Pre- and Postpartum Metabolizable Protein Supply Alters Performance of Multiparous Holstein Cows

T. A. Westhoff[1], T. R. Overton[2], M. E. Van Amburgh[2], J. N. Tikofsky[3], S. Mann[1]
[1]Department of Population Medicine and Diagnostic Sciences, Cornell University, Ithaca, NY
[2]Department of Animal Science, Cornell University, Ithaca, NY
[3]Purina Animal Nutrition, Gray Summit, MO

Introduction

Inadequate nutrient intake together with an increased nutrient demand to support fetal and mammary development as well as colostrum and milk synthesis drives transition cows into a negative energy and protein balance (Bell et al., 1995, Bell, 1995, Mann et al., 2016). Metabolizable protein (MP) requirements for multiparous, close-up dry cows were estimated at approximately 800 g/d (Husnain and Santos, 2019, NASEM, 2021). However, since the current estimate does not include the amino acid requirement for mammary growth and colostrum synthesis, prepartum MP supply might affect production of high-quality colostrum as well as early lactation milk production. Several authors have reported a positive early lactation milk production response when supplementing methionine and lysine (Batistel et al., 2017, Fehlberg et al., 2020) or increasing total prepartum MP supply (Farahani et al., 2017, Farahani et al., 2019). Milk yield during the first three weeks of lactation increased 3.4 ± 0.9 kg/d (7.5 ± 2.0 lb/d) when increasing prepartum MP supply from 849 to 1,200 g/d, but milk yield was not further increased when the prepartum diet supplied 1,387 g of MP/d (Farahani et al., 2017). Further increasing prepartum MP supply from 1,264 to 1,681 g/d resulted in a 3.7 ± 1.2 kg/d (8.2 ± 2.6 lb/d) lower milk yield from 1 to 12 wk (Zang et al., 2022). Moreover, Farahani et al., 2019 recently reported an interaction between pre- and postpartum MP supply suggesting milk yield during early lactation might depend on the supply of MP provided during both the close-up and fresh period.

During early lactation, mature Holstein cows producing a milk yield of 53 kg/d have an estimated MP requirement of 2,802 g/d. Numerous authors have demonstrated that cows respond to an increased postpartum MP supply with increased milk production (Larsen et al., 2014, Carder and Weiss, 2017, Tebbe and Weiss, 2021). Recently, Zang et al. (2021) observed a 2.5 ± 1.1 kg/d (5.5 ± 2.4 lb/d) increase in milk yield during the first three weeks of lactation when increasing MP supply from 2,227 to 2,513 g/d. However, the optimum supply of MP during the pre- and postpartum period remains unknown. Further, it is unclear whether the performance effect of altering MP supply fed during the close-up, fresh, or both periods persist beyond the treatment period. Identification of the optimum MP feeding strategy during the transition period might improve productive efficiency and increase farm profitability. As such, the objective of this study was to compare the effect of four MP feeding strategies on lactation performance, colostrum production, and the metabolic response to lactation.

Experimental design

Multiparous Holstein cows (n = 96) were enrolled in a randomized block design at the Cornell University Ruminant Center from May to November 2021. All animals were moved from a far-off dry cow pen to individual tie stalls between 35 and 42 d before expected calving and fed a far-off diet. At 28 d before expected calving, cows were blocked by calving date and balanced for parity and previous lactation 305-d mature equivalent milk production. Animals were randomly assigned within block to 1 of 4 dietary treatments groups consisting of a combination of a pre- and postpartum diet (Figure 1). The close-up TMR was formulated to contain either a control (C; 85 g of MP/kg DM) or high (H; 113 g of MP/kg DM) level of MP. Both prepartum diets were formulated to supply methionine (Met) and lysine (Lys) at 1.24 and 3.84 g/Mcal of metabolizable energy (ME), respectively. From calving to 21 days in milk (DIM), cows were fed a postpartum TMR formulated to contain either a control (C; 104 g of MP/kg DM) or high (H; 131 g of MP/kg DM) level of MP. Postpartum diets were formulated to supply Met and Lys at 1.15 and 3.16 g/Mcal of ME in both groups, respectively. The combination of a pre- and postpartum diet resulted in 4 treatment groups: 1) control-control (**CC**), 2) control-high (**CH**), 3) high-control (**HC**), and 4) high-high (**HH**), respectively. Cows were fed a common lactation diet from 22 to 42 DIM. Treatment diets were formulated using the Cornell Net Carbohydrate and Protein System v. 6.5.5 (AMTS.Cattle.Professional v. 4.17.0.0; AMTS LLC; Table 1; Van Amburgh et al., 2015).

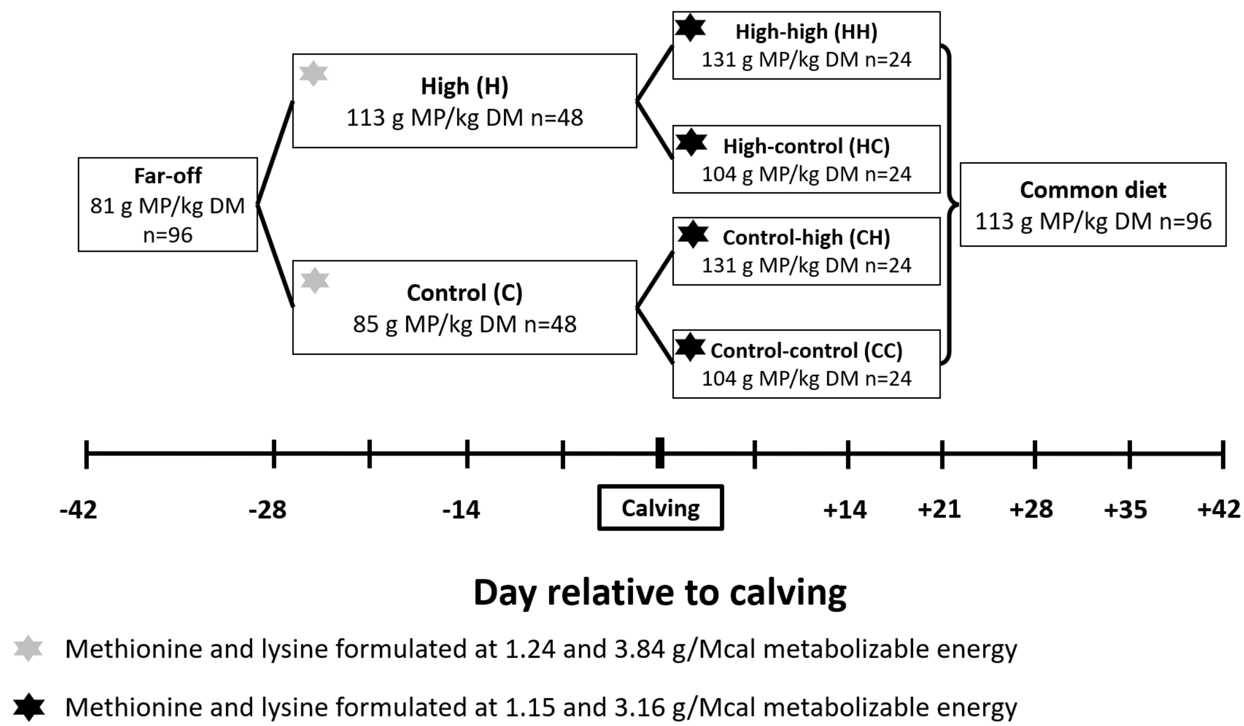

Figure 1. Schematic of treatment assignment.

Individual daily feed intake was recorded and DMI was calculated using the diet DM percentage collected weekly. Weekly, cows were weighed and BCS was determined by one investigator using a 5-point scale with 0.25-point increments (Edmonson et al., 1989). At calving, colostrum yield and Brix percentage were recorded by farm personnel. Cows were milked three times daily and milk yields were recorded until 42 DIM. Milk samples were collected once weekly for three consecutive milkings and submitted to a commercial laboratory (Dairy One Cooperative Inc.) for analysis of fat, true protein, lactose, total solids, and milk urea nitrogen (MUN) by Fourier Transform Infrared Spectroscopy (method 972.160; AOAC International, 2012). Metabolizable protein supply and balance as well as metabolizable energy balance were estimated weekly for each cow in AMTS.Cattle.Professional v. 4.17.0.0 using the calf birth weight, DCC, BW, and weekly DMI for prepartum estimates, and DIM, BW, weekly DMI, as well as milk yield, and milk composition for all postpartum estimates.

Table 1. Ingredient composition of diets.

	Treatment diets[1]				
	Prepartum		Postpartum		
Ingredient, % of DM	Control	High	Control	High	Lactation[2]
Conventional corn silage	45	45	-	-	37.1
BMR corn silage[3]	-	-	37.3	37.3	12.1
Haylage	-	-	17.8	17.8	12.5
Wheat straw	26.7	26.7	4.7	4.7	-
Corn grain	-	-	17.2	17.2	18.6
Grain mix[4]	-	-	-	-	10.8
Soybean meal solvent	3.5	3.3	3.9	0.9	8.6
Canola meal solvent	1.8	1.5	3.8	1.2	-
Amino Plus[5]	0.7	11.0	1.0	11.4	-
ProvAAl Lysine[6]	0.9	2.2	0.60	2.20	-
Smartamine M[7]	0.13	0.04	0.17	0.10	-
USA Lysine[8]	0.45	-	0.41	0.02	-
Citrus pulp dry	-	-	2.70	1.00	-
Soybean hulls ground	10.4	1.0	3.70	-	-
Wheat midds	2.9	1.2	1.6	0.90	-
Energy Booster 100[9]	-	-	1.5	1.6	-
Sodium bicarbonate	-	-	0.76	0.70	-
Salt white	0.21	0.19	0.62	0.65	-
MIN-AD[10]	0.89	0.86	-	-	-
Magnesium oxide	-	-	0.33	0.34	-
Magnesium sulfate	0.89	0.86	-	-	-
Calcium carbonate	1.83	2.23	1.33	1.42	-
Calcium sulfate dihydrate	0.78	0.10	-	-	-
Dicalcium phosphate	0.75	0.48	-	-	-
Vitamin E mix[11]	-	-	0.01	0.01	-
Monensin[12]	0.02	0.02	0.01	0.01	-
Choline[13]	0.43	0.41	0.30	0.29	-
Animate[14]	1.27	2.48	-	-	-
Vitamin/mineral mix[15]	0.45	0.43	-	-	-
Vitamin/mineral mix[16]	-	-	0.26	0.26	-

[1]Prepartum TMRs, fed starting at 28 d before expected calving, were formulated to contain methionine and lysine at 1.24 and 3.84 g/Mcal metabolizable energy (ME), respectively and supply a control (C; 85 g MP/kg DM) or high (H; 113 g MP/kg DM) level of estimated metabolizable protein (MP). Postpartum TMRs, fed from 0 to 21 DIM, were formulated to contain methionine and lysine at 1.15 and 3.16 g/Mcal ME, respectively and supply control (C; 104 g MP/kg DM) or high (H; 131 g MP/kg DM) level of estimated MP.

[2]From 22 to 42 DIM, cows were fed a lactation diet that contained methionine and lysine at 1.01 and 2.88 g/Mcal ME, respectively and supplied 113 g of estimated MP/kg DM.

[3]Brown mid-rib corn silage

[4]Contains 28.7% Amino Plus (Ag Processing Inc.), 13.1% blood meal, 11.8% sodium sesquicarbonate, 9.4% chocolate dairy mix, 9.0% soybean hulls, 7.3% calcium carbonate, 4.2% bypass fat (Cargill Animal Nutrition), 3.5% salt, 3.0% urea, 2.7% Celmanax SCP (Arm and Hammer), 1.8% mono dicalcium phosphate, 1.7% magnesium oxide 54%, 1.7% potassium magnesium sulfate, 0.50% Smartamine M (Adisseo), 0.5% Diamond V XPC (Diamond V North America), 0.40% selenium 0.06%, 0.26% Zinpro 5 (Zinpro Corp.), 0.17% Dairy ADE (Contains 30,073 kIU/kg vitamin A, 5,783 kIU/kg vitamin D, and 92,534 IU/kg vitamin E; Cargill Animal Nutrition), 0.161% trace mineral mix (Contains 2.959% Ca, 17.503% S, 0.657% Cl, 0.231% Mg, 0.141% Na, 0.128% K, 160,210.53 mg/kg Zn, 142,105.26 mg/kg Mn, 23,684.21 mg/kg Cu, 3,526.32 mg/kg Co, 3,157.90 mg/kg I, and 196.84 mg/kg Fe; Cargill Animal Nutrition), 0.057% monensin (Monovet 90; Huvepharma), 0.05% Vitamin E (Contains 510,750 IU/kg), 0.009% copper sulfate.

[5]Heat-treated soybean meal (Ag Processing Inc.)

[6]Protein supplement (Perdue AgriBusiness)

[7]Methionine protected with pH sensitive coating (Adisseo)

[8]Rumen bypass lysine (Kemin Industries Inc.)

[9]Prilled fatty acids (Milk Specialties Co.)

[10]Magnesium limestone (Papillon Agricultural Company)

[11]Contains 18.6% Ca, 0.61% Mg, and 500,444.2 ppm Vitamin E

[12]Rumensin 90 (Elanco Animal Health)

[13]Reashure (Balchem Co.)

[14]Anionic supplement (Phibro Animal Health Corp.)

[15]Contains 26.38% Ca, 0.12% P, 0.42% Mg, 0.21% K, 3.17% S, 275 ppm Fe, 17,569 ppm Zn, 1,727 ppm Cu, 5,688 ppm Mn, 55 ppm Se, 97.3 ppm Co, 98.18 ppm I, 2,341.71 kIU/kg vitamin A, 566.68 kIU/kg vitamin D, 22,658.58 kIU/kg vitamin E.

[16]Contains 28.16% Ca, 0.33% Mg, 0.09% K, 2.32% S, 870 ppm Fe, 18,235 ppm Zn, 3,291 ppm Cu, 15,995 ppm Mn, 132 ppm Se, 550 ppm Co, 390 ppm I, 1,984.5 kIU/kg vitamin A, 551.25 kIU/kg vitamin D, 9,481.5 kIU/kg vitamin E.

Blood samples were collected twice weekly from calving to 28 DIM, and once weekly from 29 to 42 DIM. Plasma concentrations of ß-hydroxybutyrate (BHB) were measured with samples warmed to 37 ºC using a Precision Xtra point-of-care device (Abbott). Samples obtained from 3 to 10 DIM with BHB ≥ 1.2 mmol/L were defined as an event of hyperketonemia (McArt et al., 2013). Plasma concentrations of nonesterified fatty acids (NEFA) were determined in duplicate by enzymatic colorimetric analysis (HR Series NEFA-HR (2), Wako Life Sciences).

The longissimus dorsi muscle diameter and subcutaneous backfat thickness were determined by ultrasound at -30, -14, -7, 7, 21, and 40 d relative. All measurements were performed on the right side of the animal using a 5-9 MHz broadband linear transducer on a portable ultrasound (IBEX PRO; E.I. Medical Imaging). The skin near the loin and thurl region were brushed and 70% alcohol was applied as a coupling agent. Longissimus dorsi diameter was measured perpendicular to the spine as the largest diameter between muscular fascial layers at the fourth transverse process and backfat was measured approximately 10 cm caudal of the tuber coxae as the distance between the profound fascia above the gluteus medius and the surface, excluding the measurement of the skin. The hair at the location of the ultrasound transducer during the first timepoint was clipped to ensure repeated placement at subsequent timepoints. Each measurement was performed in triplicate and averaged for analysis.

Statistical analysis was performed in SAS v. 9.4 (SAS Institute Inc.) for three separate periods (-28 to 0 days relative to calving, 1 to 21 DIM, and 22 to 42 DIM) with the exception that ultrasound data was analyzed separately pre- and postpartum. Mixed effects ANOVA were conducted in PROC MIXED to explore differences in outcome variables not repeated over time. Repeated measures ANOVA were performed for outcome variables repeated over time using PROC MIXED with the fixed effects of treatment, time, parity (2 vs. ≥ 3), and the interaction of treatment and time. The models included the random effect of enrollment block and repeated effect of time with the subject of cow. Baseline covariate measurements were included in all models when available. Tukey's post hoc test was used to adjust for multiple comparisons.

Results

Diet composition

The chemically analyzed composition of the diets is in Table 2. Model predicted MP concentration was 87 and 115 g/kg DM for the prepartum C and H diets and 101 and 127 g/kg DM for the postpartum C and H diets, respectively. Estimated rumen degradable protein increased approximately 1.5 % of DM in the prepartum H compared to the prepartum C diets. During the postpartum period, concentrations of rumen degradable protein were estimated at approximately 9.0 % of DM for the C and H diets, respectively. Estimated rumen undegradable protein increased from 3.7 to 7.1 % of DM and 5.0 to 8.3 % of DM in the C compared to H diets fed pre- and postpartum, respectively.

Prepartum period

Results from the prepartum period are in Table 3. Dry matter intake (% of BW) did not differ by treatment ($P = 0.29$) but feeding the H diet resulted in a greater MP supply compared to C (1,606 ± 27 vs. 1,180 ± 27 g of MP/d; $P < 0.01$), respectively. Cows fed a greater MP supply had a larger prepartum BW gain (4.78 ± 0.45 vs. 3.21 ± 0.45 % of BW; $P < 0.01$) compared to cows fed the C diet, yet BCS ($P = 0.76$), longissimus dorsi diameter (C: 42.8 ± 0.5; H: 43.1 ± 0.5 mm; $P = 0.73$), and backfat thickness (C: 5.7 ± 0.3; H: 6.2 ± 0.3 mm; $P = 0.15$) did not differ by treatment. In agreement with previously published work (Farahani et al., 2017, Farahani et al., 2019, Akhtar et al., 2022), prepartum MP supply did not affect colostrum yield or Brix percentage ($P \geq 0.76$) in the current study.

Table 2. Nutrient composition of diets.

Component[3] (mean ± SD)	Treatment diets[1]				
	Prepartum		Postpartum		
	Control	High	Control	High	Lactation[2]
DM, %[4]	46.4 ± 3.6	46.3 ± 3.3	42.6 ± 2.5	43.1 ± 2.4	42.5 ± 1.4
CP, % of DM	11.5 ± 0.4	14.6 ± 1.0	14.5 ± 0.3	15.9 ± 0.5	15.4 ± 0.6
aNDF, % of DM	43.4 ± 1.0	41.3 ± 0.4	33.3 ± 0.5	32.0 ± 0.5	27.6 ± 0.8
ADF, % of DM	28.2 ± 0.7	26.0 ± 0.3	21.7 ± 0.4	20.2 ± 0.5	17.4 ± 0.7
Lignin, % of DM	3.4 ± 0.2	3.0 ± 0.3	2.6 ± 0.2	2.2 ± 0.2	1.8 ± 0.2
Sugar, % of DM	3.8 ± 0.3	4.6 ± 0.3	4.8 ± 0.3	5.2 ± 0.5	5.0 ± 0.3
Starch, % of DM	21.8 ± 0.7	20.9 ± 0.9	24.7 ± 0.7	24.7 ± 0.9	32.1 ± 1.3
Fat, % of DM	3.4 ± 0.1	2.9 ± 0.1	4.9 ± 0.1	4.8 ± 0.1	4.2 ± 0.2
Ash, % of DM	7.7 ± 0.2	7.7 ± 0.2	8.2 ± 0.2	8.0 ± 0.2	6.8 ± 0.2
Ca, % of DM	1.52 ± 0.06	1.59 ± 0.15	1.14 ± 0.07	1.02 ± 0.13	0.93 ± 0.12
P, % of DM	0.43 ± 0.01	0.40 ± 0.02	0.39 ± 0.03	0.36 ± 0.01	0.44 ± 0.02
Mg, % of DM	0.45 ± 0.01	0.51 ± 0.03	0.43 ± 0.02	0.41 ± 0.01	0.32 ± 0.02
K, % of DM	1.03 ± 0.03	1.14 ± 0.05	1.49 ± 0.12	1.47 ± 0.05	1.47 ± 0.05
S, % of DM	0.48 ± 0.02	0.42 ± 0.02	0.23 ± 0.01	0.22 ± 0.02	0.22 ± 0.02
Na, % of DM	0.12 ± 0.02	0.11 ± 0.02	0.58 ± 0.04	0.53 ± 0.02	0.62 ± 0.11
Cl, % of DM	0.49 ± 0.03	0.58 ± 0.04	0.56 ± 0.02	0.51 ± 0.03	0.34 ± 0.03
DCAD, mEq/100 g	-12.1 ± 2.7	-8.7 ± 2.3	33.6 ± 5.1	32.6 ± 2.8	40.7 ± 6.3
ME Mcal/kg DM[5]	2.09	2.13	2.65	2.66	2.69
MP, g/kg DM[5]	87	115	101	127	113
RDP, % of DM[5]	7.46	9.04	9.06	8.92	10.46
RUP, % of DM[5]	3.71	7.08	5.00	8.25	6.39
Methionine, g/Mcal ME	1.24	1.23	1.18	1.20	1.01
Lysine, g/Mcal ME	3.88	3.84	3.27	3.31	2.88

[1]Prepartum TMRs, fed starting at 28 d before expected calving, were formulated to contain methionine and lysine at 1.24 and 3.84 g/Mcal metabolizable energy (ME), respectively and supply a control (C; 85 g MP/kg DM) or high (H; 113 g MP/kg DM) level of estimated metabolizable protein (MP). Postpartum TMRs, fed from 0 to 21 DIM, were formulated to contain methionine and lysine at 1.15 and 3.16 g/Mcal ME, respectively and supply control (C; 104 g MP/kg DM) or high (H; 131 g MP/kg DM) level of estimated MP.
[2]From 22 to 42 DIM, cows were fed a lactation diet that contained methionine and lysine at 1.01 and 2.88 g/Mcal ME, respectively and supplied 113 g of estimated MP/kg DM.

[3]Chemical composition represents the mean ± SD of 6 4-wk composite samples.

[4]Presented as mean ± SD of 22 weekly DM measurements.

[5]RDP = rumen degradable protein, RUP = rumen undegradable protein. Estimated in AMTS.Cattle.Professional v. 4.17.0.0 (AMTS LLC) using average dry matter intake for each diet.

Table 3. Prepartum dry matter intake, body weight and metabolizable energy and protein balance as well as colostrum production.

Variable	Treatment[1]		*P*-value		
	Control (n = 48)	High (n = 47)	Trt	Wk	Trt x Wk
DMI, kg/d	13.5 ± 0.3	14.0 ± 0.3	0.04	< 0.01	0.43
DMI, % BW	1.77 ± 0.03	1.80 ± 0.03	0.29	< 0.01	0.9
BW, kg	768 ± 3	778 ± 3	< 0.01	< 0.01	0.96
BW change, kg	23.9 ± 3.3	35.1 ± 3.3	< 0.01		
BW change, %	3.21 ± 0.45	4.78 ± 0.45	< 0.01		
BCS	3.39 ± 0.02	3.39 ± 0.02	0.76	0.40	0.35
BCS change, n (%)			0.14		
-0.25	4 (8.3)	2 (4.3)			
0.00	23 (47.9)	33 (70.2)			
0.25	20 (41.7)	12 (25.5)			
0.50	1 (2.1)	0 (0)			
MP supply, g/d[2]	1,180 ± 27	1,606 ± 27	< 0.01	< 0.01	0.06
MP balance, %[2]	101.1 ± 1.6	139.0 ± 1.6	< 0.01	< 0.01	< 0.01
ME balance, %[2]	96.7 ± 1.8	101.8 ± 1.8	< 0.01	< 0.01	0.79
Colostrum yield, kg	15.0 ± 1.0	15.9 ± 1.0	0.62		
Brix percentage, %	26.8 ± 0.5	27.0 ± 0.5	0.82		

[1]Prepartum TMRs, fed starting at 28 d before expected calving, were formulated to contain methionine and lysine at 1.24 and 3.84 g/Mcal metabolizable energy (ME), respectively and supply a control (C; 85 g MP/kg DM) or high (H; 113 g MP/kg DM) level of estimated metabolizable protein (MP).

[2]Estimated in AMTS.Cattle.Professional v. 4.17.0.0 (AMTS LLC) weekly using cow's DMI, BW, calf birth weight, and days carried calf.

Postpartum period

Results from the postpartum period are in Tables 4 and 5. Dry matter intake (% of BW) did not differ during the postpartum period ($P \geq 0.70$). Metabolizable protein supply from 1 to 21 DIM was greater for cows fed the postpartum H diet (CH: 2766 ± 50; HH: 2667 ± 51 g/d) compared to cows fed the C postpartum diet (CC: 2033 ± 51; HC: 2030 ± 52 g/d; $P < 0.01$). Milk yield during 1 to 21 DIM was greater in CH (42.4 ± 0.9 kg/d) compared to HC (38.0 ± 1.0 kg/d; $P < 0.01$) and milk yield in HH (44.7 ± 1.0 kg/d) was greater than CC (39.2 ± 1.0 kg/d; $P < 0.01$) and HC ($P < 0.01$), respectively. Concentrations of milk protein ($P = 0.15$), fat ($P = 0.79$), and total solids ($P = 0.90$) were not affected by treatment, but lactose was greater in CH compared to CC (4.81 ± 0.02 vs. 4.76 ± 0.02 %; $P = 0.04$). Milk urea nitrogen was lower in cows fed a control level of MP postpartum (CC: 6.37 ± 0.36; HC: 6.92 ± 0.36 mg/dL) compared to cows fed the H postpartum diet (CH: 8.83 ± 0.35; HH: 9.33 ± 0.35 mg/dL; $P < 0.01$). Cows fed CC had a greater loss in body weight (7.36 ± 0.65%) compared to cows fed CH (4.06 ± 0.64%; $P < 0.01$). Nevertheless, BCS ($P = 0.54$), the loss of BCS ($P = 0.42$), concentrations of circulating fatty acids ($P = 0.41$), and events of hyperketonemia ($P = 0.13$) did not differ by treatment during the first three weeks of lactation. Metabolizable protein supply was greater in CH at wk 4 compared to CC (*P* = 0.03) and HC (*P* = 0.03), but MP supply did not differ by treatment at wk 5 or 6 (*P* > 0.12). Milk yield remained elevated from 22 to 42 DIM in the cows fed the high postpartum diet (CH: 53.3 ± 1.0; HH: 54.1 ± 1.0 kg/d) compared to the cows fed the control postpartum diet (CC: 49.6 ± 1.0; HC: 49.3 ± 1.0 kg/d). Concentrations of milk components did not differ by treatment (*P* ≥ 0.10). Although the change in body weight did not differ from 22 to 42 DIM, BCS was lower in CC compared to CH (3.04 ± 0.03 vs. 3.17 ± 0.03; *P* = 0.02). From 7 to 40 DIM, backfat thickness (*P* = 0.99) as well as longissimus dorsi muscle diameter (*P* = 0.78) did not differ by treatment. Further, treatment did not affect projected 305-d milk production at last test (M305; CC: 12,076 ± 419; CH: 12,865 ± 410; HC: 11,797 ± 429; HH: 12,886 ± 419 kg; *P* = 0.17).

Conclusions and Implications

Increasing prepartum MP supply from 1,180 to 1,606 g/d did not affect colostrum yield or Brix percent from multiparous cows. Moreover, when feeding a control level of MP postpartum (~2,030 g/d), these data do not support increasing prepartum MP supply >1,200 g/d based on the lack of difference in milk yield during early lactation (CC vs. HC). However, lactation performance was increased in response to feeding a high level of MP (~2,700 g/d) in the fresh diet and these differences in performance persisted beyond the initial feeding period. Notably, cows fed a high level of MP during the pre- and postpartum period (HH) produced the most milk during wk 1 which resulted in a numerically higher milk yield (+2.3 ± 1.0 kg/d; 5.1 ± 2.2 lb/d) during the first three weeks of lactation when compared to cows fed CH. Overall, these data support increasing the MP supply during the postpartum period and under the correct economic situation, producers might benefit from feeding a high level of MP pre- and postpartum.

Table 4. Dry matter intake, BW, milk production, and metabolizable energy and protein balance from 1 to 21 DIM.

	Treatment[1]				*P*-value		
Variable	CC (n = 23)	CH (n = 24)	HC (n = 22)	HH (n = 23)	Trt	Wk	Trt x Wk
DMI, kg/d	20.2 ± 0.4	21.5 ± 0.4	20.5 ± 0.4	20.7 ± 0.4	0.10	< 0.01	0.68
DMI, % BW	2.98 ± 0.06	3.04 ± 0.06	3.00 ± 0.06	2.94 ± 0.06	0.71	< 0.01	0.52
Milk Yield, kg/d	39.2 ± 1.0[bc]	42.4 ± 0.9[ab]	38.0 ± 1.0[c]	44.7 ± 1.0[a]	< 0.01	< 0.01	< 0.01
ECM, kg/d	46.4 ± 1.1[bc]	50.3 ± 1.1[ab]	45.2 ± 1.1[c]	53.6 ± 1.1[a]	< 0.01	< 0.01	< 0.01
Fat, %	4.81 ± 0.11	4.82 ± 0.11	4.84 ± 0.12	4.96 ± 0.11	0.79	< 0.01	0.26
Protein, %	3.20 ± 0.04	3.20 ± 0.04	3.25 ± 0.04	3.11 ± 0.04	0.15	< 0.01	0.16
Lactose, %	4.76 ± 0.02[b]	4.81 ± 0.02[a]	4.81 ± 0.02[ab]	4.79 ± 0.02[ab]	0.04	< 0.01	0.88
Total Solids, %	13.77 ± 0.14	13.84 ± 0.13	13.90 ± 0.14	13.87 ± 0.13	0.90	< 0.01	0.81
MUN, mg/dL	6.37 ± 0.36[b]	8.83 ± 0.35[a]	6.92 ± 0.36[b]	9.33 ± 0.35[a]	< 0.01	0.30	0.38
BW, kg	678 ± 11[bc]	716 ± 11[ab]	676 ± 11[c]	720 ± 11[a]	< 0.01	< 0.01	0.05
BW change, kg	-55 ± 5[b]	-30 ± 5[a]	-42 ± 5[ab]	-46 ± 5[ab]	< 0.01		
BW change, %	-7.36 ± 0.65[b]	-4.06 ± 0.64[a]	-5.63 ± 0.67[ab]	-6.12 ± 0.67[ab]	< 0.01		
BCS	3.27 ± 0.04	3.33 ± 0.03	3.31 ± 0.04	3.33 ± 0.04	0.54	< 0.01	0.18
BCS change, n (%)					0.42		
-0.75	0 (0)	0 (0)	0 (0)	1 (4.4)			
-0.50	6 (26.1)	2 (8.4)	2 (9.1)	3 (13.0)			
-0.25	11 (47.8)	11 (45.8)	13 (59.1)	13 (56.5)			
0.00	6 (26.1)	11 (45.8)	7 (31.8)	5 (21.8)			
0.25	0 (0)	0 (0)	0 (0)	1 (4.3)			
MP supply, g/d[2]	2,033 ± 51[b]	2,766 ± 50[a]	2,030 ± 52[b]	2,667 ± 51[a]	< 0.01	< 0.01	0.20
MP balance, %[2]	70.7 ± 1.3[b]	92.1 ± 1.3[a]	71.9 ± 1.3[b]	87.4 ± 1.3[a]	< 0.01	< 0.01	0.30
ME balance, %[2]	75.3 ± 1.6[ab]	77.4 ± 1.6[a]	76.8 ± 1.6[ab]	71.6 ± 1.6[b]	0.05	< 0.01	0.56

[a-c] Least squares means with different superscripts differ (*P* ≤ 0.05; Tukey's test).

[1]Prepartum TMRs, fed starting at 28 d before expected calving, were formulated to contain methionine and lysine at 1.24 and 3.84 g/Mcal metabolizable energy (ME), respectively and supply a control (C; 85 g MP/kg DM) or high (H; 113 g MP/kg DM) level of estimated metabolizable protein (MP). Postpartum TMRs, fed from 0 to 21 DIM, were formulated to contain methionine and lysine at 1.15 and 3.16 g/Mcal ME, respectively and supply control (C; 104 g MP/kg DM) or high (H; 131 g MP/kg DM) level of estimated MP. The combination of a pre- and postpartum diet resulted in treatments: 1) CC, 2) CH, 3) HC, 4) HH. From 22 to 42 DIM, cows were fed a lactation diet that contained methionine and lysine at 1.01 and 2.88 g/Mcal ME, respectively and supplied 113 g MP/kg.

[2]Estimated in AMTS.Cattle.Professional v. 4.17.0.0 (AMTS LLC) weekly using cow's DMI, BW, DIM, milk yield, and milk composition.

Table 5. Dry matter intake, BW, milk production, and metabolizable energy and protein balance from 22 to 42 DIM.

	Treatment[1]				*P*-value		
Variable	CC (n = 23)	CH (n = 24)	HC (n = 22)	HH (n = 23)	Trt	Wk	Trt x Wk
DMI, kg/d	25.1 ± 0.5	26.8 ± 0.5	25.1 ± 0.5	25.6 ± 0.5	0.06	< 0.01	< 0.01
DMI % BW	3.86 ± 0.08	3.89 ± 0.07	3.82 ± 0.08	3.77 ± 0.08	0.70	< 0.01	< 0.01
Milk Yield, kg/d	49.6 ± 1.0[b]	53.3 ± 1.0[a]	49.3 ± 1.0[b]	54.1 ± 1.0[a]	< 0.01	< 0.01	0.21
ECM, kg/d	52.8 ± 1.1[b]	57.9 ± 1.1[a]	52.8 ± 1.1[b]	57.7 ± 1.1[a]	< 0.01	< 0.01	0.53
Fat, %	4.09 ± 0.09	4.28 ± 0.09	4.10 ± 0.10	4.11 ± 0.09	0.38	< 0.01	0.62
Protein, %	2.83 ± 0.03	2.82 ± 0.03	2.91 ± 0.03	2.79 ± 0.03	0.10	0.02	0.08
Lactose, %	4.85 ± 0.01	4.86 ± 0.01	4.88 ± 0.01	4.85 ± 0.01	0.42	< 0.01	0.69
Total Solids, %	12.69 ± 0.11	12.89 ± 0.11	12.84 ± 0.11	12.70 ± 0.11	0.45	< 0.01	0.41
MUN, mg/dL	8.96 ± 0.31	9.61 ± 0.31	9.46 ± 0.32	9.86 ± 0.32	0.15	< 0.01	0.07
BW, kg	650 ± 10[b]	689 ± 10[a]	657 ± 10[ab]	681 ± 10[ab]	0.02	0.16	0.92
BW change, kg	-0.5 ± 4.3	4.1 ± 4.2	-0.1 ± 4.4	2.2 ± 4.3	0.82		
BW change, %	-0.1 ± 0.7	0.7 ± 0.6	0.1 ± 0.7	0.4 ± 0.7	0.83		
BCS	3.04 ± 0.03[b]	3.17 ± 0.03[a]	3.08 ± 0.04[ab]	3.10 ± 0.03[ab]	0.03	< 0.01	0.74
BCS change					0.98		
-0.50	1 (4.4)	2 (8.3)	1 (4.5)	2 (8.7)			
-0.25	6 (26.1)	7 (29.2)	8 (36.4)	5 (21.7)			
0.00	13 (56.5)	13 (54.2)	10 (45.5)	14 (60.9)			
0.25	3 (13.0)	2 (8.3)	3 (13.6)	2 (8.7)			
MP supply, g/d[2]	2,801 ± 67[b]	3,079 ± 65[a]	2,798 ± 68[b]	2,926 ± 67[ab]	< 0.01	< 0.01	0.01
MP balance, %[2]	94.3 ± 1.5	98.3 ± 1.4	93.3 ± 1.5	94.1 ± 1.5	0.07	< 0.01	< 0.01
ME balance, %[2]	88.5 ± 1.7	88.2 ± 1.7	88.7 ± 1.8	85.0 ± 1.7	0.39	< 0.01	0.60

[a-c] Least squares means with different superscripts differ ($P \leq 0.05$; Tukey's test).

[1]Prepartum TMRs, fed starting at 28 d before expected calving, were formulated to contain methionine and lysine at 1.24 and 3.84 g/Mcal metabolizable energy (ME), respectively and supply a control (C; 85 g MP/kg DM) or high (H; 113 g MP/kg DM) level of estimated metabolizable protein (MP). Postpartum TMRs, fed from 0 to 21 DIM, were formulated to contain methionine and lysine at 1.15 and 3.16 g/Mcal ME, respectively and supply control (C; 104 g MP/kg DM) or high (H; 131 g MP/kg DM) level of estimated MP. The combination of a pre- and postpartum diet resulted in treatments: 1) CC, 2) CH, 3) HC, 4) HH. From 22 to 42 DIM, cows were fed a lactation diet that contained methionine and lysine at 1.01 and 2.88 g/Mcal ME, respectively and supplied 113 g MP/kg.

[2]Estimated in AMTS.Cattle.Professional v. 4.17.0.0 (AMTS LLC) weekly using cow's DMI, BW, DIM, milk yield, and milk composition.

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