# The Impact of a Monensin Controlled-Release Capsule on Subclinical Ketosis in the Transition Dairy Cow

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## ABSTRACT

An experiment was designed to examine subclinical ketosis in periparturient dairy cows and the antiketogenic effects of monensin. Subclinical ketosis was induced through a 10% feed restriction and was quantitatively determined using a blood $\beta$-hydroxybutyrate (BHBA) threshold of 1200 $\mu$mol/L. Monensin decreased the BHBA concentration by 35% and increased the glucose concentration by 15%. No effect of monensin on milk production was detected, but rumen fermentation was altered. Monensin decreased the acetate to propionate ratio, decreased the butyrate concentration, and increased pH. The lower concentration of BHBA in blood and higher concentration of blood glucose in cows treated with a monensin controlled-release capsule decreased subclinical ketosis in early lactation cows.
(**Key words**: lactating dairy cows, monensin controlled-release capsule, subclinical ketosis, $\beta$-hydroxybutyrate)

**Abbreviation key**: **CRC** = controlled-release capsule.

## INTRODUCTION

A negative energy balance in early lactation predisposes the dairy cow to metabolic imbalances and diseases such as fatty liver and ketosis. Ketosis is a disease related to the high metabolic priority of the mammary gland for glucose and the inability of the cow to meet the glucose demand through appetite (4). As a consequence, adipose tissue is mobilized, and free fatty acids are oxidized in the liver, which results in ketone body production. Overproduction and accumulation of ketone bodies is toxic (8); symptoms include a decline in BW and milk production, loss of appetite, hypoglycemia, hyperketonemia, elevated NEFA, fat deposition in the liver, and loss of liver glycogen (4). Although symptoms of subclinical ketosis are not detectable, it is potentially serious because it usually remains undetected, untreated, and could progress to a clinical condition (4). Subclinical ketosis has also been associated with a loss of milk production of 1.0 to 1.4 kg/d (11). This loss in production is economically significant to the dairy producer, particularly because the incidence level in Ontario herds has been estimated at 12% (11).

Monensin, a carboxylic ionophore, has been used extensively in the beef cattle industry to combat bloat and coccidiosis as well as improve feed efficiency and growth rate (17). The primary action of monensin is to alter the rumen fermentation pattern (i.e., decrease rumen ammonia, acetate, and butyrate production and enhance the production of the gluconeogenic precursor propionate) (38). Therefore, the potential effects of monensin on glucose production are of great importance to early lactation dairy cows in a negative energy balance and at risk for ketosis.

In previous experiments, monensin in the diets of lactating dairy cows was found to decrease the incidence of subclinical and clinical ketosis (33, 37) and to decrease blood BHBA concentrations (1, 37). In other experiments with lactating dairy cows, the effects of monensin on reproductive performance, milk production (1, 20), digestion of nutrients, and rumen changes (19) have also been examined.

The hypothesis for this experiment was that the monensin controlled-release capsule (**CRC**) would decrease the occurrence of subclinical ketosis in early lactation dairy cows.

## MATERIALS AND METHODS

### Animal Utilization and Management

A total of 41 multiparous Holstein cows, ranging from 3 to 7 yr of age, and 11 primiparous Holstein cows were used and housed at the Elora Dairy Research Station (Elora, ON, Canada). The cows were fed a 50:50 (wt/wt) combined ration of a dry

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cow total mixed diet and an early lactation total mixed diet (balanced for 40 kg of milk/d and 3.9% fat) (Table 1). After parturition, cows were fed the early lactation diet only (Table 2). Cows were fed twice daily at 0500 and 1300 h; orts were measured once daily to calculate feed intake. Cows were milked twice daily at 0500 and 1630 h.

TABLE 1. Ingredient and chemical composition of the combined TMR fed to prepartum cows on a DM basis.

| Composition | |
|---|---|
| | (% of dietary DM) |
| Ingredient | |
| Mixed dry hay | 11.93 |
| Haylage 1 | 7.73 |
| Haylage 2 | 15.09 |
| Corn silage | 23.80 |
| High moisture corn | 24.46 |
| Dry corn distillers | 2.29 |
| Dehydrated alfalfa pellet | 2.29 |
| Soybean meal pellet | 3.26 |
| 1:1 (wt/wt) Gluten and 48% soybean meal pellet | 6.59 |
| Limestone | 0.60 |
| Bicarbonate | 0.38 |
| Lactating cow premix[1] | 0.92 |
| Dry cow premix[2] | 0.33 |
| NaCl | 0.33 |
| Chemical | |
| DM, % | 46.86 |
| | (% of DM) |
| CP (N × 6.25) | 14.38 |
| Soluble protein | 6.99 |
| Soluble protein,[3] % of CP | 48.57 |
| Undegradable insoluble protein,[4] bypass estimate as a % of CP | 25.72 |
| ADF | 30.33 |
| NDF | 44.48 |
| Ca | 0.83 |
| P | 0.46 |
| K | 1.69 |
| Mg | 0.26 |
| Na | 0.39 |
| Zn, ppm | 190.68 |
| Mn, ppm | 117.20 |
| Cu, ppm | 41.59 |
| Fat | 2.58 |
| $NE_L$,[5] Mcal/kg | 1.50 |
| Ca:P | 1.83:1 |

[1]Contained 11% Ca, 15% P, 12.5% Mg, 2.0% S, 2% K, 1200 mg/kg of Fl, 100 mg/kg of I, 1500 mg/kg of Cu, 3300 mg/kg of Mn, 5000 mg/kg of Zn, 36 mg/kg of Co, 22 mg/kg of Se, 550,000 IU/kg of vitamin A, 220,000 IU/kg of vitamin D, and 3100 IU/kg of vitamin E (Floradale Feed Mill Ltd., Floradale, ON, Canada).

[2]Contained 15% Ca, 12% P, 6.0% Mg, 2.0% S, 1000 ng/kg of Fl, 100 mg/kg of I, 2200 mg/kg of Cu, 5000 mg/kg of Mn, 7500 mg/kg of Zn, 40 mg/kg of Co, 40 mg/kg of Se, 675,000 IU/kg of vitamin A, 225,000 IU/kg of vitamin D, and 7200 IU/kg of vitamin E (Floradale Feed Mill Ltd., Floradale, ON, Canada).

[3]Percentage of soluble protein/percentage of CP × 100.

[4]100 – percentage of soluble protein/percentage of CP/2.

[5]Estimated using equations and values according to the NRC (27).

## Experimental Procedures

All experimental cows were randomly assigned to one of two treatments: a monensin CRC (Rumensin CRC®; Provel Division, Eli Lilly Canada Inc., Guelph, ON, Canada) or a placebo CRC (Provel Division, Eli Lilly Canada Inc., Guelph, ON, Canada). The monensin CRC contained 32 g of monensin sodium blended into a hexaglycerol distearate matrix core. A capsule delivers ($\bar{X} \pm SD$) 335 ± 33 mg/d of monensin for approximately 100 d. The placebo CRC was identical to the monensin CRC but contained no monensin sodium in the core. The CRC was administered 3 wk prior to the expected calving date (Figure 1). Following calving, cows were allowed to eat for ad libitum intake for 2 wk. For the next 3 wk of lactation, all cows were restricted to 90% of their ad libitum intake to induce subclinical ketosis. This restriction was determined in a previous experiment (18) in which 85% of the experimental cows were induced to show subclinical ketosis with up to 10% feed restriction. Ad libitum intake was calculated using the mean feed intake during the 2nd wk of lactation. Commencing at wk 6 of lactation, all cows were returned to ad libitum intake and completed the experimental period at the end of wk 6. Subclinical ketosis was diagnosed based on the BHBA concentration in blood serum. The threshold concentration was 1200 $\mu$mol/L (11, 28).

## Feed Sample Collection and Analysis

Samples of the combined prepartum diet (Table 1) and the early lactation diet (Table 2) were collected twice weekly and frozen at –20°C until composition analysis. Samples of the TMR were composited by experimental week and analyzed using wet chemistry at the Agri-Food Laboratory (Guelph, ON, Canada). Feed samples were analyzed for DM, CP (2), soluble protein (32), ADF (2, 42), NDF (16), crude fat (3), and minerals (Ca, P, K, Mg, Na, Zn, Mn, and Cu) (2).

## Blood Collection and Analysis

Blood samples were obtained by coccygeal venipuncture at the time the CRC was administered and twice weekly for the entire experimental period. All blood samples were taken in the morning, approximately 3 h postfeeding. Blood serum was immediately sent for analysis to the Department of Pathobiology, Ontario Veterinary College (Guelph, ON,

Canada). A BHBA reagent (Sigma Diagnostics, St. Louis, MO) and Dart glucose reagent (Coulter Electronics Corp., Hialeah, FL) were used to determine serum concentrations of the respective analysates with the Coulter Dacos biochemistry analyzer (Coulter Electronics Corp.).

## Milk Collection and Analysis

Milk samples were collected once weekly and were sent immediately for composition analysis to the Ontario Ministry of Agriculture, Food and Rural Affairs, Central Milk Testing Laboratory (Guelph, ON, Canada). Concentrations of fat, protein, and lactose were determined by near infrared analysis with the Foss System 4000 (Foss Electric, Hillerød, Denmark).

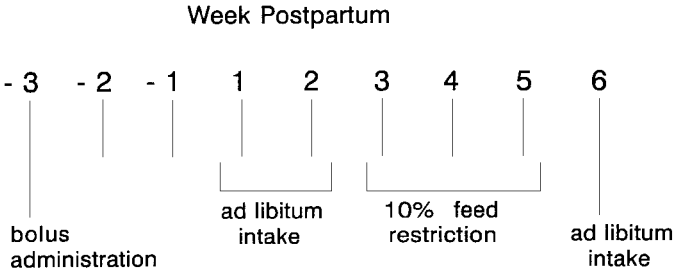


Figure 1. Schematic of the experimental protocol used to induce subclinical ketosis and examine the antiketogenic effects of the monensin controlled-release capsule.

TABLE 2. Ingredient and chemical composition of the TMR fed to lactating cows on a DM basis.

| Composition | |
|---|---|
| | (% of dietary DM) |
| Ingredient | |
| Mixed dry hay | 3.53 |
| Haylage 2 | 21.84 |
| Corn silage | 21.40 |
| High moisture corn | 31.22 |
| Dry corn distillers | 3.45 |
| Dehydrated alfalfa pellet | 3.41 |
| 1:1 (wt/wt) Gluten and 48% soybean meal pellet | 8.38 |
| Soybean meal | 3.61 |
| Limestone | 0.96 |
| Bicarbonate | 0.52 |
| Lactating cow premix[1] | 1.32 |
| NaCl | 0.36 |
| Chemical | |
| DM, % | 51.93 |
| | (% of DM) |
| CP (N × 6.25) | 17.03 |
| Soluble protein | 6.51 |
| Soluble protein,[2] % of CP | 38.25 |
| Undegradable insoluble protein,[3] bypass estimate as a % of CP | 30.88 |
| ADF | 20.93 |
| NDF | 33.14 |
| Ca | 0.96 |
| P | 0.56 |
| K | 1.32 |
| Mg | 0.31 |
| Na | 0.48 |
| Zn, ppm | 145.87 |
| Mn, ppm | 80.11 |
| Cu, ppm | 29.90 |
| Fat | 3.61 |
| $NE_L$,[4] Mcal/kg | 1.64 |
| Ca:P | 1.72:1 |

[1]Contained 11% Ca, 15% P, 12.5% Mg, 2.0% S, 2% K, 1200 mg/kg of Fe, 100 mg/kg of I, 1500 mg/kg of Cu, 3300 mg/kg of Mn, 5000 mg/kg of Zn, 36 mg/kg of Co, 22 mg/kg of Se, 550,000 IU/kg of vitamin A, 220,000 IU/kg of vitamin D, and 3100 IU/kg of vitamin E (Floradale Feed Mill Ltd., Floradale, ON, Canada).

[2]Percentage of soluble protein/CP × 100.

[3]100 – percentage of soluble protein/percentage of CP/2.

[4]Estimated using equations and values according to the NRC (27).

## BW and Body Condition Score

Body weight and body condition score were recorded at the time of CRC administration and once weekly for the duration of the experimental period. Body condition score was evaluated by three independent observers using a five-point scale with quarter-point intervals (14).

## Rumen Fluid Collection and Analysis

Rumen fluid was collected once weekly at 3 to 4 h after the morning feeding. Samples were taken prior to CRC administration, 10 d after CRC administration, and during wk 1, 2, 3, 4, and 6 of lactation. A vacuum pump and stomach tube were used to collect approximately 200 ml of rumen fluid. A 45-ml subsample was filtered, and the pH of the filtrate was immediately measured using a Fisher Accumet pH meter (model 610; Fisher Scientific, Fairlawn, NJ) calibrated with a buffer solution (pH 7.0; Fisher Scientific).

The concentration of ammonia nitrogen in the rumen fluid was determined using the indophenol-blue colorimetric method according to Novozamsky et al. (29).

The volatile fatty acid concentrations were determined on a Varian GC3400 using an 8000 autosampler and a Varian 654 data system (Varian Canada, Mississauga, ON, Canada). A fused silica capillary column was used (15 m × 0.53 mm i.d.; Nukol; Supelco Canada Ltd., Mississauga, ON, Canada). The injection and ionization temperatures were 200°C, and the carrier gas was helium. The iso-initial

column temperature was 45°C, and the initial hold time was 4 min. The final temperature was 85°C, which was reached by gradually increasing the temperature at a rate of 5°C/min with a hold time of 10 min.

### Statistical Analysis

Data were tested by repeated measures analysis of variance for the polynomial contrasts across time and the mean across time according to the following statistical model:

$$Y_{ijkl} = \mu + T_i + A_j + (T \times A)_{ij} + \beta X_{ijk} + \epsilon_{ijkl}$$

where

$Y_{ijkl}$ = mean or contrast of response variable,
$\mu$ = true mean,
$T_i$ = fixed effect of treatment (i = 1, 2),
$A_j$ = fixed effect of age (j = 0, 1),
$(T \times A)_{ij}$ = effect of interaction of treatment and age,
$\beta X_{ijk}$ = covariate (k = 1, 2, 3, 4, 5, 6, . . . , 12), and
$\epsilon_{ijkl}$ = experimental error.

The covariate was the initial measurement taken prior to administration of treatment for the blood, volatile fatty acids, pH, ammonia, BW, and body condition score response variables.

Milk production and composition, DMI, and energy balance data were tested by repeated measures analysis of variance for the polynomial contrasts across time and the mean across time according to the following statistical model:

$$Y_{ijkl} = \mu + T_i + A_j + (T \times A)_{ij} + \epsilon_{ijkl}$$

where

$Y_{ijkl}$ = mean of response variable,
$\mu$ = true mean,
$T_i$ = fixed effect of treatment (i = 1, 2),
$A_j$ = fixed effect of age (j = 0, 1),
$(T \times A)_{ij}$ = effect of interaction of treatment and age, and
$\epsilon_{ijkl}$ = experimental error.

The 2-wk prepartum period and the 6-wk postpartum period were analyzed separately. Based on residual plots, log and square root data transformations were performed on milk and blood variables prior to analysis to improve data homogenicity of variance. Least squares means were calculated for the treatment and age effects in the model for all response variables. Least squares means were plotted to explain significant treatment effects, and effects of the interaction of treatment and age were detected for the polynomial contrasts over time. Except for pH measurements, rumen fluid data were available from only 9 cows treated with a monensin CRC and 6 cows treated with a placebo CRC. In each of these treatment groups, 2 primiparous cows were included. Significant effects were declared at $P < 0.05$. All statistical models were solved using the general linear models procedure of SAS (36).

## RESULTS

### Milk Production and Milk Components

The least squares means for daily milk production and milk composition data for the postpartum experimental period are presented in Table 3, and no treatment effects were found ($P > 0.05$). The least squares mean milk production averaged 23 ± 2 kg/d for the primiparous cows and 32 ± 1 kg/d for the multiparous cows during the 6 wk of lactation (Table 3).

### DMI, Net Energy Balance, BW, and Body Condition Score

The least squares means of daily DMI, net energy balance, BW, and body condition score during the prepartum period and the postpartum period are given in Tables 4 and 5, respectively. All of these parameters were unaffected by treatment ($P > 0.05$).

### Rumen pH, Volatile Fatty Acids, and Ammonia Nitrogen

The least squares means for pH, volatile fatty acids, and ammonia concentrations for the postpartum period are given in Table 6. Rumen pH for the postpartum period was higher ($P < 0.05$) for cows treated with monensin. A treatment effect was found for the butyrate concentration; the concentration was lower ($P \leq 0.05$) for the cows treated with monensin. The rumen concentrations of acetate and propionate were not different ($P > 0.05$) for cows treated with monensin or with the placebo. The acetate to propionate ratio tended ($P < 0.06$) to be higher for cows treated with a placebo.

TABLE 3. Least squares means and treatment effects for milk production and milk components during the first 6 wk of lactation in primiparous and multiparous Holstein cows that received a monensin controlled-release capsule or a placebo controlled-release capsule at 3 wk prepartum.

| | Primiparous | | Multiparous | | | Effect | | |
|---|---|---|---|---|---|---|---|---|
| Variable | Monensin (n = 7) | Placebo (n = 4) | Monensin (n = 22) | Placebo (n = 19) | SE | Age (A) | Treatment (T) | T × A |
| Milk, kg/d | 23.53 | 23.13 | 33.27 | 32.55 | 5.24 | 0.0001 | 0.76 | 0.93 |
| Fat,[1] % | 2.06 | 2.01 | 2.10 | 2.18 | 0.19 | 0.12 | 0.79 | 0.29 |
| Protein,[2] % | 1.07 | 1.05 | 1.07 | 1.09 | 0.06 | 0.33 | 0.97 | 0.41 |
| Lactose,[1] % | 2.18 | 2.16 | 2.15 | 2.13 | 0.05 | 0.05 | 0.33 | 0.89 |

[1]Data were log-transformed.

[2]Data were transformed by square root.

## Serum Biochemistry

The least squares mean serum BHBA concentration during the 2-wk prepartum period was not different between cows on each treatment, although cows treated with monensin tended ($P < 0.06$) to have a lower concentration than did cows treated with the placebo (Table 7). The least squares mean serum BHBA concentration during the 6-wk postpartum period was lower ($P < 0.01$) in cows treated with monensin (Table 8). The changes in the mean serum BHBA concentration by week postpartum are shown in Figure 2. During wk 1, 2, and 6, the mean serum BHBA concentration was <1200 $\mu$mol/L for cows treated with monensin; these weeks corresponded to the weeks during which cows were fed for ad libitum intake. The cows treated with a placebo had a serum BHBA concentration that exceeded 1200 $\mu$mol/L throughout the postpartum period and peaked at 2700 $\mu$mol/L at the end of the restriction period (Figure 2). The maximum BHBA concentration observed in cows treated with monensin was 1800 $\mu$mol/L, which occurred during the final week of the restriction period (Figure 2).

No effect of treatment on the least squares mean serum glucose concentration was detected during the prepartum period. A decrease in serum glucose concentration occurred for cows on both treatments subsequent to parturition, and the least squares mean during the 6-wk postpartum period was higher ($P < 0.001$) for cows treated with monensin (Table 8). Changes in the mean serum glucose concentration by week postpartum are shown in Figure 3. The decline in glucose concentration was more severe in cows treated with the placebo and reached a low of 2.4 mmol/L during wk 4 of lactation; for cows treated with monensin, glucose concentration decreased to a low of 2.9 mmol/L (Figure 3). During wk 5 and 6, the glucose concentration increased for cows on both treatments. However, only the cows treated with monensin had glucose concentrations that exceeded 3.0 mmol/L after cows returned to ad libitum consumption during the last week of the experiment (Figure 3).

The least squares mean serum urea concentration during the 2-wk prepartum period was higher ($P < 0.05$) in cows treated with monensin (Table 7). No

TABLE 4. Least squares means and treatment effects for DMI, net energy balance, BW, and body condition score (BCS) during the 2-wk prepartum period in primiparous and multiparous Holstein cows that received a monensin controlled-release capsule or a placebo controlled-release capsule at 3 wk prepartum.

| | Primiparous | | Multiparous | | | | Effect | | |
|---|---|---|---|---|---|---|---|---|---|
| Variable | Monensin (n = 7) | Placebo (n = 4) | Monensin (n = 22) | Placebo (n = 19) | SE | Cov[1] | Age (A) | Treatment (T) | T × A |
| DMI, kg/d | 8.59 | 7.13 | 11.43 | 11.05 | 1.61 | . . . | 0.0001 | 0.14 | 0.39 |
| Net energy balance,[2] Mcal/d | –1.92 | –3.95 | 2.35 | 1.92 | 2.74 | . . . | 0.0001 | 0.25 | 0.45 |
| BW, kg | 747.29 | 738.47 | 764.90 | 762.75 | 21.31 | 0.0001 | 0.05 | 0.47 | 0.68 |
| BCS[3] | 3.61 | 3.70 | 3.57 | 3.62 | 0.15 | 0.0001 | 0.41 | 0.23 | 0.68 |

[1]Covariate; the initial measurement prior to bolus administration.

[2]Estimated using equations from the NRC (27).

[3]Measurements were scored on a five-point scale with quarter-point intervals where 1 = thin to 5 = fat (14).

TABLE 5. Least squares means and treatment effects for DMI, net energy balance, BW, and body condition score (BCS) during the 6-wk postpartum period in primiparous and multiparous Holstein cows that received a monensin controlled-release capsule or a placebo controlled-release capsule at 3 wk prepartum.

| | Primiparous | | Multiparous | | | | Effect | | |
|---|---|---|---|---|---|---|---|---|---|
| Variable | Monensin (n = 7) | Placebo (n = 4) | Monensin (n = 22) | Placebo (n = 19) | SE | Cov[1] | Age (A) | Treatment (T) | T × A |
| DMI, kg/d | 11.73 | 10.49 | 14.49 | 14.52 | 2.79 | . . . | 0.001 | 0.54 | 0.52 |
| Net energy balance,[2] Mcal/d | –9.14 | –10.43 | –12.61 | –13.01 | 3.41 | . . . | 0.02 | 0.48 | 0.71 |
| BW, kg | 615.75 | 607.75 | 648.82 | 627.23 | 32.62 | 0.0001 | 0.10 | 0.20 | 0.56 |
| BCS[3] | 2.97 | 3.03 | 2.90 | 2.81 | 0.21 | 0.0001 | 0.13 | 0.82 | 0.36 |

[1]Covariate; initial measurement prior to bolus administration.

[2]Estimated using equations from the NRC (27).

[3]Measurements were scored on a five-point scale with quarter-point intervals where 1 = thin to 5 = fat (14).

effect of treatment was detected on the mean serum urea concentration during the 6-wk postpartum period, but the monensin treatment tended ($P < 0.08$) to have a higher concentration (Table 8).

## DISCUSSION

The effects of the monensin CRC on milk production and milk components have not been studied extensively in dairy cows. In the current experiment, monensin had no effect on milk production, milk protein, or milk fat, which is in agreement with the results of other experiments (1, 20, 24, 37).

The lack of response in milk production in the current experiment might have been caused by the imposed period of feed restriction. During the experiment, milk production was maintained between 30 and 35 kg/d at a time when production usually increases. The decrease in DMI that was imposed over 3 wk was not severe enough to cause the dramatic decline in milk production that was found in cows during severe energy deficit (22). However, milk production was probably impaired (4). Experiments using feed restriction to induce ketosis (12, 15, 20) have shown decreases in milk production of 7 to 10 kg/d. Those studies indicated that high milk production cannot be maintained over long periods by mobilization of tissue reserves (22).

The shift in rumen fermentation and volatile fatty acid pattern as a result of monensin has been well documented in the literature. Studies with steers (31), dairy cattle (37), and sheep (30) consistently demonstrated increases in the concentration of propionate but not always a significant decrease in acetate and butyrate concentrations. In contrast, the current study found the monensin treatment had no effect on propionate ($P \geq 0.63$) and acetate ($P \geq 0.21$) concentrations but decreased butyrate concentration ($P \leq 0.05$).

TABLE 6. Least squares means and treatment effects for rumen pH, volatile fatty acids, and ammonia nitrogen during the 6-wk postpartum period in primiparous and multiparous Holstein cows that received either a monensin controlled-release capsule or a placebo controlled-release capsule at 3 wk prepartum.

| | Primiparous | | Multiparous | | | | Effect | | |
|---|---|---|---|---|---|---|---|---|---|
| Variable | Monensin (n = 7) | Placebo (n = 4) | Monensin (n = 22) | Placebo (n = 19) | SE | Cov[1] | Age (A) | Treatment (T) | T × A |
| pH | 6.85 | 6.50 | 6.66 | 6.61 | 0.24 | 0.006 | 0.67 | 0.03 | 0.11 |
| Volatile fatty acids, m*M* | | | | | | | | | |
| Total | 73.39 | 81.97 | 68.85 | 74.23 | 11.90 | 0.01 | 0.44 | 0.36 | 0.84 |
| Acetate (A) | 45.96 | 54.54 | 41.81 | 46.33 | 8.02 | 0.02 | 0.24 | 0.21 | 0.69 |
| Propionate (P) | 15.85 | 14.29 | 15.45 | 15.00 | 3.27 | 0.01 | 0.95 | 0.64 | 0.80 |
| Butyrate | 7.69 | 10.61 | 7.96 | 8.74 | 1.39 | 0.01 | 0.40 | 0.05 | 0.26 |
| Other[2] | 3.79 | 2.70 | 3.61 | 4.13 | 0.66 | 0.05 | 0.30 | 0.60 | 0.20 |
| A:P | 3.05 | 3.75 | 2.95 | 3.32 | 0.41 | 0.07 | 0.39 | 0.06 | 0.55 |
| Ammonia nitrogen, mg/dl | 56.30 | 77.39 | 42.44 | 68.39 | 18.05 | 0.61 | 0.32 | 0.12 | 0.85 |

[1]Covariate; initial measurement prior to bolus administration.

[2]Sum of isobutyrate, isovalerate, and valerate volatile fatty acids.

TABLE 7. Least squares means and treatment effects of selected serum component concentrations during the 2-wk prepartum period in primiparous and multiparous Holstein cows receiving a monensin controlled-release capsule or a placebo controlled-release capsule at 3 wk prepartum.

| | Primiparous | | Multiparous | | | | Effect | | |
|---|---|---|---|---|---|---|---|---|---|
| Variable | Monensin (n = 7) | Placebo (n = 4) | Monensin (n = 22) | Placebo (n = 19) | SE | Cov[1] | Age (A) | Treatment (T) | T × A |
| BHBA,[2] μmol/L | 6.56 | 6.68 | 6.76 | 6.93 | 0.22 | 0.77 | 0.003 | 0.06 | 0.76 |
| Glucose,[3] mmol/L | 1.94 | 1.87 | 1.82 | 1.88 | 0.08 | 0.0001 | 0.11 | 0.94 | 0.03 |
| Urea,[2] mmol/L | 1.46 | 1.24 | 1.46 | 1.41 | 0.19 | 0.0001 | 0.20 | 0.05 | 0.19 |

[1]Covariate; initial measurement prior to bolus administration.

[2]Data were log-transformed.

[3]Data were transformed by the square root.

The significantly higher pH observed for cows treated with monensin has been observed elsewhere under both in vitro (10) and in vivo conditions (26). Monensin inhibits lactate-producing bacteria, which proliferate in abundant starch conditions. Also, lactate users are resistant to monensin (34). The production of lactate is then decreased, and rumen pH increases (34, 38). Therefore, monensin could be beneficial in the reduction of lactic acidosis in dairy cows that are fed high concentrate diets in early lactation.

Another characteristic of monensin is the potential to decrease rumen ammonia concentration and protein degradation, indicating a protein sparing effect as dietary protein escapes rumen digestion (7, 38). Monensin decreases rumen ammonia concentration because Gram-positive bacteria that are sensitive to monensin have a higher specific activity for ammonia production than do Gram-negative bacteria that are resistant to monensin. (35). Experiments with sheep (30) and dairy cows (19) have observed lower rumen ammonia concentrations (63 and 53%, respectively) when monensin was administered than when a control treatment was administered. A similar trend ($P > 0.12$) appeared in the current experiment; a 32% decrease in rumen ammonia was observed for cows treated with monensin. However, the limited number of cows decreased the power of the analysis, and the result was not statistically significant.

The significantly lower BHBA concentration and higher glucose concentration in the monensin cows could have resulted from the contributions of several metabolic pathways or actions of monensin (1). These actions include a decrease in butyrate concentration, an increase in propionate concentration, a glucogenic effect that was independent of rumen fermentation, and a decrease in the circulation of free fatty acids.

A decrease in the rumen butyrate concentration, facilitated by monensin, resulted in a sequential decrease in BHBA entry into the blood. Because butyrate was lower ($P \leq 0.05$) in cows treated with monensin, this pathway could have contributed to the changes observed in the blood metabolites. Up to 75% of rumen butyrate is converted to BHBA by the rumen epithelium as it is absorbed across the rumen wall (5). In fed, lactating cows, butyrate could potentially contribute up to 50% of total ketone body production as opposed to 0% of the output during feed

TABLE 8. Least squares means and treatment effects of selected serum component concentrations during the 6-wk postpartum period in primiparous and multiparous Holstein cows receiving a monensin controlled-release capsule or placebo controlled-release capsule at 3 wk prepartum.

| | Primiparous | | Multiparous | | | | Effect | | |
|---|---|---|---|---|---|---|---|---|---|
| Variable | Monensin (n = 7) | Placebo (n = 4) | Monensin (n = 22) | Placebo (n = 19) | SE | Cov[1] | Age (A) | Treatment (T) | T × A |
| BHBA,[2] μmol/L | 6.68 | 6.99 | 7.02 | 7.45 | 0.40 | 0.16 | 0.01 | 0.01 | 0.65 |
| Glucose,[3] mmol/L | 1.86 | 1.71 | 1.72 | 1.60 | 0.11 | 0.25 | 0.003 | 0.001 | 0.71 |
| Urea,[2] mmol/L | 1.50 | 1.36 | 1.53 | 1.39 | 0.22 | 0.003 | 0.71 | 0.08 | 0.97 |

[1]Covariate; initial measurement prior to bolus administration.

[2]Data were log-transformed.

[3]Data were transformed by the square root.

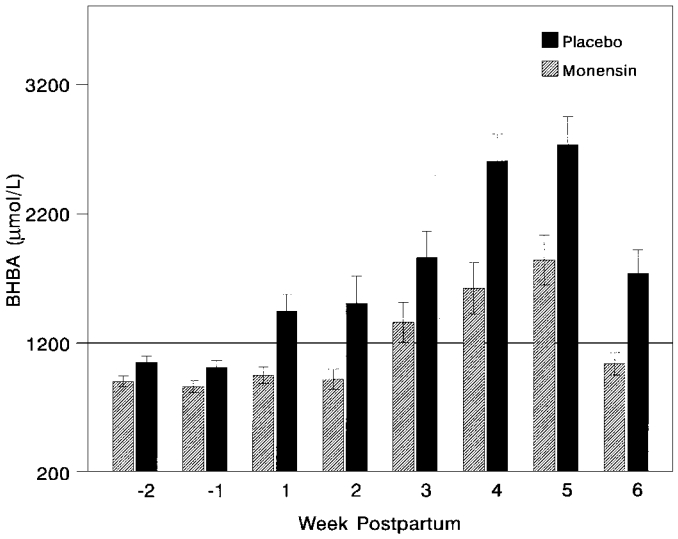


Figure 2. Mean (±SEM) serum BHBA concentration by week prepartum and postpartum for Holstein cows receiving a monensin controlled-release capsule (shaded bar; n = 29) or a placebo controlled-release capsule (solid bar; n = 23) at 3 wk prepartum. The threshold level used to determine subclinical ketosis was 1200 µmol/L.

deprivation, fasting, or when free fatty acids are the primary source of ketone body output (25).

The most common theory behind the action of monensin is the increase in rumen propionate and its subsequent role in the regulation of gluconeogenesis (43). The percentage of glucose that originates from propionate increases as more propionate is absorbed from the rumen (43). Lomax and Baird (25) calculated that propionate contributed 46% to the output of glucose by the liver in lactating cows in the fed state. However, Van Maanen et al. (43) found that the increase in glucose irreversible loss accounted for only 20% of the increase in propionate production. Therefore, the remaining 80% was thought to be oxidized to carbon dioxide or contributed carbon to the synthesis of other compounds. This alternative use of propionate would spare lactate and amino acids that are normally oxidized for processes that require energy and potentially contribute 25% to hepatic glucose output (25, 43).

Monensin may also provide a glucogenic effect that is independent of rumen fermentation to improve glucose production and decrease the BHBA concentrations (1). Haimoud et al. (19) found the intestinal digestion of undegraded rumen starch was higher when monensin was administered and found a higher concentration of glucose (3.6 vs. 3.1 m*M* for control animals). Therefore, the shift in site of starch digestion would allow more carbon to be absorbed as glucose and not as volatile fatty acids, a more efficient use of potential energy by the cow (9).

A decrease in free fatty acids that circulate in the blood is an indirect action of monensin and results from an increase in the glucose concentration and subsequently insulin concentration (1). During subclinical ketosis, Schwalm et al. (39) found that BHBA was positively correlated (0.86), and glucose was negatively correlated (–0.92), with NEFA ($P < 0.01$). Therefore, as BHBA decreases, fewer fatty acids are metabolized in the liver. The transition from subclinical to normal in the study by Schwalm et al. (39) was marked by an increase in glucose and triglycerides, but ketones, acetate, and NEFA were decreased. Free fatty acid concentrations might be assumed to be lower in the cows treated with monensin because of the significant changes observed in BHBA and glucose concentrations. However, with all of the possible mechanisms and actions of monensin considered, the changes in BHBA and glucose concentrations observed in this study center on an alteration of the rumen fermentation pattern at fixed levels of DMI.

In this experiment, the blood glucose concentration was approximately 15% higher in cows treated with monensin, which might have resulted from increased amino acid passage and absorption in the small intestine, particularly during the restriction period. Increased amino acid absorption in the small intestine increases availability for gluconeogenesis, contribut-

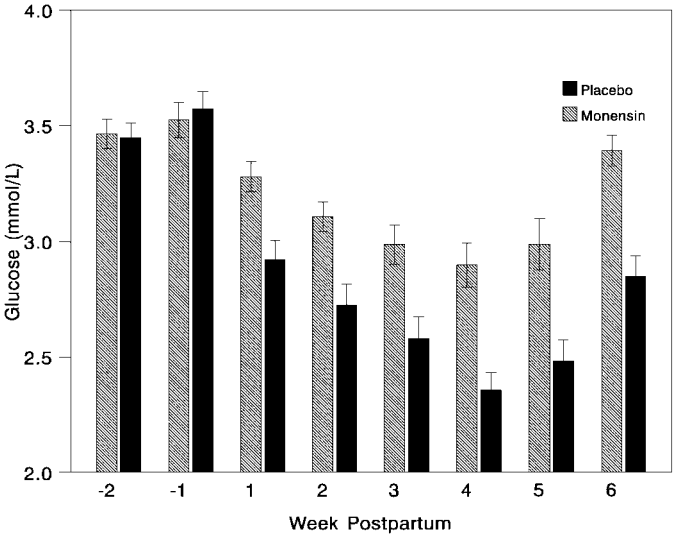


Figure 3. Mean (±SEM) serum glucose concentration by week prepartum and postpartum for Holstein cows receiving a monensin controlled-release capsule (shaded bar; n = 29) or a placebo controlled-release capsule (solid bar; n = 23) at 3 wk prepartum.

ing to the increase in urea, maintenance of glucose homeostasis, and the decrease in the amount of BHBA output. Blood urea N tended to be higher during lactation for cows treated with monensin ($P < 0.08$) than for cows treated with a placebo, which suggests that monensin increased the amount of dietary protein that reached the small intestine. In addition, Tyrrell et al. (41) found cows with a tissue energy balance of –1.1 Mcal/d and a tissue N balance of –21 g/d measured in open circuit respiration chambers at 100 d into lactation. This result suggests that the increase in urea may not only be from increased amino acid absorption in the small intestine but also from mobilization of muscle, the source of labile body protein in lactating animals (6).

Monensin was effective in decreasing the concentration of BHBA (Figure 2) and maintaining a higher glucose concentration (Figure 3) immediately postpartum throughout the restriction period and in the final week of the experiment. Similar effects have been found elsewhere under experimental and commercial conditions (1, 13, 37). In contrast, work by Stephenson et al. (40) suggested that monensin decreased plasma glucose and BHBA concentrations prior to calving in dairy cows, and no significant effects were found after parturition. An experiment using lasalocid, another ionophore, was not effective in the alteration of either BHBA or glucose concentrations using cows that were 16 d into lactation and were fed a diet varying in corn grain particle size (21). The lack of response might have been the higher DMI (19 kg/d), which subsequently resulted in an energy balance (–5 Mcal/d) that was not severely negative. In the current experiment, energy balance was between –10 and –15 Mcal/d in cows treated with the placebo and the monensin CRC, indicating the large energy deficit imposed by lactation. Inconsistencies with respect to the relationship among energy balance, BHBA, and glucose could be explained by diets that may differ in glucogenic capacity.

During the 3-wk restriction period, serum BHBA was above the 1200-$\mu$mol/L threshold for the cows treated with monensin (Figure 2). Without a decline in milk production, the restriction period was increasingly severe and augmented the mobilization of body tissues. The similar pattern of change observed in BW and body condition score has been observed elsewhere (23) in early lactating cows that were nonketotic, ketonemic, and clinically ketotic. The decrease observed in BW and body condition score during lactation indicates that body tissues were a source of energy and nutrients for cows on both treatments. However, these two parameters are not as sensitive as are blood metabolites in the demonstration of changes at the subclinical level.

The 1200-$\mu$mol/L threshold used in this experiment was very conservative, and the cows treated with monensin were subclinically ketotic only during the restriction period (Figure 2). Cows treated with the placebo were subclinically ketotic throughout the postpartum period (Figure 2). In agreement, Sauer et al. (37) found that monensin decreased the number of cows that experienced subclinical ketosis. In a commercial setting, monensin has also been beneficial in decreasing the effects of subclinical ketosis (13, 33). The effect of monensin on the decrease in the occurrence of subclinical ketosis is, therefore, not limited to the controlled experimental conditions of one herd.

## CONCLUSIONS

The ability of monensin to alter rumen fermentation pattern may benefit the high producing dairy cow that is prone to subclinical ketosis immediately postpartum. Monensin had no effect on milk production or milk composition. The rumen fermentation pattern was altered by monensin, as was evidenced by an increase in rumen pH and a decrease in butyrate concentration and the acetate to propionate ratio. Furthermore, the observed increase in serum urea in cows treated with monensin supports the protein sparing effect of monensin within the rumen and may suggest an increase in peripheral tissue protein degradation. This increase may also be because of an increase in amino acid utilization for gluconeogenesis. The biggest impact of monensin was a significantly lower blood BHBA concentration and a higher blood glucose concentration. The alteration of concentrations of these blood metabolites allowed for a decrease in the prevalence of subclinical ketosis. Results indicated that monensin may be a convenient management tool for the dairy producer. Monensin maintained energy related blood metabolites, reduced the occurrence of subclinical ketosis, and improved rumen health in the early lactation dairy cow.

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