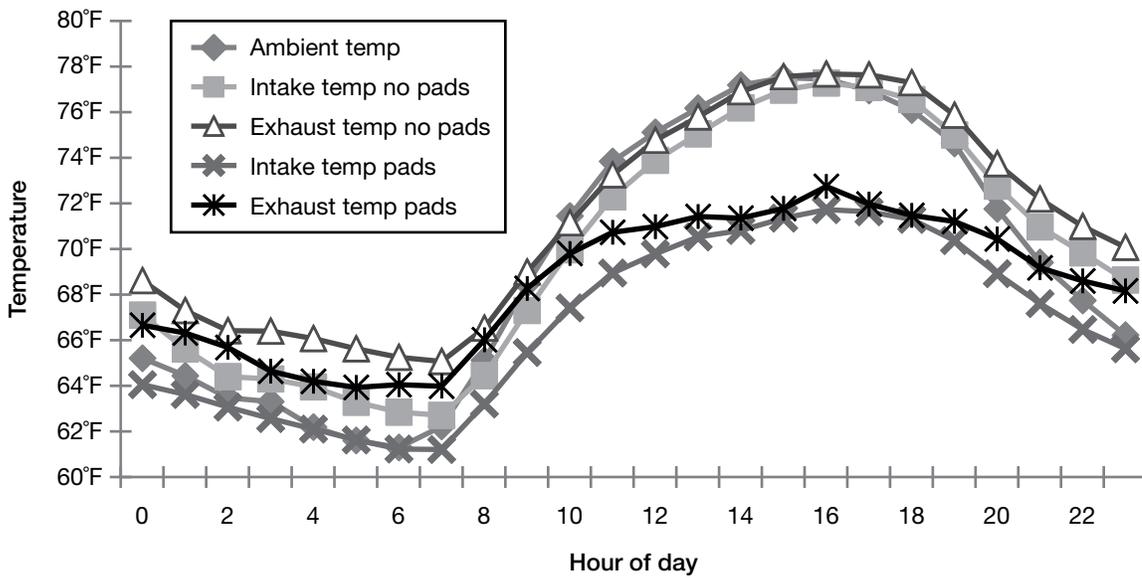


Figure 1. Ambient temperature during 9 days of the research trial.

FACILITIES AND ENVIRONMENT

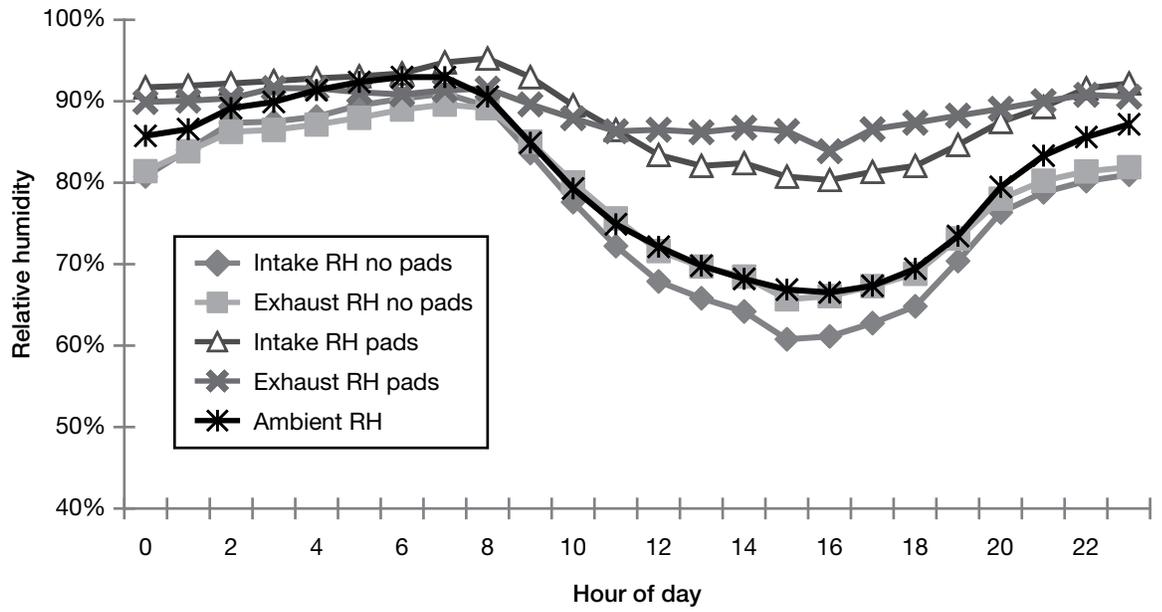


Figure 2. Ambient relative humidity during 9 days of the research trial.

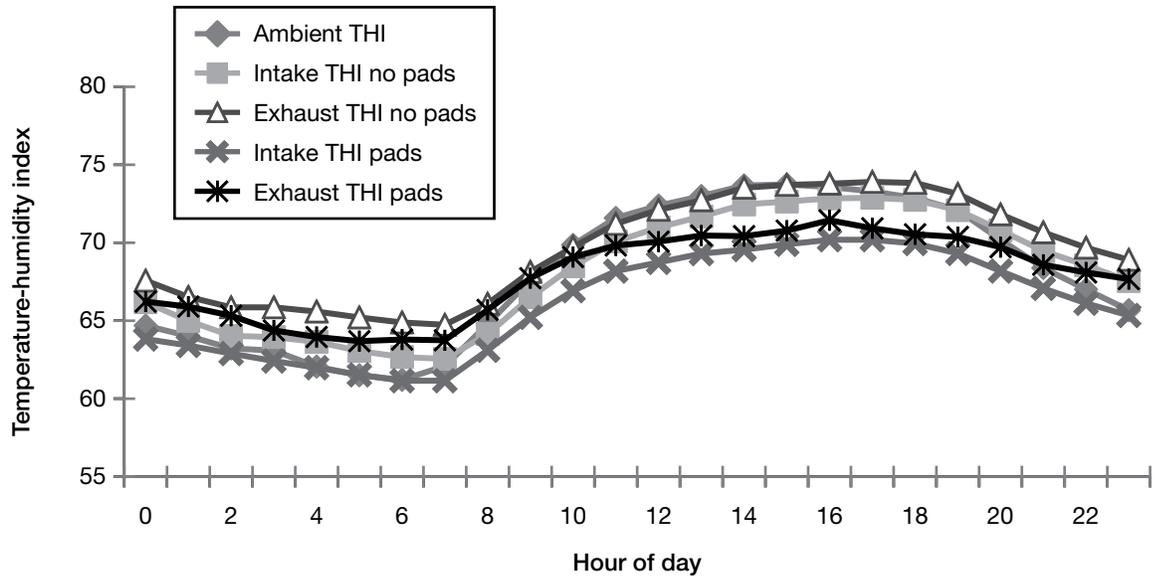


Figure 3. Ambient temperature-humidity index during 9 days of the research trial.

# Influences of Heat Stress on Serological Response and Performance of Dairy Calves

*D. M. Meyer, M. J. Brouk, and L. C. Hollis*

## Summary

Objectives of this study were to investigate the possible effects of heat stress on calf growth and the development of active immunity. Eighteen heifer calves born between July 21 and August 24, 2008, were housed in individual hutches, and half of the calves were provided supplemental shade from birth to 8 weeks of age. During this time, milk replacer intake, dry feed intake, and fecal scores were recorded daily. Calf weight and hip and shoulder heights were measured and recorded weekly. The bovine viral diarrhea portion of the vaccine given at 5 weeks of age was used as an indicator to track the development of humoral immunity. Intake, growth, temperature response after vaccination, and serum titers did not differ significantly between treatments. In contrast, differences in hutch temperature, relative humidity, and temperature-humidity index were observed between treatments. Results indicated that supplemental shade provided to calves housed in hutches does not affect their performance or ability to develop active immunity.

## Introduction

Heat stress and disease prevention for young and aged animals are two fundamental areas of concern and study in the dairy industry. Both areas have received much individual attention concerning their relationship to performance. Effect of stress on immunization efficacy, however, has seldom been considered.

Bovine viral diarrhea (**BVD**) is a single-stranded RNA virus that can decrease performance and cause a host of health-related problems in cattle, most commonly diarrhea, abortion, and immunosuppression, that generally result in secondary respiratory infection. Cattle of all ages are susceptible to BVD. It is contained in all body fluids of infected animals and transmitted primarily by aerosol. Although constant surveillance is important, immunization for BVD also is a common preventative measure in dairy herds. Vaccination generally is administered with a modified-live virus product. Furthermore, immunological response to the virus is tested easily by blood serum neutralization titers or tissue samples. These traits make the virus valuable to this study as an indicator of the calf's immune response.

Heat stress also has been clearly linked to decreased dry matter intake and other measures of performance including milk yield, average daily gain, and feed:gain ratio. Heat stress in cows commonly is thought to be a more relevant concern to the industry than heat stress in calves because calves produce less metabolic heat and have a greater surface area to body mass ratio with which to dissipate heat. Supplemental shade has been shown to decrease heat stress of calves housed in hutches and improve heifer performance in feedyards.

Most research concerning thermoregulatory responses to immunity development is related to passive immunity and colostral quality and immunoglobulin transfer. A recent study suggested that heat stress did not affect active immunity development of calves. Therefore, the objectives of this study were to determine the effect of heat stress on the development of BVD titers, study the effects of heat stress on intake and growth, and monitor environmental changes in order to

correlate them to measures of stress in Holstein dairy calves. We hypothesized that heat stress would negatively affect calf intake, growth, and immunological response and that supplemental shade would reduce these effects.

## Experimental Procedures

Each of 18 Holstein heifer calves was assigned randomly to a Calf-Tel (Hampel Corporation, Germantown, WI) hutch in either the shade or sun treatment in blocks of 2 calves each according to date of birth. Calves were weighed and hip and wither heights were recorded at birth. Calves were then weighed and measured weekly through 8 weeks of age on the day of the week they were born at about 1630 hours, just after their evening milk feeding was completed. Calves born on Sunday were weighed on Monday, and those born on Saturday were weighed on Friday. Daily morning and afternoon amounts of milk replacer and starter feed fed and refused as well as morning and afternoon fecal scores were recorded during this time. Milk replacer was fed only in the afternoon at 5 weeks of age and discontinued at 6 weeks of age.

At 8 weeks of age, calves were moved to group lots with 8 to 9 calves in each pen and housed there for the remainder of the study. Calves were weighed and measured at 10, 12, 14, and 15 weeks of age on a single day of the week as close to their actual birth date as possible.

Calves were given 2 mL TSV-2 intranasally, 2 mL Ultrabac CD subcutaneously, and 2 mL Calf-Guard orally at birth; a second 2-mL dose of Ultrabac CD was given again at 2 weeks of age. Calves were vaccinated at 5 weeks of age with 2 mL Bovi-Shield GOLD FP5L5 intramuscularly and 2 mL One Shot Ultra 7 subcutaneously. At 6 weeks of age, calves were vaccinated with 2 mL TSV-2 and 2 mL Spirovac subcutaneously. At 8 weeks of age, just before being moved to the group housing, calves also were given 2 mL of MAXI/GUARD Pinkeye Bacterin. All vaccines were provided by Pfizer Animal Health (New York, NY), except MAXI/GUARD Pinkeye Bacterin, which was provided by Addison Biological Laboratory, Inc. (Fayette, MO).

Blood samples were obtained at 5 weeks of age (immediately prevaccination), at 7 weeks of age (2 weeks postvaccination), and at 15 weeks of age (10 weeks postvaccination). Blood samples were collected into red top vacutainer (Becton Dickinson, Rutherford, NJ) tubes and allowed to clot at room temperature before centrifuging. Serum was then pipetted into 2-mL microcentrifuge tubes. Samples were frozen and analyzed together for Type 1 and 2 BVD titers by using standard serum neutralization techniques.

Temperature and humidity of each hutch and ambient temperature were monitored continuously with data loggers throughout the study. Rectal temperatures were measured and recorded at 0600, 1100, 1600, and 2100 hours for 3 days during a period of environmental heat stress. Rectal temperatures also were measured and recorded at 0600 and 1600 hours daily for 1 week before and 1 week after vaccination.

## Results and Discussion

During the study, 1 calf died because of causes unrelated to the study. Data from this calf and that of her block mate were removed before analysis. Dry feed intake did not differ significantly between treatments (Figure 1). Calf growth, as measured by weight and frame size (hip height), is shown in Figures 2 and 3. These measurements showed normal calf growth with no significant differences between treatments. Fecal scores averaged 1.2 and 1.3 for shade and sun treatments, respectively, and did not differ between treatments.

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BVD Type 1 and Type 2 titers did not differ significantly between treatments (Figures 4 and 5). Type 1 titers (Figure 4) showed no increase in titer level, but rather a titer decrease typical of maternal colostral antibody depletion. Type 2 titers (Figure 5), however, initially showed maternal antibody decrease similar to Type 1 but followed with an increase in titers, suggesting a delayed active response.

Rectal temperature did not differ between treatments (Figure 6). The temperature rise immediately after vaccination (day 7) was likely caused by the adjuvant in the One Shot *Mannheimia haemolytica* bacterin/toxoid and the 5-way *Leptospira* fraction of the Bovi-Shield GOLD FP5L5, both of which are gram-negative components known to cause a rapid postvaccination temperature response. The modified-live viral fractions of the Bovi-Shield GOLD FP5L5, containing a modified-live virus, should have shown a slower temperature response because the modified-live virus needed time to replicate before it could initiate an immune response and corresponding temperature rise.

Internal hutch temperature and relative humidity (Figure 7) were measured as an average of 8 hutches (4 from each treatment). Average temperature of the sun treatment was greater ( $P < 0.001$ ) than that of the shade treatment (77.2 vs. 75.9°F), whereas the relative humidity was lower ( $P = 0.02$ , 76.5 vs. 76.6%). Typically, a higher temperature will result in lower relative humidity if a similar amount of water is present in the air. The greater temperature observed for the sun treatment resulted in a greater ( $P < 0.001$ ) temperature-humidity index. This indicated that the environment in the sun treatment potentially could have been more stressful than the shade.

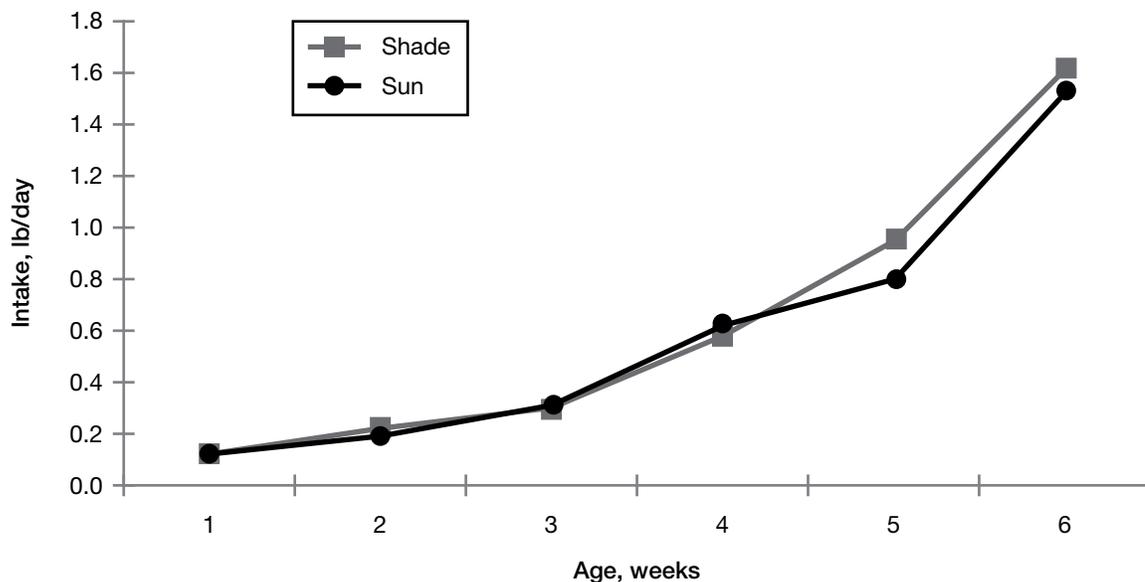


Figure 1. Dry feed intake vs. time.

### FACILITIES AND ENVIRONMENT

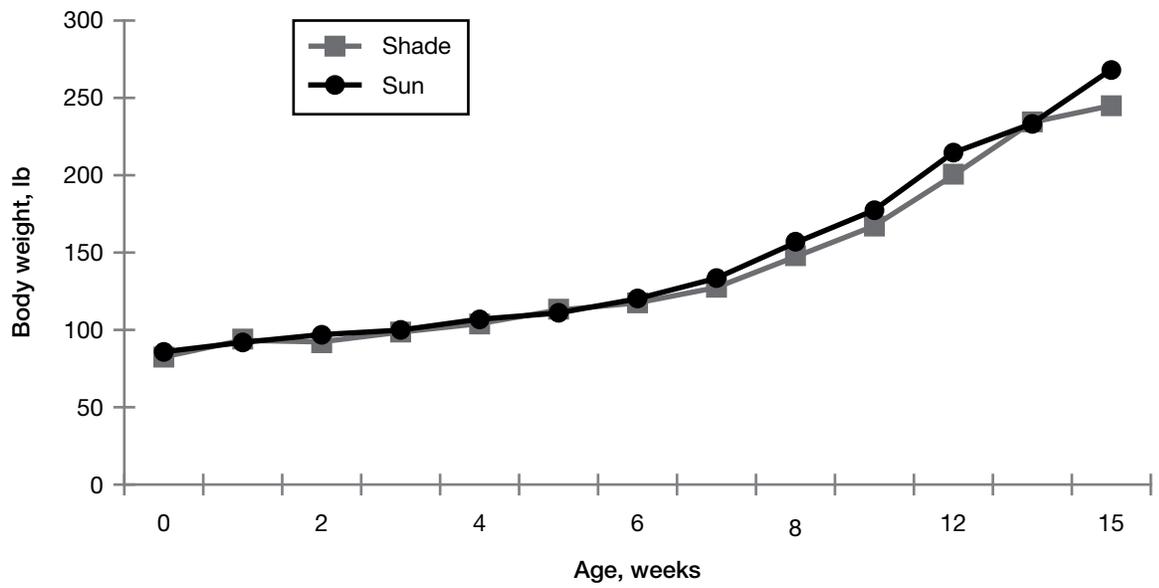


Figure 2. Calf weight vs. time.

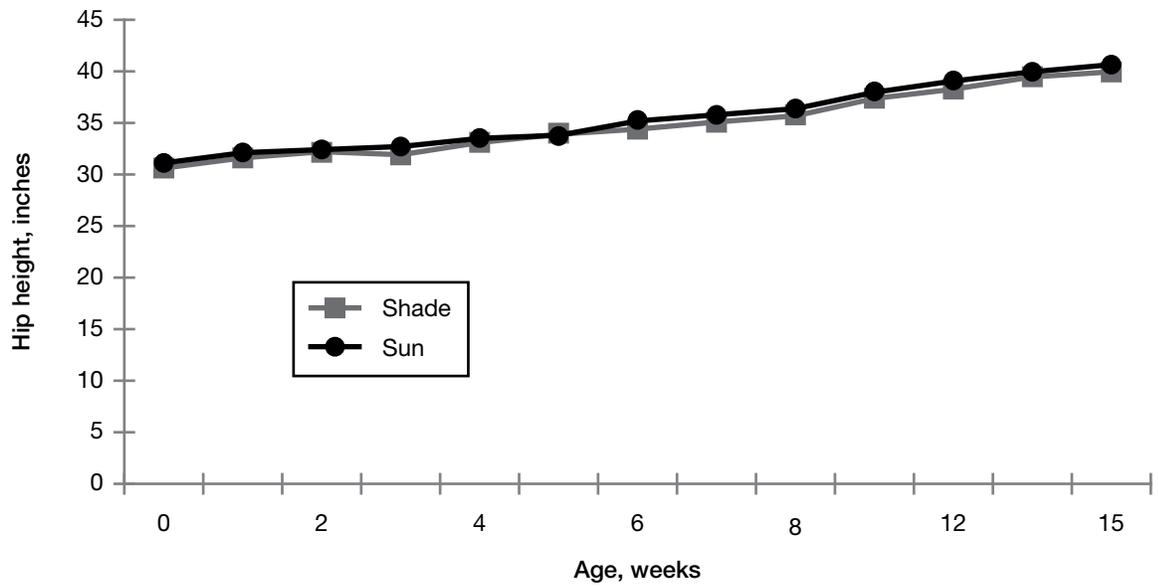


Figure 3. Calf hip height vs. time.

### FACILITIES AND ENVIRONMENT

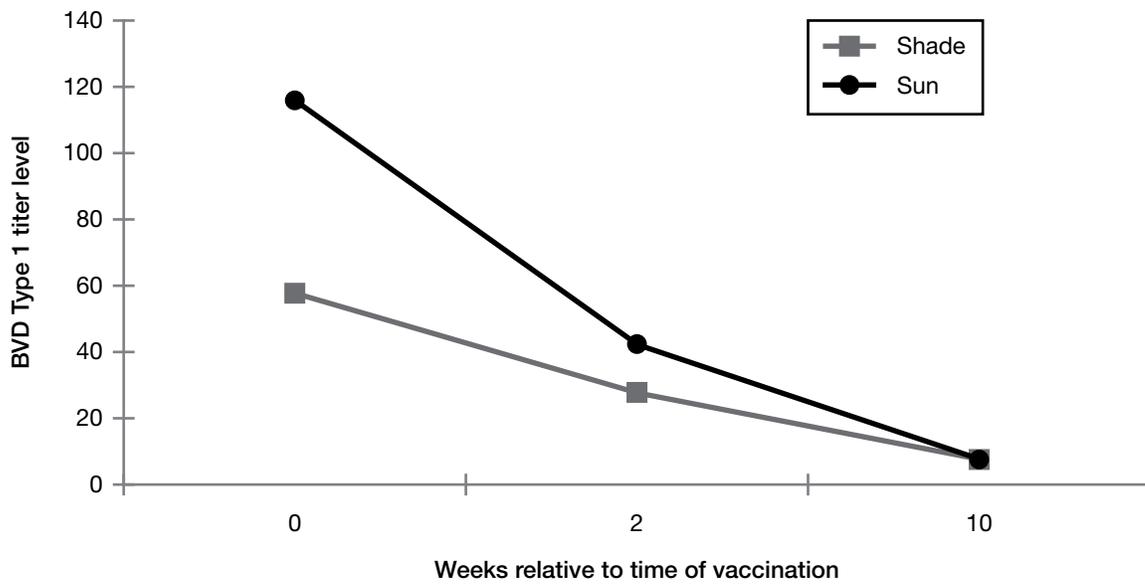


Figure 4. Bovine viral diarrhoea Type 1 titers vs. time.

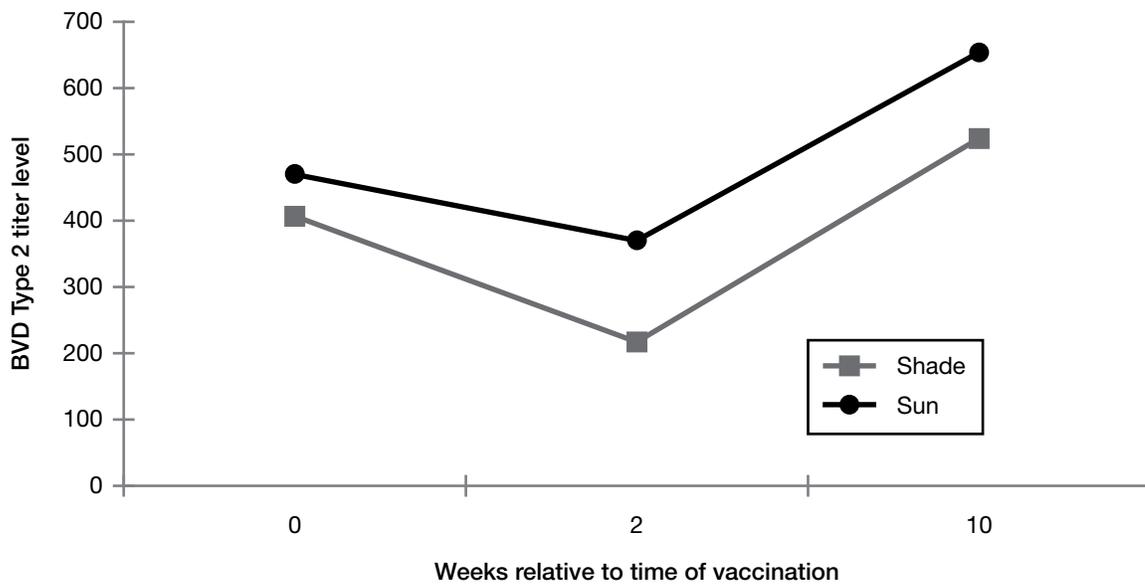


Figure 5. Bovine viral diarrhoea Type 2 titers vs. time.

FACILITIES AND ENVIRONMENT

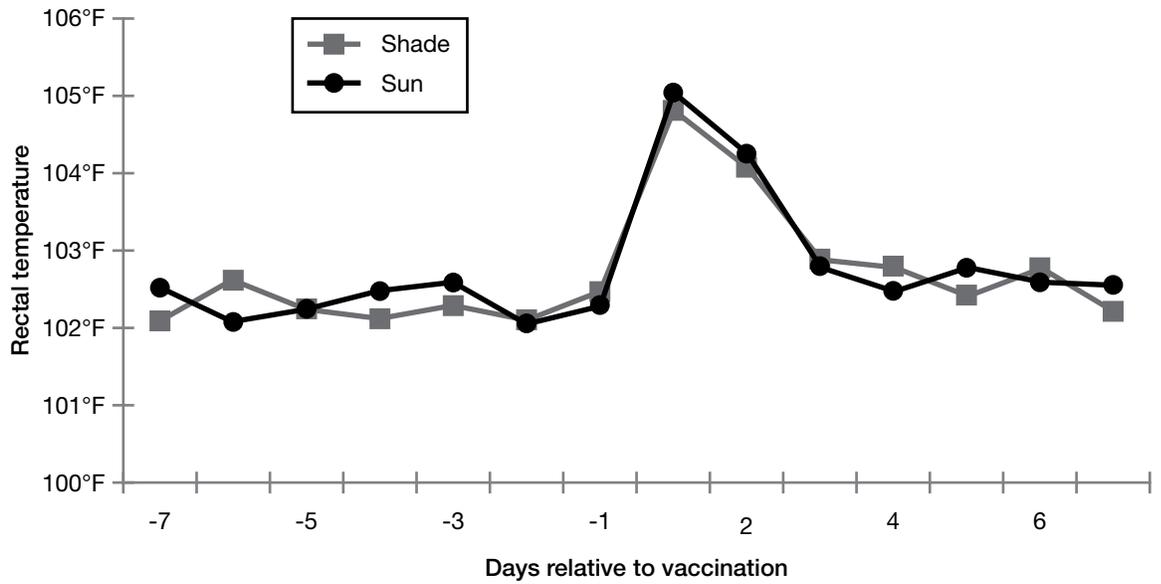


Figure 6. Average rectal temperature 1 week before and 1 week after vaccination.

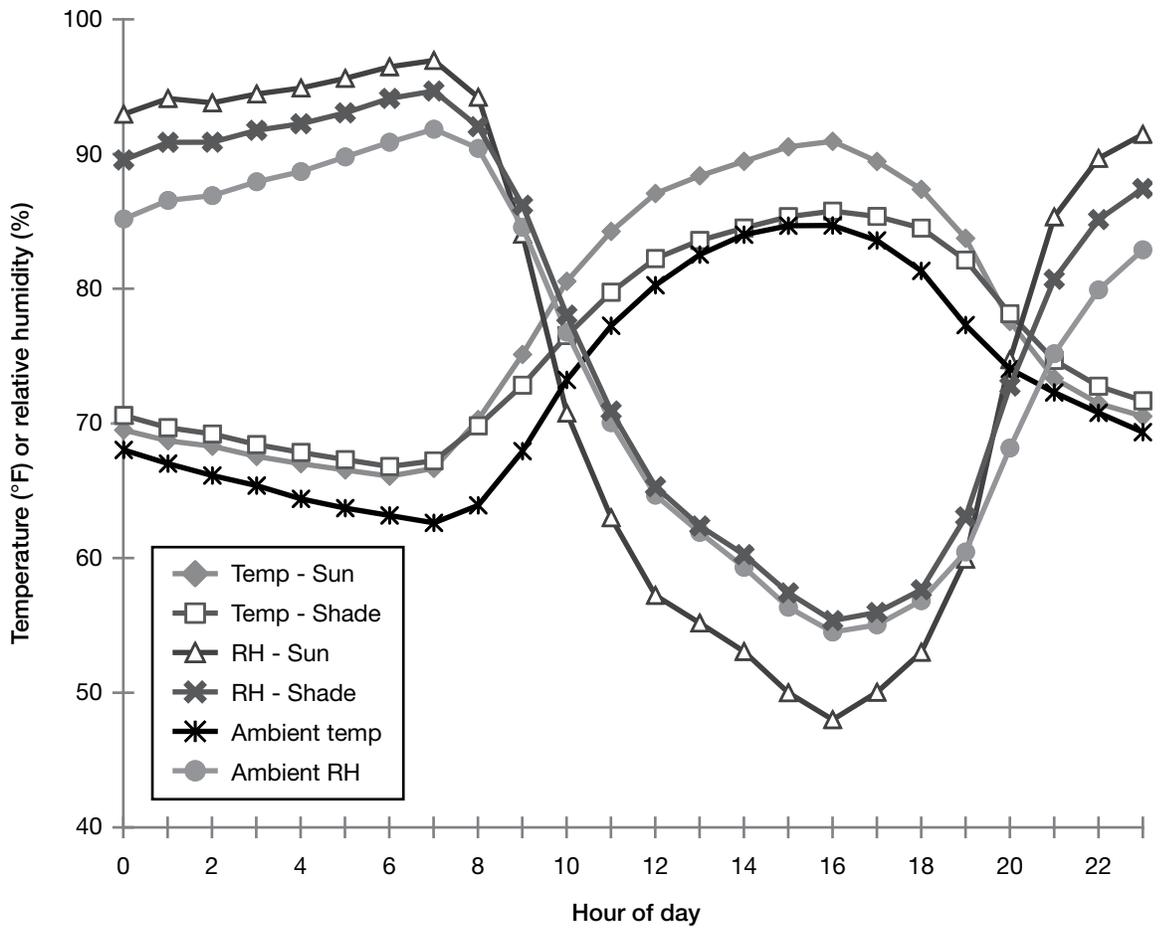


Figure 7. Average calf hutch temperature and relative humidity (July 27 to August 30, 2008).

# Effects of Acidified Fermentation By-Products and Prepartum DCAD on Feed Intake, Performance, and Health of Transition Dairy Cows

*D. J. Rezac and B. J. Bradford*

## Summary

Two commercially available acidified fermentation by-products were evaluated in the close-up period (21 days before expected calving) for their effects on feed intake, postpartum performance, and cow health. Diets were formulated to contain similar protein and energy values but differed in dietary cation anion difference and anion source. Treatments were Bio-Chlor, SoyChlor, and a control. Prepartum feed intake tended to be lower for SoyChlor than for the control, but postpartum intake did not differ among dietary treatment groups. Likewise, no significant differences were detected for milk yield between treatments. Protein percentage, milk urea nitrogen, and lactose percentage were greatest for SoyChlor-treated cows. Therefore, despite a trend for a negative effect on prepartum feed intake, SoyChlor supported similar productivity in early lactation.

## Introduction

Near the time of calving and subsequent onset of lactation, most high-producing dairy cows will experience some degree of hypocalcaemia. Although only clinical hypocalcemia (milk fever) cases are typically detected, ramifications of subclinical hypocalcaemia can reach far into the lactation of the cows affected. Studies show that cows that suffer from hypocalcemia have a much greater susceptibility to other transition disorders such as retained placenta, metritis, mastitis, ketosis, and displaced abomasum. In addition, cows may suffer from decreased milk production and impaired reproductive function long after the transition period has passed.

Preventing hypocalcemia in transition dairy cows has been the subject of much research. Traditional methods used to prevent hypocalcemia have limited calcium intake during the dry period by formulating low-calcium diets. Unfortunately, this strategy proves difficult when diets are formulated with typical feedstuffs found on dairies for transition cow nutrition. Another well-proven and more feasible option is lowering the dietary cation anion difference (**DCAD**) balance  $[(\text{Na} + \text{K}) - (\text{Cl} + \text{S})]$  in the diet during the last 21 days before expected calving. Although it is possible to achieve this goal without using feed additives, this generally does not allow for use of forages grown on the farm because they are high in potassium. To achieve a negative DCAD balance, anionic salts  $[\text{MgSO}_4, \text{MgCl}_2, \text{NH}_4\text{Cl}_2, (\text{NH}_4)_2\text{SO}_4, \text{CaCl}_2, \text{and CaSO}_4]$  have been incorporated in close-up rations. Introducing anions into the prepartum diet creates a mild state of metabolic acidosis that increases the sensitivity of tissues to parathyroid hormone, a key regulator of calcium homeostasis.

In addition to improving postpartum calcium status when fed in the close-up period, anionic salts have been found to decrease feed intake at a time when energy status of the animal is key. The need for a more palatable supplement that still has the ability to decrease DCAD in the diet led to the creation of acidified fermentation by-products (**AFBP**). The AFBP are treated with hydrochloric acid to add chloride ions but are marketed as having fewer negative effects on feed intake than anionic salts.

## Experimental Procedures

The objective of this study was to compare the effects of 2 commercial AFBP on prepartum and postpartum feed intake, milk production, and postpartum health disorders. Diets differed in DCAD and source of anionic supplement. Treatments were a control (CON; DCAD = +20 meq/100 g dry matter; 9 cows and 4 heifers), Bio-Chlor (BC; Arm & Hammer Animal Nutrition, Princeton, NJ; DCAD = -10 meq/100 g dry matter; 8 cows and 6 heifers), and SoyChlor (SC; West Central Cooperative, Ralston, IA; DCAD = -10 meq/100 g dry matter; 9 cows and 6 heifers). Treatment diets (Table 1) were fed ad libitum from 21 days before the expected calving date until parturition. After parturition, all cows received a single fresh cow total mixed ration and were fed for ad libitum intake. Cows remained in a tie-stall facility until 14 days in milk and then were moved to a freestall facility, where they continued to receive the same lactation ration. Milk yield was recorded daily through 21 days in milk, and milk samples were collected daily between 5 and 21 days in milk.

A total of 45 cows and heifers were included in the study; however, 3 (all multiparous cows) were removed because of health events unrelated to treatment. Data were analyzed by using mixed models including the fixed effects of treatment, parity, day relative to parturition, day by treatment interaction, parity by treatment interaction, and day by parity by treatment interaction, as well as the random effect of animal. Prepartum and postpartum feed intake were analyzed separately. Incidence of periparturient disorders was modeled by Fisher's exact test.

## Results and Discussion

### *Feed Intake, Milk Yield, and Composition*

A summary of prepartum and postpartum feed intake (as-fed), milk yield, and milk composition is shown in Table 2. When dietary treatments were fed (the prepartum period), SC tended ( $P = 0.09$ ) to decrease feed intake compared with CON, whereas BC was intermediate. Postpartum intake did not differ among treatments. Treatment means for protein percentage, lactose percentage, and milk urea nitrogen differed ( $P < 0.05$ ), with SC increasing concentrations of all 3 milk components compared with BC.

### *Weight, Body Condition, and Postpartum Health Disorders*

Changes in body weight and body condition score did not differ among treatments but decreased during the transition to lactation (Table 3). Clinical ketosis was identified with urinary ketone reagent strips, and cows that tested moderate or greater on the colorimetric scale for 2 consecutive days were deemed clinical cases. Incidences of displaced abomasum, retained placenta, metritis, mastitis, and milk fever were recorded according to standard clinical definitions. Incidences of all postpartum health disorders did not differ among treatments according to Fisher's exact test (Table 4).

Anionic salts are known to depress feed intake when incorporated into close-up rations, and AFBP are intended to minimize this problem. The SC treatment in this study, however, tended to decrease feed intake by 17% compared with the CON diet, which had a positive DCAD value. Feed intake of cows in the BC treatment was similar that of CON cows but did not differ from that of SC-treated cows. Milk production in early lactation, however, was not adversely affected by the trend for decreased prepartum feed intake in SC-treated cows.

Because even subclinical hypocalcaemia is associated with increased risk of numerous transition cow disorders, feeding a close-up ration with a positive DCAD, like the CON treatment, would

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be expected to cause more problems in early lactation. We did not observe a significant increase in incidence of disorders in this study, but the number of cows was quite small for determining such effects. Future analysis of blood calcium, glucose, and ketone concentrations may provide greater insight into the effects of DCAD treatments on metabolic health in early lactation.

**Table 1. Ingredient composition of experimental diets**

| Ingredient (% of diet dry matter) | Prepartum dietary treatment |           |          | Lactation diet |
|-----------------------------------|-----------------------------|-----------|----------|----------------|
|                                   | Control                     | Bio-Chlor | SoyChlor |                |
| Corn silage                       | 25.2                        | 25.1      | 25.1     | 23.8           |
| Wheat straw                       | 35.9                        | 35.9      | 35.8     | —              |
| Wet corn gluten feed              | 11.0                        | 11.0      | 11.0     | 32.4           |
| Alfalfa hay                       | —                           | —         | —        | 12.1           |
| Rolled corn                       | 10.6                        | 10.5      | 10.6     | 19.4           |
| Soy hulls                         | 4.8                         | 4.5       | 1.6      | 1.8            |
| Soybean meal                      | 9.9                         | 5.2       | 8.5      | —              |
| SoyBest                           | —                           | —         | —        | 6.2            |
| Blood meal                        | 0.8                         | 0.8       | 0.8      | —              |
| Bio-Chlor                         | —                           | 5.3       | —        | —              |
| SoyChlor                          | —                           | —         | 6.1      | —              |
| Molasses                          | 0.4                         | 0.4       | 0.4      | —              |
| Micronutrient premixes            | 1.7                         | 1.7       | 0.5      | 4.2            |

**Table 2. Dietary treatment effects on feed intake and performance**

| Item  | Prepartum dietary treatment |           |           |       | <i>P</i> value |        |       |
|---|-----------------------------|-----------|-----------|-------|----------------|--------|-------|
|   | Control                     | Bio-Chlor | Soy-Chlor | SEM   | Day            | Parity | Diet  |
| Prepartum feed intake, lb/day               | 40.2                        | 37.9      | 33.4      | 2.7   | <0.001         | 0.68   | 0.09  |
| Postpartum feed intake, lb/day <sup>1</sup> | 44.0                        | 46.6      | 50.9      | 5.5   | <0.001         | 0.24   | 0.63  |
| Milk yield, lb/day <sup>2</sup>             | 70.3                        | 74.7      | 76.2      | 5.3   | <0.001         | <0.01  | 0.70  |
| Energy-corrected milk, lb/day               | 77.8                        | 82.8      | 82.0      | 5.1   | 0.08           | <0.001 | 0.74  |
| Fat yield, lb/day                           | 2.91                        | 3.22      | 2.95      | 0.18  | 0.13           | <0.001 | 0.41  |
| Fat, %                                      | 4.33                        | 4.46      | 4.01      | 0.002 | <0.001         | 0.03   | 0.27  |
| Protein yield, lb/day                       | 2.23                        | 2.23      | 2.49      | 0.19  | <0.01          | <0.01  | 0.46  |
| Protein, %                                  | 3.18                        | 2.99      | 3.29      | 0.001 | <0.001         | 0.55   | 0.03  |
| Lactose yield, lb/day                       | 3.35                        | 3.50      | 3.70      | 0.28  | <0.001         | <0.01  | 0.62  |
| Lactose, %                                  | 4.70                        | 4.66      | 4.87      | 0.001 | <0.01          | 0.08   | 0.02  |
| Somatic cell count                          | 162                         | 131       | 107       | 68    | 0.98           | 0.40   | 0.86  |
| Milk urea nitrogen, mg %                    | 8.82                        | 8.57      | 9.11      | 0.12  | <0.001         | 0.19   | <0.01 |

<sup>1</sup>Through 14 days in milk.

<sup>2</sup>Through 21 days in milk.

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**Table 3. Effects of dietary treatment on body weight and body condition score**

| Item                 | Prepartum dietary treatment |           |           | SEM  | <i>P</i> value |        |      |
|----------------------|-----------------------------|-----------|-----------|------|----------------|--------|------|
|                      | Control                     | Bio-Chlor | Soy-Chlor |      | Day            | Parity | Diet |
| Weigh, lb            | 1369                        | 1505      | 1479      | 49   | <0.001         | <0.001 | 0.12 |
| Body condition score | 3.10                        | 3.15      | 3.20      | 0.08 | <0.001         | 0.04   | 0.68 |

**Table 4. Incidence of postpartum health disorders<sup>1</sup>**

| Item               | Prepartum dietary treatment |           |          |
|--------------------|-----------------------------|-----------|----------|
|                    | Control                     | Bio-Chlor | SoyChlor |
| Number of cows     | 13                          | 14        | 15       |
| Ketosis            | 7                           | 6         | 4        |
| Displaced abomasum | 3                           | 1         | 2        |
| Retained placenta  | 2                           | 0         | 1        |
| Metritis           | 1                           | 0         | 0        |
| Mastitis           | 1                           | 1         | 0        |
| Milk fever         | 1                           | 1         | 0        |

<sup>1</sup>No significant treatment effects were detected using Fisher's exact test.

# Dietary Molasses Increases Ruminal pH and Enhances Ruminal Biohydrogenation During Milk Fat Depression

*C. A. Martel, E. C. Titgemeyer, and B. J. Bradford*

## Summary

Molasses has long been used in animal feeds for palatability and as a binding agent to ensure uniform consumption of essential nutrients. Recent work with molasses in highly fermentable diets has revealed that molasses might offer additional benefits in dairy rations. Feeding high-concentrate diets increases the risk of milk fat depression by disrupting the normal pathway of fatty acid biohydrogenation in the rumen. Preliminary research conducted at Kansas State University and other universities has indicated that dietary sugars have the potential to increase milk fat synthesis during milk fat depression. In this study, we sought to understand the reasons for this beneficial effect of molasses on milk fat synthesis. Despite the fact that molasses provides readily fermentable sugar, replacing 5% of dietary corn grain with molasses increased ruminal pH, improved fatty acid biohydrogenation, and shifted the profile of fermentation acids in a manner suggesting that growth of fiber-digesting bacteria was improved. Results of several studies suggest that 5% dietary molasses can increase milk fat yield by 5 to 10%, and the current study indicates that this effect is driven by a stabilization of ruminal pH and biohydrogenation.

## Introduction

Satisfactory milk fat production by lactating dairy cows has great economic value to dairy producers. Fat is the most variable component of milk, and it can be affected by many factors including diet, genetics, physiological state, and environment. Milk fat depression (**MFD**) is a decrease in milk fat yield of up to 50% with no change in milk yield or yield of other milk components. Milk fat depression occurs when dietary factors influence ruminal fermentation and biohydrogenation of unsaturated fatty acids (**FA**), resulting in the production of unique FA that decrease synthesis of milk fat in the mammary gland. Normal biohydrogenation converts linoleic acid (*cis*-9, *cis*-12 C<sub>18:2</sub>) to stearic acid (C<sub>18:0</sub>) using *cis*-9, *trans*-11 conjugated linoleic acid and *trans*-11 C<sub>18:1</sub> FA as intermediates. When MFD occurs, conversion of linoleic acid to stearic acid goes through an alternative pathway that produces *trans*-10, *cis*-12 conjugated linoleic acid, and *trans*-10 C<sub>18:1</sub> FA as intermediates, both of which are capable of inducing MFD.

Because of increasing feed costs, use of by-products like distillers grains has increased during the past few years. Although these by-products can provide less expensive dietary feedstuffs, they are imperfect. Unlike many other corn-derived feedstuffs, distillers grains contain about 10% unsaturated fat, which can impair ruminal biohydrogenation and promote MFD. As a result, few commercial dairies feed more than 10% distillers grains.

A preliminary study conducted at Kansas State University in 2008 found that inclusion of 5% molasses in a high-concentrate diet could decrease production of MFD-inducing FA. We hypothesized that molasses enhances milk fat synthesis by supporting the growth of bacteria responsible for FA biohydrogenation in the rumen, resulting in decreased absorption of FA that induce MFD. In addition to the signs of improved milk fat synthesis in molasses-fed cows, however, our previous study also showed a decrease in milk protein yield with the 5% molasses treatment. Our overall objective in this study was to evaluate the effects of adding molasses to a high-concentrate diet containing 20% distillers grains, with or without supplemental amino acids.

## Experimental Procedures

Six rumen-cannulated, multiparous, late-lactation Holstein cows ( $220 \pm 18$  days in milk) were used to evaluate effects of adding molasses, with or without supplemental amino acids, on ruminal traits and milk composition. The control diet was formulated with the intention of causing MFD and included 37% forage and 20% corn dried distillers grains with solubles, resulting in a diet with 24.5% neutral detergent fiber, 48.7% non-fiber carbohydrate, and 4.7% ether extract (Table 1). Dietary treatments were formulated such that a 5% inclusion rate of cane molasses replaced a portion of the corn grain. Dietary treatments were fed for 28 days, allowing 16 days for diet adaptation and the final 12 days for 2 abomasal infusion periods (Figure 1). In the preliminary study, increasing the inclusion rate of molasses decreased milk protein yield, which suggested that metabolizable protein supply may be inadequate, thus limiting milk protein synthesis. As a result of those findings, either water or the 3 most limiting amino acids were infused into the abomasum to test the effect of additional amino acid supply. Throughout the experiment, cows were housed in a tie-stall facility, milked thrice daily (0500, 1300, and 2100 hours), and fed twice daily (0630 and 1500 hours) for ad libitum intake.

Milk production data and samples were collected during the final 4 days of each infusion period for analysis of milk components and FA profiles (Figure 1). Use of cannulated cows facilitated ruminal digesta collection to assess measures of ruminal fermentation. Ruminal contents were collected at 9-hour intervals on days 26 to 28 of each dietary period, which represented every 3 hours of a 24-hour period. Rumen samples were analyzed for pH as well as volatile FA and ammonia concentrations.

## Results and Discussion

### *Milk Protein*

Our preliminary study showed that increasing the inclusion rate of dietary molasses linearly decreased milk protein yield. This prompted further investigations into the effects of molasses on milk protein. In the present study, essential amino acids were infused into the abomasum to test whether molasses created a metabolizable protein limitation that could be overcome with additional amino acids. Amino acid infusions had no interaction with molasses for milk protein content or yield (Table 2). In addition, molasses had no direct effect on milk protein content or yield. Therefore, these results do not support previous findings that dietary molasses negatively affects milk protein yield during MFD.

### *Productivity and Milk Fat*

Consistent with previous findings, 5% molasses had no effect on dry matter intake during MFD (Table 2). Molasses tended ( $P = 0.06$ ) to decrease body weight gain compared with the control diet ( $-7$  vs.  $+35 \pm 20$  lb during 28 days). Dietary molasses increased ( $P < 0.01$ ) milk fat concentration with no significant effect on milk yield. Neither content nor yield of protein, lactose, or urea nitrogen was changed by addition of molasses.

To further understand the role of molasses in FA biohydrogenation, milk FA analysis was conducted. Dietary molasses decreased the yield of *trans*-10 C<sub>18:1</sub> and increased ( $P < 0.01$ ) the yield of *trans*-11 C<sub>18:1</sub> in milk, key intermediates in alternative and normal biohydrogenation pathways, respectively (Table 3). Shifts in FA profiles indicated that dietary molasses had a positive influence on biohydrogenation by promoting normal biohydrogenation and decreased use of the alternative pathway.

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***Ruminal Metabolism***

Analysis of ruminal fluid showed that dietary molasses increased ( $P < 0.01$ ) pH and decreased total volatile FA concentration in the rumen (Table 4). Molasses increased ( $P < 0.04$ ) molar proportions of acetate and butyrate but decreased ( $P < 0.01$ ) the proportion of propionate in ruminal fluid.

Replacing 5% dietary corn with cane molasses (on a dry matter basis) in a low-forage, high-concentrate diet increased ruminal pH, promoted the normal pathway of ruminal biohydrogenation, and decreased the production of a key MFD-inducing FA. Dietary molasses increased milk fat content in a diet used to induce MFD without changing milk component yield. Considered along with our prior results, these data indicate that molasses has the potential to mildly increase milk fat yield by promoting normal ruminal biohydrogenation and decreasing absorption of MFD-inducing FA.

**Table 1. Diet composition (% of dry matter)**

| Item                                       | Dietary molasses, % |      |
|--|---------------------|------|
|  | 0                   | 5    |
| Ingredient                                 |                     |      |
| Corn silage                                | 25.5                | 25.5 |
| Alfalfa hay                                | 12.6                | 12.6 |
| Corn dried distillers grains with solubles | 19.8                | 19.8 |
| Ground corn grain                          | 33.6                | 28.8 |
| Molasses                                   | —                   | 4.8  |
| Soybean meal                               | 4.1                 | 4.1  |
| Expeller soybean meal                      | 2.7                 | 2.7  |
| Limestone                                  | 1.1                 | 1.1  |
| Trace mineral salt                         | 0.4                 | 0.4  |
| Micronutrient premix                       | 0.2                 | 0.2  |
| Nutrient                                   |                     |      |
| Dry matter, % as is                        | 66.4                | 65.8 |
| Crude protein                              | 16.2                | 16.1 |
| Neutral detergent fiber                    | 24.5                | 24.5 |
| Non-fiber carbohydrate                     | 48.7                | 48.6 |
| Starch                                     | 36.3                | 32.9 |
| Sugars (by invertase)                      | 6.4                 | 8.9  |
| Ether extract                              | 4.7                 | 4.6  |
| Ash  | 5.9                 | 6.2  |

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**Table 2. Effects of treatment on productivity of lactating dairy cows**

| Item                          | Infusion         |      |             |      | SEM  | <i>P</i> value <sup>1</sup> |      |
|-------------------------------|------------------|------|-------------|------|------|-----------------------------|------|
|                               | Water            |      | Amino acids |      |      | Molasses                    | AA   |
|                               | Dietary molasses |      |             |      |      |                             |      |
|                               | 0%               | 5%   | 0%          | 5%   |      |                             |      |
| Dry matter intake, lb/day     | 54.0             | 53.8 | 52.7        | 54.2 | 2.9  | 0.41                        | 0.66 |
| Milk yield, lb/day            | 62.4             | 63.1 | 63.9        | 61.9 | 6.2  | 0.70                        | 0.86 |
| Solids-corrected milk, lb/day | 53.4             | 54.9 | 55.1        | 54.7 | 5.7  | 0.67                        | 0.53 |
| Energy-corrected milk, lb/day | 58.6             | 60.2 | 60.6        | 60.2 | 6.2  | 0.18                        | 0.45 |
| Milk fat, %                   | 2.68             | 2.90 | 2.75        | 2.98 | 0.21 | 0.01                        | 0.23 |
| Milk protein, %               | 3.45             | 3.37 | 3.44        | 3.44 | 0.13 | 0.41                        | 0.37 |
| Milk lactose, %               | 4.91             | 4.92 | 4.93        | 4.90 | 0.08 | 0.74                        | 0.99 |
| Milk fat, lb/day              | 1.74             | 1.83 | 1.80        | 1.87 | 0.24 | 0.25                        | 0.49 |
| Milk protein, lb/day          | 2.14             | 2.09 | 2.16        | 2.12 | 0.20 | 0.43                        | 0.73 |
| Milk lactose, lb/day          | 3.11             | 3.13 | 3.13        | 3.08 | 0.31 | 0.81                        | 0.89 |
| Milk urea nitrogen, mg/dL     | 11.6             | 11.2 | 11.4        | 11.3 | 0.7  | 0.30                        | 0.97 |

<sup>1</sup> Molasses = effect of dietary molasses; AA = effect of amino acid infusion. All molasses × AA effects were nonsignificant for reported variables (*P* > 0.15).

**Table 3. Effects of treatment on yield of selected milk fatty acids**

| Fatty acid, g/day                                 | Infusion         |      |             |      | SEM  | <i>P</i> value <sup>1</sup> |      |
|---|------------------|------|-------------|------|------|-----------------------------|------|
|   | Water            |      | Amino acids |      |      | Molasses                    | AA   |
|   | Dietary molasses |      |             |      |      |                             |      |
|   | 0%               | 5%   | 0%          | 5%   |      |                             |      |
| <i>trans</i> -10 C <sub>18:1</sub>                | 16.8             | 10.9 | 12.8        | 10.4 | 3.3  | 0.01                        | 0.15 |
| <i>trans</i> -11 C <sub>18:1</sub>                | 12.2             | 14.7 | 12.3        | 14.1 | 3.0  | <0.01                       | 0.76 |
| total <i>trans</i> C <sub>18:1</sub>              | 32.3             | 28.7 | 27.8        | 27.8 | 3.0  | 0.28                        | 0.13 |
| <i>trans</i> -10, <i>cis</i> -12 CLA <sup>2</sup> | 0.19             | 0.18 | 0.18        | 0.15 | 0.03 | 0.37                        | 0.42 |

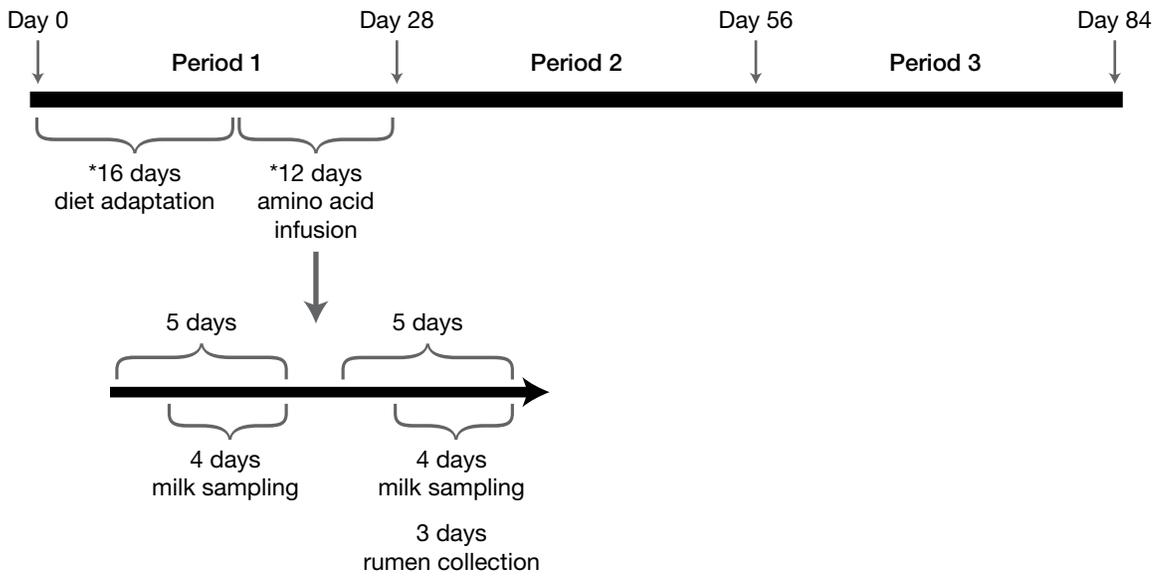
<sup>1</sup> Molasses = effect of dietary molasses; AA = effect of amino acid infusion. All molasses × AA effects were nonsignificant for reported variables (*P* > 0.15).

<sup>2</sup> Conjugated linoleic acid.

**Table 4. Effects of molasses inclusion rate on measures of ruminal fermentation**

| Item                           | Dietary molasses, % |       | SEM  | <i>P</i> value |
|--------------------------------|---------------------|-------|------|----------------|
|                                | 0                   | 5     |      |                |
| Ruminal pH                     | 5.73                | 5.87  | 0.06 | 0.02           |
| Total volatile fatty acids, mM | 140.8               | 132.7 | 4.6  | <0.01          |
| Acetate, mol/100 mol           | 46.3                | 46.9  | 0.9  | 0.04           |
| Propionate, mol/100 mol        | 28.7                | 27.4  | 1.4  | 0.01           |
| Butyrate, mol/100 mol          | 16.7                | 17.7  | 1.0  | 0.04           |
| Valerate, mol/100 mol          | 4.9                 | 4.9   | 0.3  | 0.78           |
| Ammonia, mg/dL                 | 7.86                | 7.35  | 0.88 | 0.32           |

### NUTRITION AND FEEDING



**Figure 1. Experimental protocol.**

Each period included 16 days of diet adaptation and 12 days of amino acid infusion and data collection.

# Effects of Feeding Increased Amounts of Wet Corn Gluten Feed on Dairy Cow Metabolism and Milk Production

*C. R. Mullins, K. N. Grigsby, D. E. Anderson, E. C. Titgemeyer, and B. J. Bradford*

## Summary

An experiment was conducted to evaluate the effects of feeding increasing dietary amounts of wet corn gluten feed (**WCGF**). Eight lactating Holstein cows were housed in a tie-stall facility and fed 1 of 4 diets containing 0, 11, 23, or 34% WCGF on a dry matter basis. To maintain similar nutrient concentrations, alfalfa hay, corn silage, corn grain, soybean meal, expeller soybean meal, and mineral supplements varied across diets. Feed intake, milk production, body weight, and body condition score were monitored, and effects of WCGF inclusion rate were assessed. Increasing the level of WCGF in the diet led to increased feed intake, milk production, and body condition. Concentrations of milk components did not differ among treatments; therefore, yield of energy-corrected milk and solids-corrected milk increased as well. In contrast, increasing dietary WCGF decreased efficiency of production and also decreased ruminal pH, possibly because treatments with greater proportions of WCGF had a decreased mean particle size. As expected, the decreased ruminal pH coincided with changes in ruminal volatile fatty acid concentrations. Furthermore, the rate of fiber digestion after 24 hours decreased when more WCGF was incorporated into diets. Results indicate that adding WCGF to dairy rations can increase energy-corrected milk yield, and this increase seems to be driven, at least in part, by an increase in feed intake.

## Introduction

The large demand for cereal grains for purposes beyond feeding livestock has contributed to rising feed costs. Because many rations are formulated on a least-cost basis, researchers and producers are pressured to devise novel strategies to help keep feed costs in check. Recently, focus has turned to the use of milling coproducts, particularly wet corn gluten feed (**WCGF**). Wet corn gluten feed is a rapidly digestible non-forage source of fiber and protein. Feeding WCGF to dairy cattle can be a low-cost method of providing energy and nutrients needed for milk production.

Defining the optimum amount of WCGF to incorporate into a diet is complex because this substance interacts with other feed ingredients. Formulating diets to complement the characteristics of WCGF, rather than substituting WCGF for a single ingredient, will increase the likelihood of optimizing its use in lactation diets. The physically effective neutral detergent fiber (**peNDF**) value of WCGF is very low; therefore, sources of peNDF must be included when evaluating feedstuffs to complement WCGF. Rumination is stimulated when peNDF is provided, leading to longer chewing times, greater saliva production, and normal ruminal pH values. If the total mixed diet does not provide adequate peNDF, rumen health may be compromised and milk fat depression can occur.

The objective of this study was to feed increasing amounts of WCGF to lactating dairy cows and monitor effects on production traits. We also measured the effects of WCGF on the rumen environment and the rate of ruminal fiber digestion.

## Experimental Procedures

Eight lactating cows averaging 90 days in milk were housed in a tie-stall facility and fed 1 of 4 diets that contained 0, 11, 23, or 34% WCGF (Sweet Bran; Cargill, Inc., Blair, NE) on a dry matter basis. Nutrient composition of the WCGF used in this study is shown in Table 1. This study was designed so each cow received all 4 treatment diets, allowing us to evaluate individual responses to diets. Alfalfa hay, corn silage, corn grain, soybean meal, expeller soybean meal, and mineral supplements varied among diets so that similar nutrient concentrations were maintained. Ingredients and nutrient compositions of diets are shown in Table 2.

Cows were fed a total mixed ration (**TMR**) twice daily, and amounts fed and refused were recorded daily for each cow during each of the four 28-day periods. Feed samples of individual ingredients were collected for analysis on days 25 to 28. Cows were milked thrice daily, and milk yield was recorded. Milk samples were collected from all milkings on days 25 to 28, and these samples were used to determine milk composition. Body weight and body condition score were measured at the beginning and end of each period. Particle size of the TMR and refusals were measured with the Penn State Particle Separator. Rumen samples and ruminal pH data were collected every 9 hours from days 26 to 28 so that 8 samples were collected from each cow during each period, representing every 3 hours of a 24-hour period to account for diurnal variation. Soybean hulls were incubated in Dacron bags suspended in the rumen on days 21 to 24 of each period to assess dietary effects on rate of ruminal fiber digestion.

## Results and Discussion

### *Feed Intake and Milk Production*

As the inclusion rate of WCGF increased, feed intake increased ( $P = 0.03$ ) from 58.9 to 65.5 lb/day (Table 3). Because intake is influenced by specific gravity of the feedstuffs, particle size, and rate of fermentation, it is no surprise that intake increased in diets with greater concentrations of WCGF.

Milk yield increased ( $P = 0.007$ ) from 81.1 to 85.8 lb/day as greater proportions of WCGF were offered (Table 3); however, concentrations of milk components were not affected ( $P > 0.13$ ; Table 4). Because of the increases in milk yield, there were greater yields of fat, protein, and lactose as WCGF was added (Table 4). The increases in milk and milk component yields from increased WCGF led to greater ( $P < 0.01$ ) solids-corrected milk and energy-corrected milk production (Table 3). The total energy being used for productivity, which includes production of milk and changes in body condition score, increased as WCGF inclusion rate increased ( $P < 0.001$ ; Figure 1). This energy increase is not surprising because feed intake was greater for these cows, which likely resulted in greater consumption of energy. Feed efficiency, measured as energy-corrected milk yield divided by intake, declined ( $P = 0.007$ ) as more WCGF was added (Table 3). Treatments did not affect energy efficiency, measured as total net energy for productive use over intake. Therefore, when milk production efficiency decreased in diets with greater WCGF, energy was not lost, rather it contributed to increased body condition. Body condition score increased ( $P < 0.02$ ) as greater proportions of WCGF were fed (Table 3). Body weight change was not affected by treatment and did not correspond with changes noted in body condition, likely because diets led to differences in gut fill, which would affect measured weight change.

Substituting WCGF for portions of corn silage, alfalfa, corn grain, and soybean meal kept dietary neutral detergent fiber (**NDF**) values similar ( $\approx 30\%$ ), but the effectiveness of that fiber

can be questioned. Table 5 shows the particle size data of the TMR and Orts. All 4 diets had a relatively small mean particle size. In all diets, the mean percentage of particles >19 mm was about 3%. This proportion of particle size is small, but it is within current recommendations for a lactating cow TMR. In contrast, according to these same guidelines, all 4 treatment diets contained an insufficient proportion of particles between 8 and 19 mm. Comparing particle sizes of the refusals with that of the TMR showed no differences in the 3 fractions tested, suggesting that cows did not sort feed components.

An adequate supply of long particles is necessary for healthy rumen function and maintenance of ruminal pH. Diets lacking long particles are generally more fermentable, which can lead to greater acid production; this is a potential explanation for the decrease ( $P < 0.001$ ) in ruminal pH as WCGF was added to the diet (Table 6). Excessive acid production is often attributed to starch in dairy rations; however, the diet with the lowest pH also had the lowest starch concentration, suggesting that acid production may not be related to dietary starch content. In contrast, highly fermentable fiber (i.e., WCGF) also replaced less-fermentable forage fiber, so total diet fermentability may have increased with WCGF additions. Even though adding WCGF depressed ruminal pH, milk fat production was not adversely affected. This suggests the diet with 34% WCGF still provided enough effective fiber to maintain rumen function and promote ruminal biohydrogenation.

### ***Ruminal Metabolism***

Measures of ruminal fermentation are presented in Table 6. As expected, the lower pH observed as WCGF was added coincided with decreased ( $P < 0.001$ ) ruminal acetate and isovalerate concentrations and increased ( $P < 0.001$ ) propionate and valerate concentrations. Differences in diet particle size or ruminal fiber digestibility may account for these effects. Quadratic effects were detected for concentrations of total volatile fatty acids (VFA) (trend:  $P < 0.09$ ) and ammonia ( $P < 0.01$ ); cows fed 0 and 34% WCGF tended to have greater overall VFA and ammonia concentrations. In general, this study shows that WCGF significantly affected the VFA profile.

### ***Rate of Fiber Digestion***

Soybean hulls have a highly digestible fiber fraction and minimal associative effects; therefore, they were used to measure rate of fiber digestion in the rumen as influenced by each diet. Digestibility of soybean hulls showed a significant diet by time interaction (Figure 2;  $P < 0.001$ ). Increasing WCGF quadratically affected ( $P < 0.001$ ) NDF disappearance at 24 hours (Table 6), showing the diet with 23% WCGF to have the lowest disappearance of soybean hull NDF. There was no correlation ( $R^2 = 0.0002$ ) between pH and 24-hour NDF disappearance, suggesting that pH does not seem to be the primary cause of change in fiber digestion. It is unclear how increasing WCGF inclusion negatively affected rate of NDF digestion.

Results from this study demonstrate responses to WCGF that are consistent with recently published research. However, rather than indicating that WCGF improves the rumen environment for fiber-digesting bacteria, production responses to WCGF in this study seem to have been driven by increased feed intake. As a whole, adding WCGF to dairy rations will likely increase milk yield; however, this increase in production is driven, at least in part, by an increase in feed intake.

NUTRITION AND FEEDING

**Table 1. Nutrient composition of wet corn gluten feed used in experiment<sup>1</sup>**

| Nutrient                | % of dry matter | Standard deviation |
|-------------------------|-----------------|--------------------|
| Dry matter (% as-fed)   | 56.1            | 0.9                |
| Crude protein           | 24.5            | 0.4                |
| Neutral detergent fiber | 35.3            | 1.1                |
| Acid detergent fiber    | 11.0            | 0.9                |
| Ether extract           | 2.3             | 0.2                |
| Starch                  | 11.2            | 0.5                |
| Ash                     | 5.8             | 0.4                |

<sup>1</sup>Samples collected on days 25 to 28 of all 4 periods.

**Table 2. Ingredient and nutrient compositions (% of dry matter) of diets containing increasing amounts of wet corn gluten feed (WCGF)**

| Item                              | Dietary WCGF |      |      |      |
|-----------------------------------|--------------|------|------|------|
|                                   | 0%           | 11%  | 23%  | 34%  |
| <b>Ingredient</b>                 |              |      |      |      |
| WCGF <sup>1</sup>                 | 0.0          | 11.4 | 23.2 | 33.6 |
| Corn silage                       | 25.2         | 25.5 | 22.1 | 18.4 |
| Alfalfa                           | 24.4         | 24.6 | 21.2 | 17.7 |
| Cottonseed                        | 6.1          | 6.2  | 6.2  | 6.1  |
| Corn grain                        | 23.5         | 19.9 | 17.3 | 14.6 |
| Soybean meal                      | 8.6          | 4.9  | 2.2  | 2.2  |
| Molasses                          | 0.4          | 0.4  | 0.4  | 0.4  |
| Expeller soybean meal             | 3.3          | 3.7  | 4.0  | 3.6  |
| Soybean hulls                     | 5.0          | —    | —    | —    |
| Limestone                         | 1.00         | 1.08 | 1.28 | 1.36 |
| Magnesium oxide                   | 0.26         | 0.24 | 0.21 | 0.17 |
| Micronutrient premix <sup>2</sup> | 1.33         | 1.32 | 1.33 | 1.31 |
| <b>Nutrient</b>                   |              |      |      |      |
| Dry matter, % as fed              | 65.4         | 60.0 | 61.3 | 61.2 |
| Crude protein (CP)                | 19.3         | 18.8 | 19.1 | 20.1 |
| Rumen degradable protein, % of CP | 63.5         | 65.3 | 63.9 | 66.6 |
| Neutral detergent fiber           | 28.8         | 28.8 | 30.4 | 31.0 |
| Starch                            | 24.3         | 27.9 | 25.5 | 24.2 |
| Non-fiber carbohydrate            | 39.1         | 40.9 | 38.6 | 37.6 |
| Ether extract                     | 3.4          | 3.3  | 3.6  | 3.6  |
| Ash                               | 9.4          | 8.3  | 8.3  | 7.7  |

<sup>1</sup>Wet corn gluten feed; Sweet Bran, Cargill, Inc., Blair, NE.

<sup>2</sup>Premix consists of 61.0% sodium bicarbonate, 27.3% trace mineral salt, 3.90% 4-plex, 3.90% Se premix, 2.60% vitamin E, 1.30% vitamin A, and 0.21% vitamin D.

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**Table 3. Effects of dietary wet corn gluten feed (WCGF) on performance of lactating cows**

| Item                                | Dietary WCGF <sup>1</sup> |      |      |      | SEM  | P value |           |
|-------------------------------------|---------------------------|------|------|------|------|---------|-----------|
|                                     | 0%                        | 11%  | 23%  | 34%  |      | Linear  | Quadratic |
| Dry matter intake, lb/day           | 58.9                      | 57.1 | 64.6 | 65.5 | 3.4  | 0.03    | 0.55      |
| Milk, lb/day                        | 81.1                      | 81.6 | 88.4 | 85.8 | 5.7  | 0.007   | 0.28      |
| Solids-corrected milk, lb/day       | 77.6                      | 78.7 | 84.9 | 82.0 | 5.6  | 0.01    | 0.19      |
| Energy-corrected milk, lb/day       | 84.2                      | 85.5 | 91.9 | 89.1 | 6.2  | 0.01    | 0.19      |
| Efficiency <sup>1</sup>             | 1.44                      | 1.50 | 1.34 | 1.29 | 0.06 | 0.007   | 0.20      |
| Body weight change, lb/28 days      | 100                       | 31   | 20   | 65   | 38.8 | 0.65    | 0.73      |
| Body condition score change/28 days | -0.02                     | 0.09 | 0.15 | 0.25 | 0.07 | 0.02    | 0.92      |

<sup>1</sup> Measured as energy-corrected milk divided by feed intake.

**Table 4. Effects of dietary wet corn gluten feed (WCGF) on milk components**

|  | Dietary WCGF |      |      |      | SEM  | P value |           |
|--|--------------|------|------|------|------|---------|-----------|
|  | 0%           | 11%  | 23%  | 34%  |      | Linear  | Quadratic |
| Milk fat, %                              | 3.65         | 3.76 | 3.72 | 3.67 | 0.11 | 0.93    | 0.23      |
| Milk protein, %                          | 3.02         | 3.07 | 3.05 | 3.11 | 0.08 | 0.13    | 0.80      |
| Milk lactose, %                          | 5.02         | 5.00 | 5.03 | 5.01 | 0.03 | 0.94    | 0.96      |
| Somatic cell count, <sup>1</sup> 1000/mL | 40.6         | 64.1 | 31.9 | 50.2 | 14.8 | 0.96    | 0.87      |
| Urea nitrogen, mg/dL                     | 17.2         | 16.3 | 16.3 | 17.3 | 0.90 | 0.83    | 0.08      |
| Yield, lb/day                            |              |      |      |      |      |         |           |
| Milk fat                                 | 3.02         | 3.06 | 3.28 | 3.17 | 0.24 | 0.06    | 0.21      |
| Milk protein                             | 2.45         | 2.51 | 2.67 | 2.67 | 0.18 | 0.01    | 0.49      |
| Milk lactose                             | 4.18         | 4.08 | 4.45 | 4.30 | 0.29 | 0.01    | 0.32      |

<sup>1</sup> Three outliers were removed.

**Table 5. Effects of dietary wet corn gluten feed (WCGF) on particle size of diets and feed refusals (% as-fed basis)<sup>1</sup>**

| Sample                     | Size                    | Dietary WCGF      |                   |                   |                   |
|----------------------------|-------------------------|-------------------|-------------------|-------------------|-------------------|
|                            |                         | 0%                | 11%               | 23%               | 34%               |
| Total mixed ration         | >19 mm                  | 3.9 <sup>a</sup>  | 3.3 <sup>ab</sup> | 3.0 <sup>ab</sup> | 2.4 <sup>b</sup>  |
|                            | 19 to 8 mm              | 29.6 <sup>a</sup> | 29.6 <sup>a</sup> | 27.2 <sup>b</sup> | 24.2 <sup>c</sup> |
|                            | <8 mm                   | 66.6 <sup>a</sup> | 67.2 <sup>a</sup> | 69.9 <sup>b</sup> | 73.4 <sup>c</sup> |
| Feed refusals <sup>2</sup> | >19 mm                  | 4.2               | 5.1               | 3.9               | 1.5               |
|                            | 19 to 8 mm <sup>3</sup> | 27.4              | 30.0              | 30.6              | 23.7              |
|                            | <8 mm <sup>3</sup>      | 70.6              | 64.9              | 65.5              | 74.7              |

<sup>abc</sup> Means within row without a common superscript letter differ ( $P < 0.05$ ).

<sup>1</sup> Measured using a 3-compartment Penn State Particle Separator.

<sup>2</sup> No significant differences were detected ( $P > 0.15$ ) between the total mixed ration and feed refusals for each fraction across dietary treatments.

<sup>3</sup> One outlier was removed.

NUTRITION AND FEEDING

**Table 6. Effects of dietary wet corn gluten feed (WCGF) on rumen environment**

| Item                              | Dietary WCGF |       |       |       | SEM  | <i>P</i> value |           |
|-----------------------------------|--------------|-------|-------|-------|------|----------------|-----------|
|                                   | 0%           | 11%   | 23%   | 34%   |      | Linear         | Quadratic |
| Total VFA <sup>1</sup> , mM       | 168.6        | 163.8 | 160.1 | 165.0 | 4.5  | 0.26           | 0.09      |
| Acetate, mM                       | 97.2         | 90.6  | 87.1  | 84.6  | 2.4  | <0.001         | 0.15      |
| Propionate, mM                    | 34.4         | 37.8  | 36.8  | 43.1  | 1.3  | <0.001         | 0.15      |
| Butyrate, mM                      | 25.9         | 26.0  | 25.3  | 26.9  | 1.1  | 0.32           | 0.43      |
| Isobutyrate, mM                   | 2.10         | 2.11  | 2.08  | 2.16  | 0.08 | 0.59           | 0.60      |
| Valerate, mM                      | 4.38         | 4.76  | 4.90  | 5.84  | 0.26 | <0.001         | 0.02      |
| Isovalerate, mM                   | 3.45         | 3.36  | 3.11  | 2.56  | 0.31 | 0.001          | 0.16      |
| Ammonia, mM                       | 16.2         | 13.1  | 12.9  | 15.7  | 1.2  | 0.69           | 0.01      |
| Ruminal pH                        | 6.18         | 6.12  | 6.14  | 5.91  | 0.06 | 0.001          | 0.07      |
| 24-hour in situ                   |              |       |       |       |      |                |           |
| NDF <sup>2</sup> disappearance, % | 58.0         | 48.0  | 37.6  | 46.4  | 2.64 | <0.001         | <0.001    |

<sup>1</sup> Volatile fatty acid.

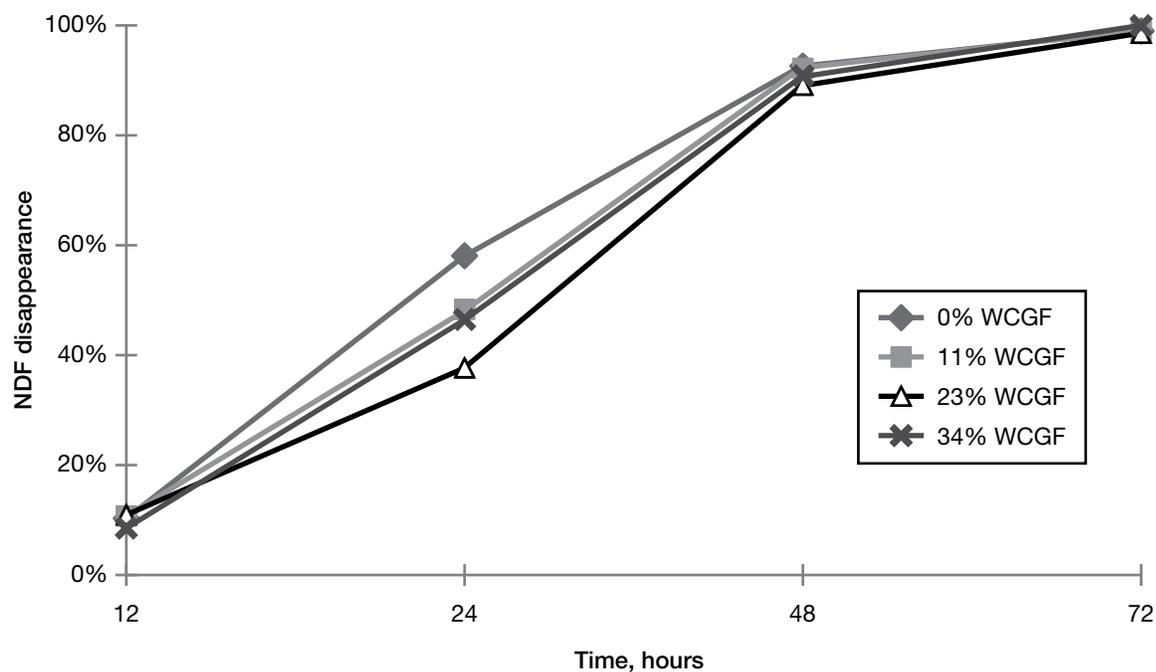
<sup>2</sup> Neutral detergent fiber.



**Figure 1. Total energy partitioned to milk production and body condition score (BCS) change in cows fed increasing amounts of WCGF.**

As WCGF was added, total productive energy increased ( $P < 0.001$ ) linearly. Body condition score loss was assigned an energetic value of 368 Mcal/unit, and BCS gain was assigned 459 Mcal/unit (National Research Council, 2001, Natl. Acad. Sci., Washington, DC.). Milk energy was calculated according to the equation: [Milk energy =  $(41.63 \times \% \text{ fat}) + (24.13 \times \% \text{ protein}) + (21.60 \times \% \text{ lactose}) - 11.72$ ].

### NUTRITION AND FEEDING



**Figure 2. In situ neutral detergent fiber (NDF) disappearance of soybean hulls in diets with increasing amounts of wet corn gluten feed (WCGF) measured over a 72-hour period.**

A time by treatment interaction ( $P < 0.001$ ) was detected for dry matter digestibility. After 24 hours of ruminal degradation, a quadratic effect ( $P < 0.001$ ) was detected for in situ NDF disappearance of soybean hulls. The quadratic response was driven by lower disappearance in the diet with 23% WCGF.

# Effects of Encapsulated Niacin on Metabolism and Production of Periparturient Holstein Cows

*S. D. Morey, B. J. Bradford, L. K. Mamedova, and D. E. Anderson*

## Summary

Niacin (nicotinic acid) can suppress lipolysis, but responses to dietary niacin have been inconsistent in cattle. A widely used commercial feed additive, niacin is thought to reduce heat stress and decrease postpartum plasma nonesterified fatty acid (NEFA) concentration. Raw niacin has poor stability in the rumen, however, and it is estimated that only 5% is bioavailable. Recently, an encapsulated niacin (EN) product with an estimated 40% bioavailability became commercially available, but its effects on health and metabolism in transition cows have not been tested previously. Twenty-two Holstein cows were used in a study beginning 21 days before expected calving; cows were assigned to the EN treatment (24 g/day) or control group until 21 days postpartum. Results showed that EN decreased peak plasma NEFA and ketone concentrations after calving but also caused a 9 lb/day decrease in dry matter intake during the final 4 days before calving in multiparous cows. These results indicate that a high dose of EN can decrease postpartum plasma NEFA and ketones but also may decrease prepartum dry matter intake.

## Introduction

Use of niacin in dairy cattle is widely studied; however, most results are inconclusive or contradictory. Niacin is a B vitamin that is required in very small amounts to maintain cellular metabolism. At much higher inclusion rates, niacin also has the ability to suppress the release of fat stores. Degradation within the rumen likely destroys almost 95% of niacin fed to ruminants, making additional supplementation inefficient. A rumen-protected form of niacin (encapsulated niacin; EN) became commercially available several years ago, providing a more effective option for dietary supplementation of niacin.

Fatty liver affects up to 50% of postpartum dairy cattle; this can be costly to producers because of milk production losses and secondary diseases, such as ketosis. Fatty liver occurs when cattle enter a negative energy balance, usually during the first 2 weeks of lactation. Lipolysis occurs as a response to the negative energy balance and results in the liver being overwhelmed by high blood concentrations of nonesterified fatty acids (NEFA). In experiments, niacin has been shown to have antilipolytic effects when given to cattle with induced fatty liver. Until this experiment, no known studies have been done to explore the metabolic and production responses to EN in peripartum dairy cows. The purpose of this study was to determine if 24 g/day of dietary EN could reduce lipolysis enough to control plasma NEFA in postpartum dairy cattle. Other metabolic and production responses measured were body condition score, milk production, liver composition, and dry matter intake.

## Experimental Procedures

Primiparous ( $n = 9$ ) and multiparous ( $n = 13$ ) Holstein cows were assigned randomly within parity to receive either 24 g/day EN or none (control) beginning 21 days before expected calving date and continuing until 21 days postpartum. Dry matter intake and milk production were measured daily until day 21 postpartum. Throughout the study, liver biopsies were collected for

triglyceride (TG) analysis and blood samples were collected for NEFA and  $\beta$ -hydroxybutyrate (BHBA) analyses. Cattle were housed in a tie-stall facility, milked thrice daily (0400, 1100, and 2100 hours), and fed twice daily (0700 and 1500 hours). Data were analyzed using mixed models with repeated measures over time.

## Results and Discussion

### *Dry Matter Intake*

There was a treatment by time by parity effect ( $P < 0.07$ , Figure 1) on prepartum dry matter intake caused by a 9 lb/day decrease in dry matter intake of EN-treated cows compared with control cows during the final 4 days before calving. There were no treatment effects on postpartum feed intake.

### *Plasma Traits*

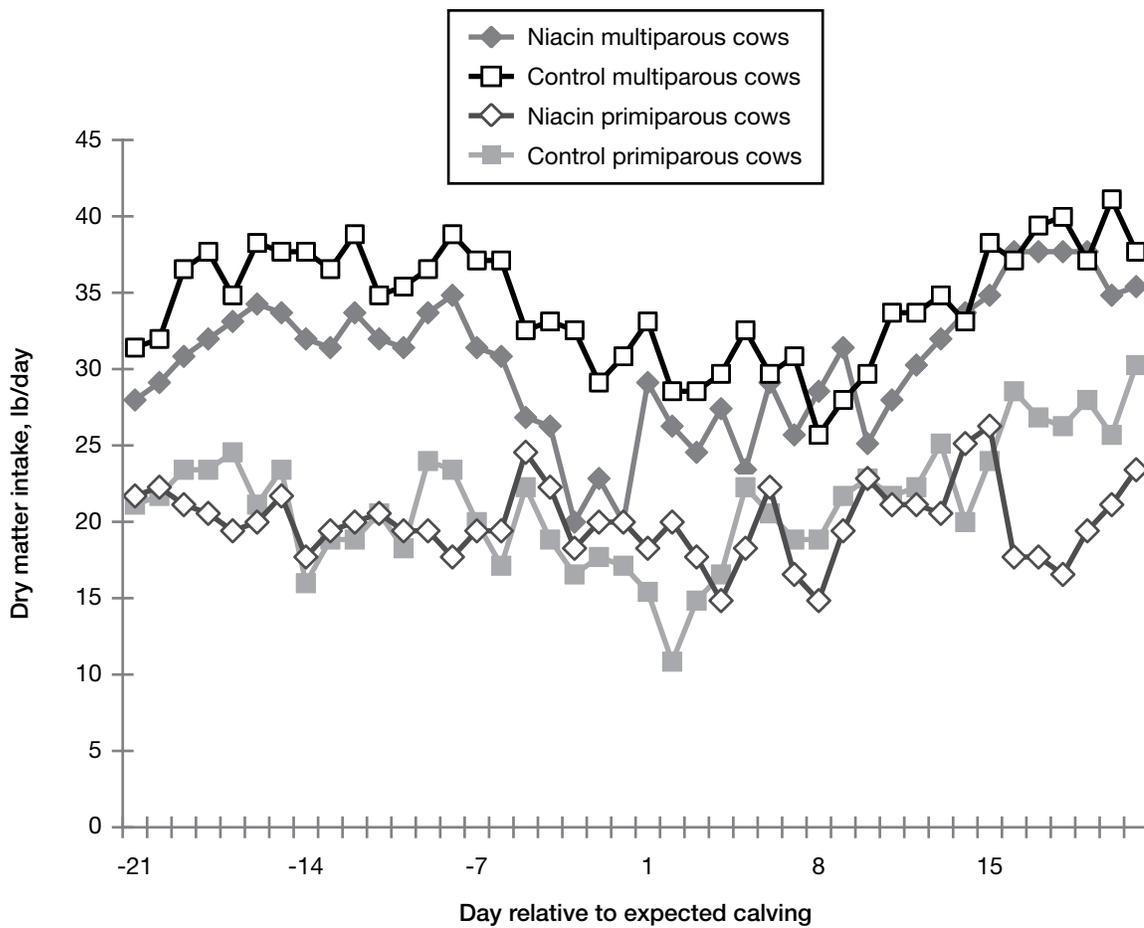
No prepartum treatment effects were detected for any of the plasma traits measured. In contrast, treatment by time by parity effects were detected for NEFA ( $P = 0.09$ , Figure 2) and BHBA concentrations ( $P = 0.03$ , Figure 3) during the postpartum period. Plasma NEFA peaked at 1700 and 1300  $\mu\text{M}$  for control primiparous and multiparous cows, respectively, compared with 700 and 800  $\mu\text{M}$  for treated primiparous and multiparous cows, respectively. Niacin treatment also suppressed peak BHBA concentrations in both parity groups.

### *Other Measures*

No treatment effects were observed for liver TG concentration, body condition score, body weight, or milk and milk component production.

In this study, 24 g/day of EN inhibited lipolysis in postpartum cows as demonstrated by a decrease in postpartum NEFA and BHBA concentrations. Depression of prepartum dry matter intake in multiparous cows is a novel finding and is difficult to explain. However, the results of this study also indicate that when EN reduced dry matter intake in multiparous cows, it still suppressed plasma NEFA and BHBA after calving. Although significant alterations in plasma lipid metabolism were detected after EN treatment, this did not result in decreased liver TG content. Lack of changes in liver TG content likely reflects the fact that no cows suffered from severe fatty liver disease in this study regardless of treatment. In summary, a high dose of EN can decrease postpartum plasma NEFA and BHBA. Further work is needed to understand the effects of niacin on prepartum dry matter intake and mechanisms involving postpartum metabolism.

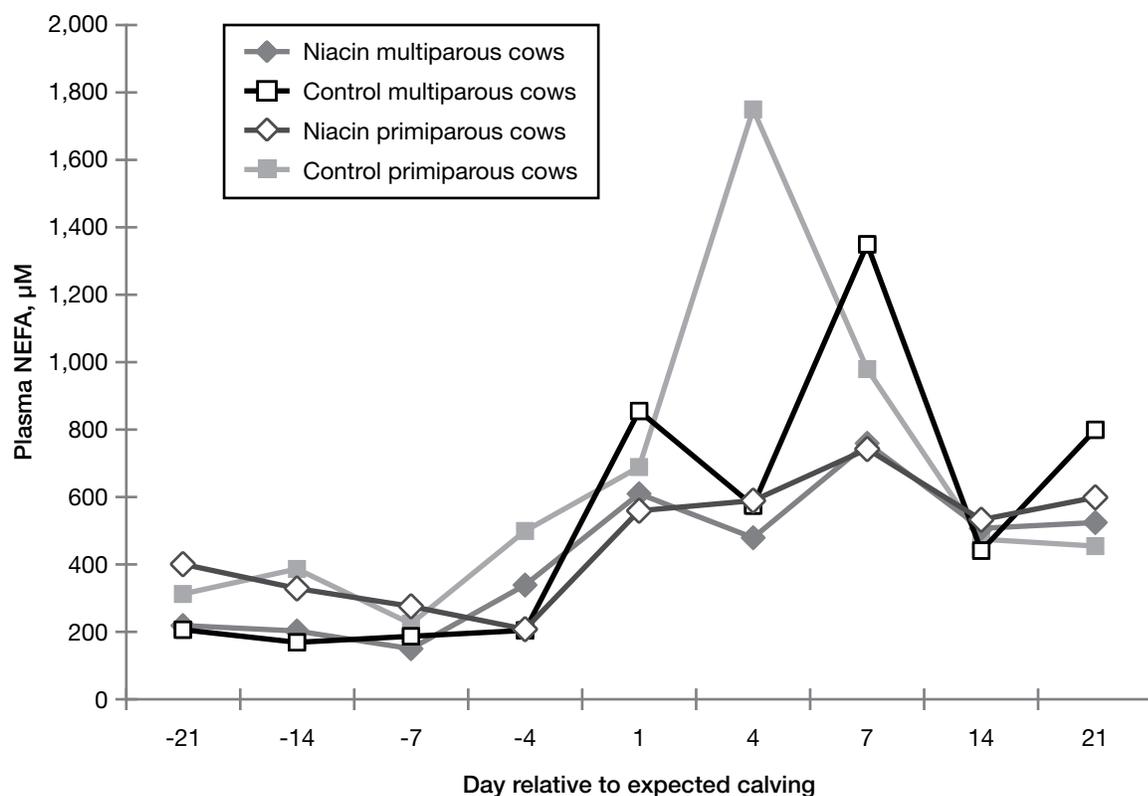
### NUTRITION AND FEEDING



**Figure 1. Dry matter intake of periparturient Holstein cows with or without 24 g/day of encapsulated niacin (EN) in the diet.**

Dry matter intake in EN-treated multiparous cows decreased ( $P < 0.01$ ) by 9 lb/day during the final 4 days before calving.

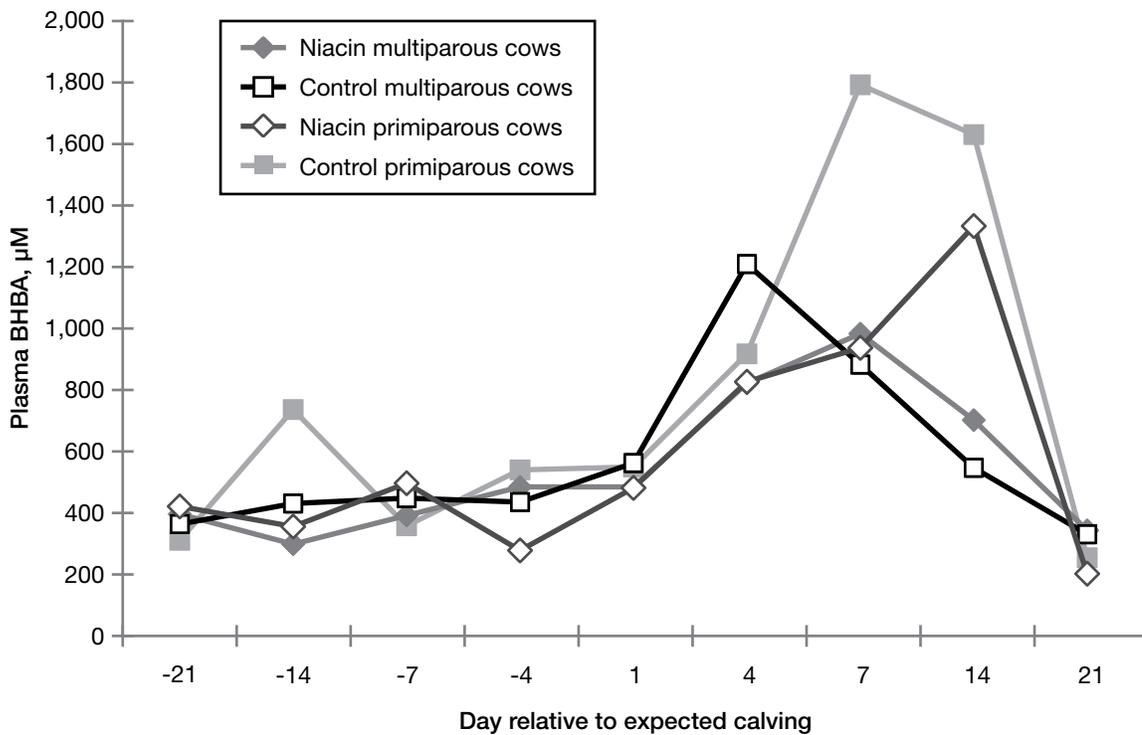
### NUTRITION AND FEEDING



**Figure 2. Plasma concentrations of nonesterified fatty acids (NEFA) in periparturient Holstein cows with or without 24 g/day of encapsulated niacin (EN) in the diet.**

A treatment by time by parity interaction was detected ( $P = 0.09$ ) after calving. Plasma NEFA peaked at 1700 and 1300  $\mu\text{M}$  for primiparous and multiparous control cows, respectively, compared with 700 and 800  $\mu\text{M}$  for EN-treated primiparous and multiparous cows, respectively.

### NUTRITION AND FEEDING



**Figure 3. Plasma concentrations of  $\beta$ -hydroxybutyrate (BHBA) in periparturient Holstein cows with or without 24 g/day of encapsulated niacin (EN) in the diet.**

A treatment by time by parity interaction was detected ( $P = 0.02$ ) after calving. Plasma NEFA peaked at 1800 and 1200  $\mu\text{M}$  for primiparous and multiparous control cows, respectively, compared with 1200 and 900  $\mu\text{M}$  for EN-treated primiparous and multiparous cows, respectively.

# Impact of Supplemental Phosphorus Source on Phosphorus Utilization in Lactating Dairy Cattle

*K. J. Lager, M. J. Brouk, B. J. Bradford and J. P. Harner*

## Summary

Supplemental phosphorus (P) in various forms and sources (pellet, meal, liquid, and corn dried distillers grains with solubles; **DDGS**) were compared in 12 multiparous Holstein cows producing 94.8 lb of milk ( $115 \pm 55$  days in milk) in a  $4 \times 4$  Latin square with 21-day periods. The pellet and meal diets contained monocalcium phosphate with a wheat middlings carrier, and the liquid diet contained ammonium polyphosphate in a cane molasses base. The DDGS supplied an organic P source. Cows were blocked by parity, days in milk, and milk production and randomly assigned to treatments. Phosphorus intakes were similar among all 4 diets (116, 116, 119 and 118 g/day for pellet, meal, liquid and DDGS diets, respectively). Cows consuming the liquid diet experienced greater ( $P < 0.001$ ) sugar intakes. Milk yield differed ( $P = 0.05$ ) among diets, with the DDGS diet yielding the most milk (76.3, 78.1, 75.2 and 80.5 lb/day for pellet, meal, liquid, and DDGS diets, respectively). There were no differences in milk fat and milk protein percentages or in daily lactose production. Excretion of P in feces tended ( $P = 0.07$ ) to differ among treatments (67.4, 66.3, 57.5, and 60.0 g/day for pellet, meal, liquid and DDGS diets, respectively), resulting in a trend ( $P = 0.10$ ) for greater P retention from diets, resulting in less P excretion. Secretion of P in milk did not differ among treatments. These data show that supplemental P source does not affect dry matter intake or P intake. Phosphorus source resulted in slight differences in P utilization, but it was not related to sorting of the diet. The DDGS diet showed responses similar to those of inorganic P mineral supplements and had favorable production yields, indicating that DDGS is an adequate substitute for mineral sources of P.

## Introduction

Developing feeding strategies that reduce phosphorus (P) excretion is crucial to alleviating the potential environmental hazards of P entering surface water and the resulting algal growth and eutrophication. The 2001 Nutrient Requirements for Dairy Cattle recommended P feeding level is 0.32 to 0.42%, depending on the physiological status of the animal, and research has shown that levels beyond this range are unnecessary. Overfeeding P often is practiced to ensure adequate P absorption to meet the cow's needs for milk production and reproduction; however, reproductive efficiency may decrease only when dietary levels fall below 0.2%. It is well known that feeding excess dietary P results in increased P in the manure and increased feed expense. The objective of this experiment was to analyze the effects of various sources of supplemental P presented in 3 forms on milk production and composition, P partitioning in the cow, and diet sorting in lactating dairy cows.

## Experimental Procedures

Twelve multiparous lactating dairy cows ( $115 \pm 55$  days in milk;  $98 \pm 14$  lb milk) were fed 1 of 4 diets with similar P levels but different forms and sources of P in a  $4 \times 4$  Latin square design with 3 replications. Treatment periods were 21 days in duration; the first 14 days served as an adaptation period, and the last 7 days were used to collect data. Cows were assigned randomly to treatments after treatments were balanced by days in milk, milk yield, and parity. Inorganic P forms and sources were a wheat middlings-based meal diet and a pellet diet containing monocalcium phosphate and a molasses-based liquid diet containing ammonium polyphosphate.

## NUTRITION AND FEEDING

The fourth diet used corn dried distillers grains with solubles (**DDGS**) as an organic source of supplemental P for comparison with inorganic sources and was presented in meal form.

Cows were milked twice daily at 0500 and 1600 hours. Milk weights were recorded at each milking, and duplicate milk samples were collected for 6 consecutive milkings beginning on day 1 of the collection week each period. One sample was collected in a vial with preservative; the second was collected in a vial without preservative and frozen for later analysis. Fecal grab samples were collected every 8 hours for 4 days beginning on day 1 of the collection week, and sampling time advanced 2 hours each day to account for diurnal variation.

Data were analyzed using the MIXED procedure of SAS version 9.1 (SAS Institute Inc., Cary, NC). The model statement included the effects of diet, replication, and the interaction between diet and replication as fixed effects. Random effects were cow and period. Treatment means were determined by using the LSMEANS option, and orthogonal contrasts were performed. Significance was determined at  $P \leq 0.05$ .

### Results and Discussion

Ingredients included in the diets are listed in Table 1. Diets were formulated to contain similar amounts of all nutrients, and adjustments were made for the inclusion of DDGS in the DDGS diet. Corn grain and Soy Best (Grain States Soya, West Point, NE) were reduced to account for the protein and energy supplied by the DDGS. Because the pellet, meal, and liquid diets differed only in P form, a base total mixed ration was mixed daily, and sufficient amounts were obtained to feed the cows on the selected treatment. Supplemental P was added to the base ration and mixed in a stand-alone drum tumble mixer (Data Ranger; American Calan, Northwood, NH). The DDGS diet was mixed separately, and the DDGS was added as a component of a grain mixture.

Dietary nutrient composition analysis (Table 2) showed dietary P concentrations of 0.46, 0.47, 0.49, and 0.47% for the pellet, meal, liquid, and DDGS diets, respectively. Fat and starch content of the DDGS diet were more than 17% greater and 11% less, respectively, than those of the other diets. There seemed to be small differences in mineral composition, but all other nutrients were similar across all diets.

Dry matter intake was similar across treatments, which explains the similarities in nutrient intake across treatments (Table 3). Fat intake was greater ( $P < 0.001$ ) for the DDGS diet than for the other treatments. The solubles portion of DDGS is added back to the distillers grains after starch is extracted from the corn endosperm for ethanol production. The solubles contain oil from the corn germ and increase the fat content of the distillers grains. Decreasing corn grain in the DDGS diet decreased ( $P = 0.004$ ) starch intake compared with the pellet and meal diets and tended ( $P = 0.08$ ) to reduce starch intake compared with the liquid diet. The additional molasses supplied by the P supplement in the liquid diet increased ( $P < 0.001$ ) sugar intake above that of all other treatments. Values for net energy for lactation intake were not different among treatments. Similarly, intakes of nonstructural carbohydrates were not different despite differences in starch and sugar intakes. Ash intake was not different across treatments, but instances of differing mineral intakes exist because of differences in mineral content of the diets. All minerals, however, were within sufficient ranges that would not affect experimental treatments.

Milk production was greater for the DDGS diet than for the pellet and meal diets (Table 3). Contrasts were performed to compare the inorganic P source supplied in the pellet and meal diets with the organic P source in the DDGS diet and the inorganic P source in the liquid diet as well as to compare the inorganic P source in the liquid diet with the organic P source. Phosphorus source affected milk production ( $P = 0.05$ ); the DDGS diet supported 3.3 lb/day more milk production than the inorganic P sources ( $P = 0.05$ ) and 5.3 lb/day more milk production than the liquid diet ( $P = 0.01$ ). The increase in milk production is not necessarily due to P intake. Milk fat and milk protein percentages were unaffected by treatment. In contrast, the liquid diet produced less fat than the inorganic ( $P = 0.05$ ) and organic ( $P = 0.01$ ) sources. Small decreases in milk fat percentage can occur with molasses supplementation. Sugar from molasses is readily soluble by ruminal microbes in the rumen, which increases propionate production. This leads to a decrease in the amount of acetate available for milk fat synthesis in the mammary gland. Daily milk protein production favored the DDGS diet, which had greater protein production than the inorganic ( $P = 0.03$ ) and liquid ( $P = 0.01$ ) diets, whereas no difference existed between inorganic and liquid P sources. Increases in daily protein and fat production followed an increase in milk yield. Fat-corrected milk was lower ( $P = 0.009$ ) for the liquid diet compared with the organic P supplement. Fat-corrected milk for the liquid diet was also lower ( $P = 0.05$ ) than for the inorganic P source.

As formulated, and along with similarities in dry matter intake, P intakes were similar across diets, but P utilization was different in some instances (Table 4). Dietary levels of P in this experiment would be sufficient for lactating cows producing more than 88 lb/day (based on the 2001 National Research Council-recommended value for dairy). Mean milk production over the length of the trial was less than 88 lb/day, resulting in cows being over supplemented with P near the end of the trial, when milk production was decreasing. Fecal P excretion tended ( $P = 0.07$ ) to be greater for the inorganic diets than for the organic diet and was greater ( $P = 0.02$ ) for the inorganic diet than for the liquid diet, but no difference existed between the liquid and organic dietary P supplements. Calculating P balance as P intake minus fecal and milk P resulted in a tendency ( $P = 0.10$ ) for the organic supplement to retain approximately 9 g/day more P than the inorganic supplement. The liquid diet also retained ( $P = 0.02$ ) more P than (13.5 g/day) than diets supplemented with inorganic P.

NUTRITION AND FEEDING

**Table 1. Ingredient composition of total mixed rations**

| Ingredient, % of diet dry matter | Treatments <sup>1</sup> |       |        |       |
|----------------------------------|-------------------------|-------|--------|-------|
|                                  | Pellet                  | Meal  | Liquid | DDGS  |
| Corn silage                      | 33.28                   | 33.28 | 33.56  | 32.38 |
| Alfalfa hay                      | 25.66                   | 25.66 | 25.97  | 24.98 |
| Whole cottonseed                 | 5.14                    | 5.14  | 5.14   | 5.00  |
| Corn grain, ground               | 18.40                   | 18.40 | 18.68  | 14.65 |
| Soy Best <sup>2</sup>            | 10.15                   | 10.15 | 10.15  | 2.88  |
| Sodium bicarbonate               | 0.76                    | 0.76  | 0.76   | 0.74  |
| Magnesium oxide                  | 0.13                    | 0.13  | 0.13   | 0.13  |
| MFP <sup>3</sup>                 | 0.09                    | 0.09  | 0.09   | 0.09  |
| Zinpro 4-Plex <sup>4</sup>       | 0.05                    | 0.05  | 0.05   | 0.05  |
| Sodium selenite, 0.06%           | 0.04                    | 0.04  | 0.04   | 0.04  |
| Vitamin A premix, 30,000/g       | 0.02                    | 0.02  | 0.02   | 0.02  |
| Vitamin E premix, 20,000/g       | 0.19                    | 0.19  | 0.19   | 0.18  |
| XP Yeast <sup>5</sup>            | 0.21                    | 0.21  | 0.21   | 0.21  |
| Rumensin <sup>6</sup>            | 0.01                    | 0.01  | 0.01   | 0.01  |
| Cane molasses                    | 0.90                    | 0.90  | 2.88   | 0.68  |
| DDGS                             | —                       | —     | —      | 16.43 |
| Wheat middlings                  | 3.55                    | 3.55  | —      | —     |
| Salt                             | 0.28                    | 0.28  | 0.32   | 0.28  |
| Calcium carbonate                | 0.76                    | 0.76  | 1.00   | 1.10  |
| Monocalcium phosphate            | 0.38                    | 0.38  | —      | 0.15  |
| Ammonium polyphosphate           | —                       | —     | 0.80   | —     |

<sup>1</sup> Supplemental phosphorus sources: wheat middling base containing monocalcium phosphate in pelleted (Pellet) and meal (Meal) forms; cane molasses base containing ammonium polyphosphate (Liquid); corn dried distillers grains with solubles (DDGS).

<sup>2</sup> Grain States Soya, West Point, NE.

<sup>3</sup> Dry source of 84% active methionine. Novus International, Inc., St. Charles, MO.

<sup>4</sup> Nutrient premix containing 2.58% zinc, 1.43% manganese, 0.90% copper, 0.18% cobalt, 8.21% methionine, 3.80% lysine, 11.5% protein, 1.5% fat, 22.0% fiber, and 26.5% ash; Zinpro Corp., Eden Prairie, MN.

<sup>5</sup> Diamond V Mills, Inc., Cedar Rapids, IA.

<sup>6</sup> Elanco, Greenfield, IN.

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**Table 2. Nutrient composition of total mixed rations containing various phosphorus supplements**

| Nutrient (dry matter basis)            | Treatments <sup>1</sup> |      |        |      |
|--|-------------------------|------|--------|------|
|  | Pellet                  | Meal | Liquid | DDGS |
| Crude protein, %                       | 16.6                    | 16.6 | 16.8   | 17.2 |
| Acid detergent fiber, %                | 18.6                    | 18.5 | 18.5   | 19.0 |
| Neutral detergent fiber, %             | 29.9                    | 29.9 | 29.2   | 31.3 |
| Fat, %                                 | 4.7                     | 4.6  | 4.6    | 5.6  |
| Sugar, %                               | 5.2                     | 5.2  | 6.4    | 4.7  |
| Nonstructural carbohydrates, %         | 43.4                    | 43.3 | 42.4   | 41.2 |
| NE <sub>L</sub> <sup>2</sup> , Mcal/kg | 1.70                    | 1.69 | 1.66   | 1.73 |
| Ash, %                                 | 7.7                     | 8.0  | 8.0    | 7.8  |
| Calcium, %                             | 1.0                     | 1.1  | 1.1    | 1.2  |
| Phosphorus, %                          | 0.46                    | 0.47 | 0.49   | 0.47 |
| Magnesium, %                           | 0.29                    | 0.29 | 0.29   | 0.28 |
| Potassium, %                           | 1.6                     | 1.6  | 1.7    | 1.5  |

<sup>1</sup> Supplemental phosphorus sources: wheat middling base containing monocalcium phosphate in pelleted (Pellet) and meal (Meal) forms; cane molasses base containing ammonium polyphosphate (Liquid); corn dried distillers grains with solubles (DDGS).

<sup>2</sup> Net energy of lactation.

**Table 3. Effect of supplemental phosphorus source and form in total mixed rations on lactating dairy cattle performance**

| Item                      | Treatment <sup>1</sup> |                    |                   |                   | SEM  | P    | Contrast             |                      |                      |
|---------------------------|------------------------|--------------------|-------------------|-------------------|------|------|----------------------|----------------------|----------------------|
|                           | Pellet                 | Meal               | Liquid            | DDGS              |      |      | I vs. O <sup>2</sup> | I vs. L <sup>3</sup> | L vs. O <sup>4</sup> |
| Dry matter intake, lb/day | 54.5                   | 54.0               | 53.4              | 54.0              | 1.8  | 0.97 | 0.90                 | 0.67                 | 0.78                 |
| Milk, lb/day              | 76.3 <sup>a</sup>      | 78.1 <sup>ab</sup> | 75.2 <sup>a</sup> | 80.5 <sup>b</sup> | 2.80 | 0.05 | 0.05                 | 0.25                 | 0.01                 |
| Milk fat, %               | 3.68                   | 3.83               | 3.58              | 3.68              | 0.16 | 0.26 | 0.47                 | 0.11                 | 0.43                 |
| Milk protein, %           | 3.05                   | 3.09               | 3.08              | 3.12              | 0.06 | 0.33 | 0.13                 | 0.67                 | 0.33                 |
| Milk fat, lb/day          | 2.80 <sup>ab</sup>     | 2.98 <sup>b</sup>  | 2.69 <sup>a</sup> | 2.95 <sup>b</sup> | 0.18 | 0.05 | 0.50                 | 0.05                 | 0.03                 |
| Milk protein, lb/day      | 2.31 <sup>a</sup>      | 2.40 <sup>ab</sup> | 2.32 <sup>a</sup> | 2.49 <sup>b</sup> | 0.09 | 0.04 | 0.03                 | 0.34                 | 0.01                 |
| SNF <sup>5</sup> , lb/day | 6.6 <sup>a</sup>       | 6.8 <sup>ab</sup>  | 6.6 <sup>a</sup>  | 7.1 <sup>b</sup>  | 0.29 | 0.05 | 0.06                 | 0.24                 | 0.01                 |
| Log somatic cell count    | 2.09                   | 1.90               | 2.03              | 2.21              | 0.18 | 0.29 | 0.14                 | 0.82                 | 0.27                 |
| Milk urea N, mg/dL        | 14.9                   | 14.7               | 15.4              | 14.3              | 0.73 | 0.12 | 0.19                 | 0.14                 | 0.02                 |
| FCM <sup>6</sup> , kg/day | 32.8 <sup>ab</sup>     | 34.4 <sup>b</sup>  | 32.0 <sup>a</sup> | 34.6 <sup>b</sup> | 1.55 | 0.03 | 0.21                 | 0.05                 | 0.009                |

<sup>abc</sup> Means within a row with differing superscripts differ ( $P < 0.05$ ).

<sup>1</sup> Supplemental phosphorus (P) sources: wheat middling base containing monocalcium phosphate in pelleted (Pellet) and meal (Meal) forms; cane molasses base containing ammonium polyphosphate (Liquid); corn dried distillers grains with solubles (DDGS).

<sup>2</sup> Inorganic P source vs. organic P source = pellet and meal vs. DDGS.

<sup>3</sup> Inorganic P source vs. liquid P source = pellet and meal vs. liquid.

<sup>4</sup> Liquid P source vs. organic P source = liquid vs. DDGS.

<sup>5</sup> Solids nonfat.

<sup>6</sup> Fat-corrected milk =  $(0.4 \times \text{lb of milk}) + (15 \times \text{lb of milk fat})$ .

**Table 4. Effect of supplemental phosphorus source and form on phosphorus partitioning in lactating dairy cattle**

| Item                        | Treatment <sup>1</sup> |      |        |      | SEM  | P    | Contrast             |                      |                      |
|-----------------------------|------------------------|------|--------|------|------|------|----------------------|----------------------|----------------------|
|                             | Pellet                 | Meal | Liquid | DDGS |      |      | I vs. O <sup>2</sup> | I vs. L <sup>3</sup> | L vs. O <sup>4</sup> |
| P intake, g/day             | 116                    | 116  | 119    | 118  | 5.05 | 0.93 | 0.75                 | 0.53                 | 0.79                 |
| Fecal P excretion, g/day    | 67.4                   | 66.3 | 57.5   | 60.0 | 3.54 | 0.07 | 0.07                 | 0.02                 | 0.54                 |
| Milk P concentration, g/day | 36.6                   | 39.0 | 36.4   | 37.2 | 1.84 | 0.51 | 0.72                 | 0.39                 | 0.66                 |
| P balance, g/day            | 11.7                   | 11.1 | 25.1   | 20.3 | 5.23 | 0.08 | 0.10                 | 0.02                 | 0.43                 |

<sup>1</sup> Supplemental phosphorus (P) sources: wheat middling base containing monocalcium phosphate in pelleted (Pellet) and meal (Meal) forms; cane molasses base containing ammonium polyphosphate (Liquid); corn dried distillers grains with solubles (DDGS).

<sup>2</sup> Inorganic P source vs. organic P source = pellet and meal vs. DDGS.

<sup>3</sup> Inorganic P source vs. liquid P source = pellet and meal vs. liquid.

<sup>4</sup> Liquid P source vs. organic P source = liquid vs. DDGS.

# Effect of Acidulant Addition on Yogurt Fermentation

*T. A. Boomgaarden and K. A. Schmidt*

## Summary

Yogurt was manufactured by pre-acidifying the yogurt mix with citric acid, lactic acid, or concentrated lemon juice either before or after pasteurization to a target pH of 6.2, and then the traditional manufacturing process was continued. Adding citric acid or lemon juice to the yogurt mix after pasteurization resulted in a 13% reduction in fermentation time compared with the control. This reduction in fermentation time may result in greater efficiency for yogurt manufacturers, allowing for a more sustainable manufacturing process.

## Introduction

In recent years, some consumers have expressed interest in purchasing products that have minimal impact on the environment, society, and the economy, which in short, is a definition of sustainable products. Consumers actively seek both food and nonfood sustainable products in the marketplace. In a recent survey, 50% of consumers responded that food products should contain labels that reflect their foods' impact on the environment and that they would buy food products that had less impact on the environment, provided that food costs did not increase. In the same study, however, only a small percentage of consumers viewed themselves as "most responsible" for their carbon footprint, instead placing most of the responsibility to reduce carbon footprints on manufacturers, government, and retailers. This insight into the consumer mindset leads to a basic conclusion: consumers expect products that fit the sustainable profile in the marketplace; thus, processors and producers should meet this need.

Yogurt is a growing food category; sales increased 6.5% from 2004 to 2005. Interestingly, sales of natural and organic yogurt accounted for much of this growth. In a comparison of the environmental life cycle of dairy products, yogurt had the second greatest energy consumption per ton, only behind cheese, with 25% of the energy usage attributed to the manufacturing process. Fermentation generally is the rate-limiting step in yogurt manufacture because typical fermentation times can range from 2 to 6 hours for the bacteria to reduce the yogurt mix pH from 6.4 to 4.6 (legal definition of yogurt). If fermentation time could be reduced, companies might reduce the amount of non-renewable resources, such as energy and water, required to produce a unit of yogurt. Thus, yogurt manufacture, particularly yogurt fermentation, is an excellent model for studying sustainable practices in a processing facility.

Adding the enzyme  $\beta$ -galactosidase to yogurt mix decreased fermentation time decreased while increasing yogurt digestibility and sweetness. Other researchers combined an external acidulant, glucano-delta lactone, with the starter cultures to decrease fermentation time by about 30%, but the resulting yogurt was firmer and exhibited more syneresis (i.e., separation of liquid from the gel). Adding ingredients such as casein hydrolysates, whey protein concentrates, and fructooligosaccharides to yogurt mix stimulated the growth of probiotic bacteria in yogurt and decreased fermentation time by 50%.

Other researchers developed a protocol of adding a concentrated mother culture (15% starter cultures) to the yogurt mix and reported that the resulting yogurt was similar to one made traditionally but the fermentation time was 50% less. General Mills, Inc. was issued a patent that

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described the process for directly acidifying yogurt when the pH reaches 4.8 to 5.2. Yogurt was successfully produced by adding a variety of acidulants such as citric acid, lactic acid, malic acid, gamma delta lactone, tartaric acid, and combinations thereof during the fermentation process. The yogurts had quality similar to that of yogurt made by the traditional process, but the fermentation time was reduced by 50%. This patent focused on minimizing the lag phase of the starter cultures by adding an acidulant (or acidulants) to quickly decrease pH to 4.6.

Reducing fermentation time has several advantages for yogurt producers including decreased labor cost, increased production efficiency, and more importantly, decreased non-renewable resource usage. The objective of this research was to determine a method of manufacturing yogurt in less time by pre-acidifying the mix with citric acid, lactic acid, or lemon juice, to jump-start acid production in the inoculated yogurt mix.

### Experimental Procedures

To make yogurt mix, 13.5% nonfat dry milk was reconstituted in water, rehydrated overnight, pasteurized at 194°F for 10 minutes and quickly cooled to 10°F within 10 minutes. Yogurt mixes were acidified with citric acid, lactic acid, or lemon juice (80 ppm) either before or after pasteurization to a target pH of  $6.2 \pm 1$ . A direct set culture of 2:1 *Streptococcus thermophilus* (ST) and *Lactobacillus delbrueckii* ssp. *bulgaricus* (LB) was added at 0.2%. Addition of ST and LB was considered to be the start of fermentation.

Yogurt mixes were fermented in a BioFlo 3000 (New Brunswick Scientific Co., Edison, NJ) fermentation unit until a pH of 4.6 and 0.9% titratable acidity were reached as per federal regulations. To monitor the fermentation process, pH and titratable acidity were measured every hour starting at inoculation following standardized methods. Microbiological analyses for ST and LB were assayed 10 minutes after inoculation and at the end of fermentation following standard methods. Seven treatments were prepared and 3 replications were done. As the experiment was designed to include a control, data were analyzed using procedure GLM and Dunnett's method in SAS 9.1 (SAS Institute Inc., Cary, NC) to determine differences from the control.

### Results and Discussion

Table 1 presents the results for fermentation time and LB and ST counts. Fermentation time was significantly affected by the acidulant treatment as well as by when the acidulant was added (before or after pasteurization). Neither citric acid nor lemon juice added before pasteurization affected fermentation time, but adding citric acid or lemon juice after pasteurization decreased fermentation time by 13.4% and 13.6%, respectively.

There were no differences in ST or LB counts expressed as colony forming units (cfu) per milliliter at the beginning or end of fermentation for any treatments, indicating that the experimental treatments did not affect the microbial populations compared with the control. This could be important to a yogurt manufacturer who uses the Live & Active Cultures seal, a program assuring that yogurt contains  $10^6$  bacteria per milliliter at the time of manufacture. The experimental treatments did not affect the viability of starter cultures and would meet the Live & Active Cultures requirements.

Adding citric acid or lemon juice to yogurt mixes after pasteurization resulted in shorter fermentation times and a more efficient yogurt manufacturing process. Changes in titratable acid

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and pH during fermentation were very similar to the control yogurt, with the exception of citric acid and lemon juice added after pasteurization (Figure 1). Yogurt mixes that were pre-acidified with citric acid or lemon juice after pasteurization developed acidity at an accelerated rate compared with control yogurt.

Lactic acid addition either before or after pasteurization resulted in a longer fermentation time than the control. Lactic acid is the result of lactose fermentation by ST and LB, and its external addition slowed fermentation in this study. This could be due to the low acid tolerance of ST, which may have prevented ST growth during the early phases of fermentation.

Overall, the order of acidulant addition affected fermentation time. Researchers have reported that if yogurt mix is pasteurized at pH <6.55, the whey proteins tend to self-aggregate rather than aggregate with the casein proteins during heat treatment. Perhaps the self-aggregation process of the whey proteins inhibited some of the proteolytic reactions necessary to induce the desired symbiotic relationship between ST and LB that allows for accelerated fermentation.

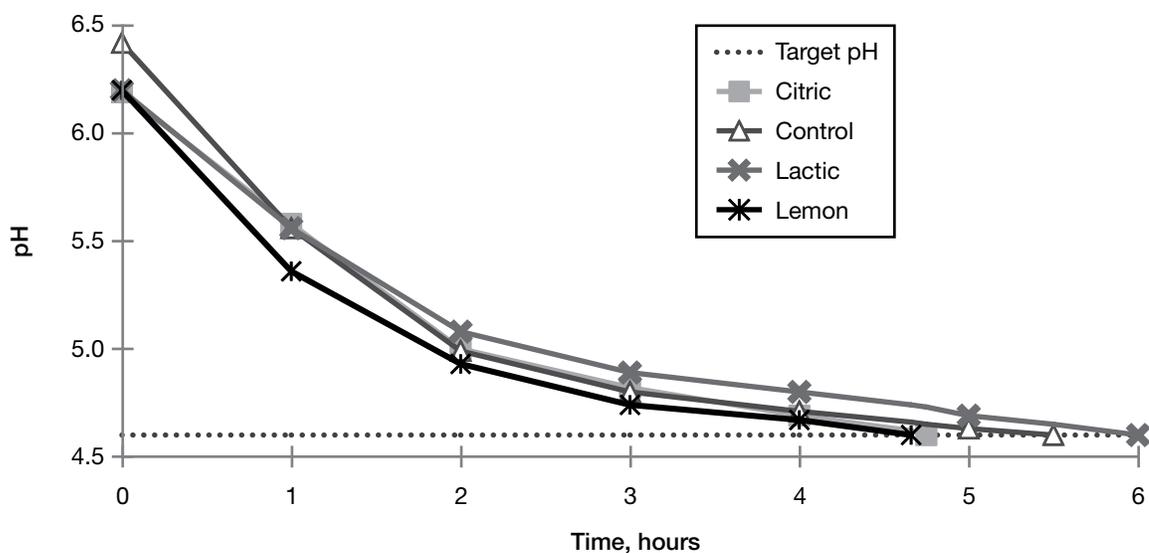
In summary, fermentation time could be reduced by adding certain acidulants to yogurt mix after pasteurization. It is important to note that although the fermentation time decreased about 45 minutes and the pre-acidification process required an additional 2 minutes, an overall reduction in the time required to manufacture yogurt was demonstrated. Decreasing the fermentation time of yogurt while maintaining desirable sensory and physiochemical properties, should appeal to both yogurt manufacturers and consumers seeking sustainable products. A decreased process time could potentially save energy, thus reducing the carbon footprint of the yogurt manufacturer. Further testing is warranted to ensure that the functional and sensory properties of these yogurts are not adversely affected.

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**Table 1. Average fermentation times and counts of *Lactobacillus bulgaricus* and *Streptococcus thermophilus***

| Acidulant treatment | Addition order | Fermentation, hours | <i>L. bulgaricus</i> log, cfu/mL |                     | <i>S. thermophilus</i> log, cfu/mL |                     |
|---------------------|----------------|---------------------|----------------------------------|---------------------|------------------------------------|---------------------|
|                     |                |                     | Start of fermentation            | End of fermentation | Start of fermentation              | End of fermentation |
| Control             | —              | 5.44 <sup>b</sup>   | 6.83                             | 7.62                | 7.78                               | 7.77                |
| Citric              | Before         | 5.33 <sup>b</sup>   | 7.23                             | 7.14                | 7.55                               | 7.48                |
| Lemon               | Before         | 5.50 <sup>b</sup>   | 6.93                             | 7.00                | 7.77                               | 7.96                |
| Lactic              | Before         | 6.00 <sup>a</sup>   | 7.15                             | 7.22                | 7.65                               | 7.72                |
| Citric              | After          | 4.75 <sup>c</sup>   | 6.81                             | 7.54                | 7.68                               | 7.54                |
| Lemon               | After          | 4.66 <sup>c</sup>   | 6.92                             | 6.98                | 7.78                               | 7.65                |
| Lactic              | After          | 6.00 <sup>a</sup>   | 6.46                             | 6.52                | 7.90                               | 7.68                |

<sup>abc</sup> Means having different superscript letters differ ( $P < 0.05$ ).



**Figure 1. pH development during fermentation of yogurt samples with acidulant added after heat treatment.**

# Luteolysis and Pregnancy Outcomes in Dairy Cows after Treatment with Estrumate or Lutalyse

*J. S. Stevenson and A. P. Phatak<sup>1</sup>*

## Summary

In Experiment 1, lactating dairy cows ( $n = 1,230$ ) in 6 herds were treated with 2 injections of prostaglandin  $F_{2\alpha}$  (**PGF<sub>2 $\alpha$</sub>** ) 14 days apart (**Presynch**), with the second injection administered 12 to 14 days before the onset of a timed AI protocol (**Ovsynch**). Cows were inseminated when detected in estrus after the Presynch PGF<sub>2 $\alpha$</sub>  injections. Cows not inseminated were enrolled in the Ovsynch protocol and were assigned randomly to be treated with either Estrumate or Lutalyse as part of a timed artificial insemination (**AI**) protocol. Blood samples were collected before treatment injection (0 hour) and 48 and 72 hours later. In cows having progesterone concentrations  $\geq 1$  ng/mL at 0 hour and potentially having a functional corpus luteum (**CL**) responsive to a luteolytic agent, Lutalyse increased ( $P < 0.05$ ) luteal regression from 83.9 to 89.3%. Despite a significant increase in luteolysis, pregnancy rate per AI did not differ between treatments. Fertility was improved in both treatments in cows having reduced progesterone concentrations at 72 hours and in those showing signs of estrus. In Experiment 2, an ovulation resynchronization (**Ovsynch-Resynch**) program was initiated with gonadotropin-releasing hormone (**GnRH**) or saline in 427 previously inseminated lactating dairy cows of unknown pregnancy status in 1 herd. Seven days later, pregnancy was diagnosed and nonpregnant cows were blocked by number of CL and assigned randomly to receive Estrumate or Lutalyse. Diameter of each CL was recorded and blood samples were collected at 0 and 72 hours after treatment to assess serum progesterone. A fixed-time AI was given at 72 hours after treatment and approximately 16 hours after a GnRH injection to induce ovulation. Lutalyse increased ( $P < 0.05$ ) luteal regression from 69.1 to 78.5% regardless of the number of CL present or the total luteal volume per cow exposed to treatment. Pregnancy rate per AI did not differ between treatments. Although Lutalyse was slightly more effective than Estrumate in inducing luteolysis in lactating dairy cows exposed to an Ovsynch or Ovsynch-Resynch protocol, resulting pregnancy outcomes did not differ between products.

## Introduction

Since the first prostaglandin  $F_{2\alpha}$  (**PGF<sub>2 $\alpha$</sub>** ) product (Lutalyse, The Upjohn Co., Kalamazoo, MI) was introduced in the United States in 1979, several agonists and generic PGF<sub>2 $\alpha$</sub>  products have become available by prescription. The major difference in available products is between those that are chemically similar to uterine-derived PGF<sub>2 $\alpha$</sub>  (Lutalyse, ProstaMate, and In Synch) and its agonists (Estrumate and estroPLAN).

Different physiological responses of bovine females to administration of either Estrumate or Lutalyse have been reported for luteolysis, receptor binding, intrauterine pressure, estrus expression, conception rates, and pregnancy rates. An unpublished meta-analysis of some of these factors did not find significant differences in conception rate and pregnancy rate or overall differences in detected estrus. Odds ratios, however, consistently were greater than 1.0, indicating trends in the combined studies that numerically, but not significantly, favored Estrumate over Lutalyse.

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Strict timed artificial insemination (**AI**) programs have become common in dairy operations because they are reliable and not dependent on visual or other means of detecting estrus in cattle. The **Ovsynch** protocol (injection of gonadotropin-releasing hormone (**GnRH**) 7 days before and 48 or 72 hours after treatment with prostaglandin  $\text{PGF}_{2\alpha}$ ; timed AI at 72 hours) synchronizes follicular maturation and luteal regression, resulting in about 20 to 30% of cows having at least 2 luteal structures at the time of  $\text{PGF}_{2\alpha}$  injection. A good test of luteolytic efficacy between product types (Lutalyse vs. Estrumate) is possible in lactating cows to which the Ovsynch protocol is applied because a larger proportion of cows have more than 1 corpus luteum to regress at the time of the  $\text{PGF}_{2\alpha}$  injection.

The present study consisted of 2 experiments. The objective of the first experiment was to determine the efficacy of luteal regression in response to 2 chemically different luteolytic products (Estrumate vs. Lutalyse) as determined by changes in progesterone concentrations in blood and subsequent pregnancy outcome of lactating dairy cows exposed to either of the 2 products before first postpartum AI. The objective of the second experiment was similar to that of the first, except the number of corpora lutea (**CL**) and total luteal tissue volume were quantified in previously inseminated nonpregnant dairy cows before treatment injections were administered.

## Experimental Procedures

### *Experiment 1*

Lactating dairy cows were enrolled at multiple sites in Merced and Stanislaus counties in the Central Valley of California. Cows were enrolled in a **Presynch** protocol (2  $\text{PGF}_{2\alpha}$  injections administered 14 days apart; Lutalyse; Pfizer Animal Health, New York, NY). Cows detected in estrus in response to the Presynch  $\text{PGF}_{2\alpha}$  injections were inseminated, and the residual cows were then enrolled in a Cosynch-72 timed AI program (GnRH injection administered 7 days before and 72 hours after treatment with  $\text{PGF}_{2\alpha}$ ; timed AI at 72 h) that was initiated 12 to 14 days after the second Presynch injection (Figure 1). Cows were assigned randomly to either of 2 luteolytic product injections as the treatment  $\text{PGF}_{2\alpha}$  that preceded AI. Cows received i.m. 2 mL of Estrumate (0.5 mg of Cloprostenol, Schering Plough Animal Health, Union, NJ) or 5 mL of Lutalyse (Dinoprost, Pfizer Animal Health) before AI as part of the Cosynch-72 procedure. Body condition scores (**BCS**; 1 = emaciated, 5 = obese) were assigned at treatment in 1,019 of 1,230 (82.8%) cows studied.

Cows detected in estrus after the treatment injection and before the scheduled AI were inseminated while restrained in feed line lockups. Estrus detection included visual observation but also relied on tail chalk removal when cows were examined each morning while restrained in feed line lockups. Inseminations made during the breeding week that followed treatment injections included those made after detected estrus and at 72 hours posttreatment by appointment. A small proportion of cows in 1 herd detected in estrus after AI were reinsmated when still in estrus 12 hours later. Breeding codes at the time of AI were as follows: (1) timed AI-coded cows had no diagnosed signs of estrus before or at the time of AI, (2) estrus-coded cows were inseminated before the scheduled timed AI (83%) or double inseminated (timed AI and then reinsmated because of estrus expression; 17%), and (3) timed AI + estrus-coded cows were diagnosed in estrus at the timed AI.

Pregnancy was diagnosed weekly in 5 herds by transrectal palpation of the uterus beginning at 35 days after AI; in the sixth herd, pregnancy was diagnosed by transrectal ultrasonography at

day 32 after AI. At all locations, blood samples were collected before treatment injection (0 hour) and at 48 and 72 hours. Progesterone was quantified in serum by radioimmunoassay.

### ***Experiment 2***

In our Kansas State University herd, 333 lactating dairy cows of unknown pregnancy status were enrolled in an ovulation resynchronization (**Ovsynch-Resynch**) procedure (GnRH injection administered 7 days before a not-pregnant diagnosis before treatment with PGF<sub>2α</sub> followed in 56 hours by a GnRH injection and timed AI at 72 h; Figure 1). Cows were eligible to be treated randomly with either Estrumate or Lutalyse as in Experiment 1 when 1 or more CL was present upon transrectal ultrasonography before treatment. An additional 94 cows not preenrolled in an Ovsynch-Resynch protocol and having a CL at a not-pregnant diagnosis were administered randomly either Estrumate or Lutalyse followed in 56 hours by a GnRH injection and timed AI at 72 hours.

Ovarian follicles and CL were mapped and sized by transrectal ultrasonography at the time of the not-pregnant diagnosis for purposes of counting of follicles  $\geq 10$  mm in diameter and the number of CL before treatment. All CL were assumed to be spherical. Diameter of structures was determined by averaging their largest cross-sectional width and height, measured by ultrasound electronic calipers. When a CL contained a fluid-filled cavity, volume of the cavity was subtracted from the calculated CL volume.

Body condition scores were assigned at treatment as in Experiment 1. Blood samples were collected at 0 and 72 hours after treatment injection and later assayed for progesterone content. Pregnancy diagnosis subsequent to AI occurred 32 to 39 days after timed AI. A positive pregnancy outcome required presence of uterine fluid and a large CL or uterine fluid and presence of an embryo.

## **Results and Discussion**

### ***Experiment 1***

When considering only cows having pretreatment progesterone concentrations  $\geq 1$  ng/mL and potentially eligible to respond to either Estrumate or Lutalyse, the proportion of cows having successful luteolysis (progesterone  $< 1$  ng/mL at 72 hours) was greater ( $P < 0.05$ ) in cows treated with Lutalyse (Table 1). Further, in 5 of 6 herds, the proportion of cows having luteal regression was numerically greater after Lutalyse. The odds ratio indicated that the odds for successful luteolysis were 95% greater (odds ratio = 1.95; 95% confidence interval = 1.27 to 2.45;  $P < 0.05$ ) for cows treated with Lutalyse than for cows treated with Estrumate. In the reduced set of cows with recorded BCS, thinner cows (BCS  $\leq 2.5$ ) were 2.15 times (95% confidence interval = 1.31 to 3.54;  $P < 0.05$ ) more likely to have luteolysis than cows having BCS  $> 2.25$  (93.7 vs. 86.8%, respectively).

Pregnancy rate per AI (**PR/AI**) did not differ between treatments in cows with or without luteolysis or in cows with different BCS. Differences in PR/AI were detected among locations (Table 2). At 4 of 6 locations, PR/AI was numerically greater for cows treated with Lutalyse than for cows treated with Estrumate. Although PR/AI did not differ between treatments, breeding codes indicated that cows in estrus at the timed AI had greater ( $P < 0.05$ ) PR/AI than those receiving timed AI without estrual symptoms. Cows inseminated after detected estrus before the scheduled timed AI or those double inseminated because estrus was detected after timed AI had intermediate PR/AI (Table 2). Most of the cows that were estrus coded before AI

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expressed estrus (83%) during 1 to 3 days before the scheduled timed AI compared with 17% of cows reinseminated within 24 hours after the timed AI because of detected signs of estrus. Further, similar proportions of cows displayed estrus after Estrumate and Lutalyse (49.6 vs. 51.7%, respectively).

### **Experiment 2**

The proportion of 427 cows having 1, 2, or 3 CL before treatment was 75.2% ( $n = 321$ ), 22.7% ( $n = 97$ ), and 2.1% ( $n = 9$ ), respectively. Among factors analyzed (treatment, number of CL, lactation number, energy-corrected milk yield, injection of GnRH 7 days before treatment, number of ovarian follicles  $\geq 10$  mm in diameter, BCS, days in milk, season, and pretreatment progesterone concentration in cows having 1 or more than 1 CL), only treatment, BCS, and season were significant (Table 3). Luteal regression was 1.64 times more ( $P < 0.001$ ) likely with Lutalyse than with Estrumate. Regression of CL in cows having 1 CL was 74.7% and did not differ from that for cows having more than 1 CL (71.2%). Corpora lutea in cows having greater BCS ( $>2.25$  vs.  $\leq 2.25$ ) were 2.72 times less likely to regress (83.2 vs. 62.5%, respectively). The poorest CL regression occurred during summer (57.5%); the best occurred during winter (80.8%).

Among factors tested that may influence pregnancy outcome (treatment, number of CL, lactation number, energy-corrected milk yield, injection of GnRH 7 days before treatment, number of ovarian follicles  $\geq 10$  mm in diameter, BCS, days in milk, season, and pretreatment progesterone concentration in cows having 1 or more than 1 CL), only GnRH injection 7 days before treatment ( $P = 0.059$ ) and BCS ( $P = 0.09$ ) tended to be significant (Table 4). Pregnancy rate per AI varied little between Estrumate and Lutalyse treatments (31.3 vs. 32.8%, respectively). Likewise, no advantage for either product was detected for PR/AI whether cows had 1 or more than 1 CL (31.5 vs. 33.7%, respectively). Injecting GnRH 7 days before treatment tended ( $P = 0.059$ ) to increase the odds of PR/AI (34.6 vs. 23%) by 1.71 times (for none vs. GnRH, respectively), and greater ( $>2.25$  vs.  $\leq 2.25$ ) BCS tended ( $P = 0.09$ ) to decrease PR/AI (27.1 vs. 35.8%, respectively).

Pretreatment progesterone concentrations did not differ before treatments of Lutalyse or Estrumate were applied ( $4.75 \pm 0.2$  vs.  $4.57 \pm 0.2$  ng/mL, respectively). By 72 hours posttreatment, concentrations were similar between treatments ( $0.89 \pm 0.2$  vs.  $1.03 \pm 0.2$  ng/mL, respectively). Pretreatment progesterone concentrations in serum were greater ( $P < 0.001$ ) for cows having more than 1 CL ( $5.92 \pm 0.31$  ng/mL;  $n = 106$ ) than for cows having only 1 CL ( $4.22 \pm 0.19$  ng/mL;  $n = 321$ ).

On the basis of our definition for luteolysis, which required progesterone concentrations to be  $\geq 1$  ng/mL before treatment and  $< 1$  ng/mL by 72 hours after treatment, Lutalyse was slightly more effective as a luteolytic product than Estrumate. This was true in both experiments, including cows known to have 1 or more than 1 CL before treatment. Although a slight difference in luteolytic efficacy was observed, no differences in pregnancy outcome were detected in either experiment. In both experiments, luteolysis was less effective in cows with BCS exceeding 2.25 or 2.50 compared with thinner cows. We concluded that both products were equally effective luteolysins for producing similar pregnancy outcomes in lactating dairy cows.

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**Table 1. Proportion of cows having luteal regression in response to Estrumate or Lutalyse treatment (Experiment 1)<sup>1</sup>**

| Location | Treatment         |            | Overall      |
|----------|-------------------|------------|--------------|
|          | Estrumate         | Lutalyse   |              |
|          | ----- % (n) ----- |            |              |
| 1        | 93.5 (46)         | 91.7 (48)  | 92.6 (94)    |
| 2        | 82.8 (64)         | 96.7 (61)  | 89.6 (125)   |
| 3        | 88.1 (42)         | 92.1 (38)  | 90.0 (80)    |
| 4        | 78.0 (50)         | 88.0 (50)  | 83.0 (100)   |
| 5        | 74.5 (98)         | 77.6 (98)  | 76.0 (196)   |
| 6        | 92.1 (252)        | 98.2 (217) | 94.9 (469)   |
| Total    | 86.4** (552)      | 92.0 (512) | 89.1 (1,064) |

\*\* Different from Lutalyse ( $P < 0.01$ ). Odds ratio = 1.95 (95% confidence interval = 1.29 to 2.94).

<sup>1</sup> Only cows having luteolysis (pretreatment progesterone concentrations  $\geq 1$  ng/mL and 72-hour posttreatment concentrations  $< 1$  ng/mL) were analyzed.

**Table 2. Pregnancies per artificial insemination (AI) in response to Estrumate or Lutalyse injection as part of the Cosynch-72 protocol (Experiment 1)**

| Item                           | Treatment <sup>1</sup> |            | Overall                  |
|--------------------------------|------------------------|------------|--------------------------|
|                                | Estrumate              | Lutalyse   |                          |
| Luteal regression <sup>2</sup> |                        |            |                          |
| No                             | 8.0 (75)               | 7.3 (41)   | 7.8 <sup>a</sup> (116)   |
| Yes                            | 44.3 (469)             | 43.1 (459) | 43.8 <sup>b</sup> (928)  |
| Body condition <sup>3,4</sup>  |                        |            |                          |
| $\leq 2.5$                     | 38.0 (234)             | 34.1 (226) | 36.1 <sup>a</sup> (460)  |
| $> 2.5$                        | 37.2 (293)             | 42.5 (266) | 39.7 <sup>a</sup> (559)  |
| Location <sup>3</sup>          |                        |            |                          |
| 1                              | 43.9 (57)              | 40.0 (55)  | 42.0 <sup>ab</sup> (112) |
| 2                              | 25.0 (80)              | 30.1 (83)  | 27.6 <sup>a</sup> (163)  |
| 3                              | 34.0 (53)              | 17.0 (47)  | 26.0 <sup>a</sup> (100)  |
| 4                              | 30.9 (55)              | 32.7 (55)  | 31.8 <sup>ab</sup> (110) |
| 5                              | 37.7 (130)             | 39.4 (127) | 38.5 <sup>b</sup> (257)  |
| 6                              | 40.0 (260)             | 44.7 (228) | 42.2 <sup>b</sup> (488)  |
| Breeding code <sup>3,5</sup>   |                        |            |                          |
| Timed AI                       | 33.6 (453)             | 33.5 (400) | 33.5 <sup>a</sup> (853)  |
| Estrus                         | 38.3 (107)             | 43.1 (102) | 40.7 <sup>ab</sup> (209) |
| Timed AI + estrus              | 53.3 (80)              | 50.5 (93)  | 51.8 <sup>b</sup> (168)  |
| Total                          | 36.7 (635)             | 37.8 (595) |                          |

<sup>ab</sup> Mean percentages within column having different superscript letters differ ( $P \leq 0.05$ ).

<sup>1</sup> Treatment was applied 3 days before scheduled timed AI.

<sup>2</sup> Includes only cows eligible for luteal regression (progesterone concentrations  $\geq 1$  ng/mL before treatment).

<sup>3</sup> Includes all cows regardless of whether luteal regression occurred.

<sup>4</sup> Body condition score was assessed in 1,019 of 1,230 (82.8%) cows.

<sup>5</sup> Timed AI-coded cows had no diagnosed signs of estrus before AI, estrus-coded cows were inseminated after treatment injection but before the scheduled timed AI (83%) or double inseminated (17%; showed estrus after timed AI and were reinseminated), and timed AI + estrus-coded cows were in estrus at the timed AI.

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**Table 3. Factors affecting luteal regression after treatment with Estrumate and Lutalyse in lactating dairy cows having 1 or more than 1 corpus luteum before treatment (Experiment 2)**

| Treatment | n <sup>1</sup> | Luteal regression (%) | Odds ratio | Confidence limits | P value |
|-----------|----------------|-----------------------|------------|-------------------|---------|
| Estrumate | 191            | 69.1                  | Referent   |                   | 0.001   |
| Lutalyse  | 205            | 78.5                  | 1.64       | 1.01-2.68         |         |

<sup>1</sup> Although a corpus luteum was visible, 31 of 427 cows that did not have pretreatment progesterone concentrations  $\geq 1$  ng/mL (not eligible for luteolysis) were excluded.

**Table 4. Factors affecting pregnancy rate per artificial insemination (PR/AI) after treatment with Estrumate and Lutalyse in dairy cows having 1 or more than 1 corpus luteum before treatment (Experiment 2)**

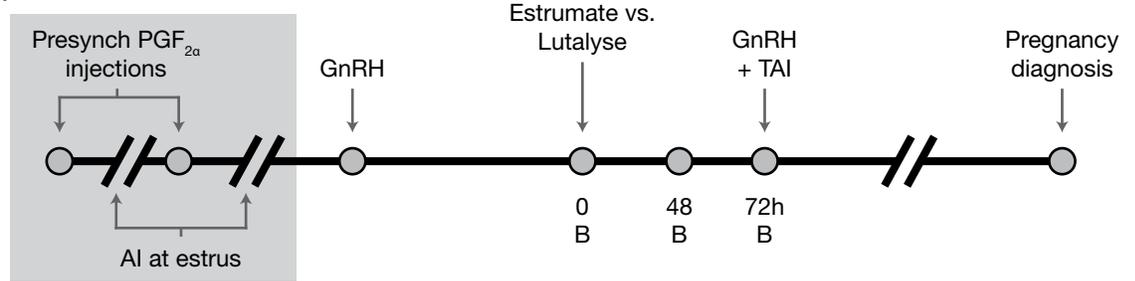
| Treatment | n <sup>1</sup> | PR/AI <sup>2</sup> (%) | Odds ratio | Confidence limits | P value |
|-----------|----------------|------------------------|------------|-------------------|---------|
| Estrumate | 198            | 31.3                   | Referent   |                   | 0.707   |
| Lutalyse  | 201            | 32.8                   | 1.09       | 0.71-1.66         |         |

<sup>1</sup> Excludes 28 of 427 cows for which pregnancy outcome was not known before culling.

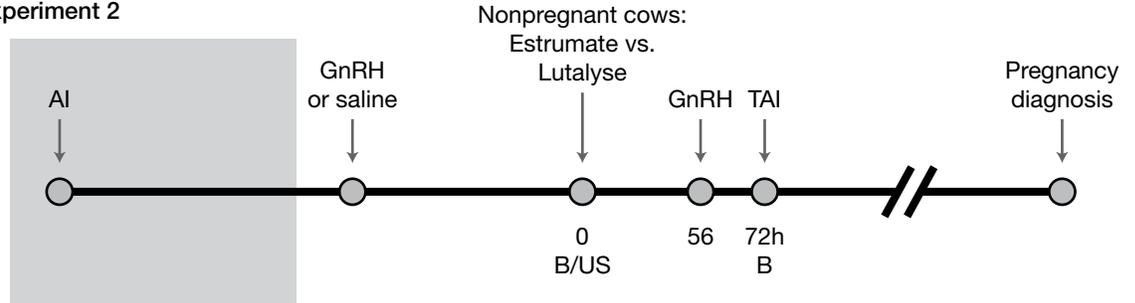
<sup>2</sup> Includes all cows regardless of luteal regression status.

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Experiment 1



Experiment 2



**Figure 1. Experimental design of treatments.**

**Experiment 1.** Lactating dairy cows in 6 California dairy herds were enrolled in a Presynch protocol (2 PGF<sub>2α</sub> injections administered 14 days apart). Cows detected in estrus in response to the Presynch PGF<sub>2α</sub> injections were inseminated, and the remaining cows were treated with the Cosynch-72 timed AI protocol beginning 12 or 14 days after the second Presynch injection. Alternate cows were administered 2 mL of Estrumate or 5 mL of Lutalyse before timed AI as part of Cosynch-72.

**Experiment 2.** At 1 Kansas location, cows of unknown pregnancy status were enrolled in an Ovsynch-Resynch procedure (GnRH injection or none administered 7 days before a not-pregnant diagnosis before treatment with Estrumate or Lutalyse followed in 56 hours by a GnRH injection and timed AI at 72 hours). Ovarian structures were mapped and sized by transrectal ultrasonography (US) at the time of the not-pregnant diagnosis.

At all locations, blood samples (B) were collected before treatment injection (0 hour) and at 48 and 72 hours (Experiment 1) or at 0 and 72 hours (Experiment 2). Shaded area for Experiment 1 represents pretreatment Presynch injections. Cows not inseminated were then assigned randomly to the experiment. Shaded area for Experiment 2 represents pretreatment AI in which only resulting nonpregnant cows were enrolled in the experiment.

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