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Supplementing sows with the leucine metabolite beta-hydroxy-beta-methylbutyrate and vitamin D_3 improves piglet birth weights that may lead to increased weaning weights

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Previous university studies demonstrated that supplementing sows with calcium beta-hydroxy-beta-methylbutyrate (CaHMB) in late gestation and/or lactation improved piglet weights through weaning. Two studies were conducted at commercial farrowing operations to test if the results would translate to commercial operations. Sows in both trials were randomized to receive either 3 g/day CaHMB plus 500 IU/day vitamin D_3 (HMB/D) or a calcium carbonate containing control top-dressed to the feed from day 104 of gestation through weaning. Sows were randomly assigned to either HMB/D (n = 41 trial 1 and n = 26 trial 2) or control (n = 46 trial 1 and n = 26 trial 2). Data were analyzed using a general linear model with main effects of group, treatment, and group by treatment interaction. Treatment with HMB/D had no effect on sow weights, lactational weight loss, and stillborn or mummified piglets. In trial 1, the control group had an increased number of live born piglets, which at 24 h tended to be greater, and no difference in liveborn or 24-h piglet numbers was seen in trial 2. In trial 1, HMB/D increased piglet live birth ($P < 0.03$) and 24-h weights (1,490 ± 30.1 vs. 1,390 ± 28.8 g in HMB/D and control piglets, respectively, $P < 0.02$). Farm practices were to equalize piglet numbers across sows by cross-fostering. After cross-fostering, the 24-h average piglet weights were not different, and further advantages to supplementation were not observed ($P = 0.21$). In trial 2, birth and 24-h weights of the piglets from HMB/D-supplemented sows were increased ($P < 0.0001$). Piglets from sows supplemented with HMB/D were 9.7% heavier at birth and 9.2% heavier at 24 h (1,549 ± 22.0 and 1,419 ± 21.2 in HMB/D and control, respectively). A difference was observed in weaning age ($P < 0.0001$), and weaning weights were adjusted to 21-day weights (5,426 ± 103.5 and 5,205 ± 99.5 for HMB/D and control piglets, respectively, $P = 0.12$). Analysis by group showed that HMB/D tended to increase weaning weights in younger sows (second and third parity), 5,432 ± 150.7 and 5,074 ± 142.7 in HMB/D and control piglets, respectively ($P < 0.09$). In conclusion, these results agree with previous university studies demonstrating that CaHMB supplementation increased early piglet weights with a tendency to improve weaning weights.

KEYWORDS

Sow nutrition, CaHMB, piglet weight, weaning weight, sow lactation nutrition, sow late gestation nutrition

1 Introduction

Beta-hydroxy-beta-methylbutyrate (HMB) is an endogenous metabolite of the essential branched chain amino acid leucine (Van Koevering and Nissen, 1992). Decades of research in animals and humans have demonstrated that supplementing endogenously produced HMB can have positive effects on animal growth and health (Szczesniak et al., 2015) and, in humans, preserves or increases muscle mass and strength in exercise and age-related muscle loss (Wilson et al., 2013; Gepner et al., 2019; Oktaviana et al., 2019). Use of HMB as a potential feed ingredient to promote animal protein metabolism, growth, and general health has been identified for over three decades (Nissen et al., 1990a; Nonnecke et al., 1991). A review of HMB studies discussed the benefits of supplementing HMB across many species (Szczesniak et al., 2015).

Swine genetics and sow selection have greatly increased the number of piglets born per litter. Variation within litter birth weights leads to greater pre- and post-parturition mortality (Yuan et al., 2015). More uniform birth weight has the potential to improve survivability and also reduce management costs in trying to optimize production efficiency and piglet survival (Yuan et al., 2015).

Nissen et al. (1994a) first identified benefits in supplementing HMB to sows from 2 days prior to farrowing through the lactation and to weaning. Since then, several studies have been conducted, but thus far, published studies have been relatively small and conducted in university research settings. However, many university studies have shown the benefit of supplementing calcium beta-hydroxy-beta-methylbutyrate (CaHMB) to sows and have reported the following benefits to the piglet: reduction in low-birth weight piglets and tendency for greater-birth weight piglets (Wan et al., 2016b), increased piglet birth weight (Tatara et al., 2012), and increased weaning weights (Nissen et al., 1994a; Krakowski et al., 2002). Recently, a relatively large university study was reported by Davis et al. (2021) where a total of 279 multiparous sows were studied in four groups. Over all of the groups, they found that feeding sows 15 mg/kg CaHMB per day (approximately 3.4 g) starting at day 100 of gestation until farrowing resulted in overall increased litter birth weight and tendencies for increased 24-h and weaning weights of the piglets.

Studies in humans have shown that vitamin D_3 combined with HMB has a synergistic effect on muscle function and muscle health (Fuller et al., 2011; Rathmacher et al., 2020). While gestation and lactation diets generally contain adequate vitamin D_3 or 25(OH)-vitamin D_3, there is still debate over what constitutes an optimal level for the desired effects of muscle development and piglet birth and weaning weights (Flohr et al., 2016b; Flohr et al., 2016a; Thayer et al., 2019; Zhang and Piao, 2021). To ensure additional vitamin D_3 with the HMB, 500 additional units of vitamin D_3 per day were added to the HMB supplement.

Two trials were conducted, one in Brazil and one in the United States to investigate the effects of supplementing CaHMB and vitamin D_3 to sows in late gestation and lactation on piglet birth, 24-h, and weaning weights in the commercial production setting. The objective of these studies was to determine if previously observed benefits of supplementing HMB to sows in the university setting would be observed in a commercial sow production setting. The hypothesis that supplementation with HMB would increase birth and 24-h piglet weights and would result in an improved piglet weaning weight was evaluated.

2 Materials and methods

2.1 Animals

Trial 1 was conducted at a commercial farrow to finish operation in Brazil (Patos de Minas, Minas Gerais, Brazil), and a total of 87 sows from DanBred Pig Genetics (Minas, Minas Gerais, Brazil) with sows having Landrace and Large White bred to Duroc boars were used for the study. DanBred Pig Genetics are bred for increased piglet numbers, and sows were from third to fifth parity. The number of sows by parity is shown in

Supplementary Table S1. Sows were studied in two farrowing groups, with 58 sows in the first group and 29 in the second. Data from all 87 sows were included in the analysis.

Trial 2 was conducted at a commercial swine breeding farm in the United States (Intelipig, Bloomkest, MN, USA) and on a total of 52 sows from DNA Genetics (Columbus, NE, USA) bred to PIC boars (Hendersonville, TN, USA). Sows were studied in two farrowing groups of 26 sows each. Group 1 consisted of fourth and fifth parity sows, while in group 2, sows were second and third parity. The number of sows by parity for trial 2 is also shown in Supplementary Table S1. In group 1, one Control sow was not pregnant, and one HMB/D sow farrowed late, and her piglets were weaned at day 14. In group 2, one Control sow failed to farrow, and one HMB/D died pre-farrowing. Additionally, in group 2, one HMB/D sow was weaned early and the sow died shortly after. Data from these sows were excluded from the analysis so a total of 24 control sows and 23 HMB/D sows were included in the analyses.

The animal protocol was reviewed and approved by the company internal ethics committee, Metabolic Technologies, LLC. All procedures used in the studies were standards of practice and care for production animals, and a licensed veterinarian performed the blood and milk sampling. Additionally, all animal procedures followed the Guide for the Care and Use of Agricultural Animals in Research and Teaching (Tucker et al., 2020).

2.2. Treatments and experimental design

In both trials, sows were randomized to receive either a control [maltodextrin plus $CaCO_3$ (Fisher Scientific, Pittsburgh, PA, USA)] or CaHMB (Metabolic Technologies, Missoula, MT, USA) plus vitamin D_3 (Lycored, Branchburg, NJ, USA) (HMB/D, maltodextrin plus 3.0 g CaHMB and 500 IU vitamin D_3 per day per sow) treatments. The added vitamin D was about 30% more than the National Research Council (NRC)-recommended daily vitamin D intake of 1,680 IU per day and was added due to the recent awareness that vitamin D and HMB can improve muscle function (NRC, 2012). The experimental treatments were assayed for CaHMB and vitamin D_3 in both trials by High Pressure Liquid Chromatography (HPLC) (Heartland Assays, Ames, IA, USA). The analyses showed 19.6% and 23.9% CaHMB and 49.5 and 57.1 IU vitamin D_3 per gram for trials 1 and 2, respectively. The control was assayed for calcium in trial 2 (3.3%; Iowa State University, Ames, IA, USA) that was the same as the calcium provided by the CaHMB (3.3%). Based upon the scoop size used in the trials (approximately 30 ml), at minimum, the active ingredients were provided as described above. Treatments were top dressed to the feed in the morning beginning 10 days pre-farrowing, approximately Day 104 of gestation, and continued until weaning. Calculated analyses of the diets for trials 1 and 2 are shown in Supplementary Tables S2, S3, respectively.

2.3. Measurements and sampling

Weights of the sows were recorded at day 103 from breeding, at the time the sows were moved to the farrowing rooms, and again at weaning. Individual piglet weights were taken at birth, 24 h after birth, and at weaning.

For both trials, the sows farrowed without needing induction. Farrowing progress was monitored, and in trial 1, if the birthing interval was longer than 20 min, the sow's belly was massaged to help the labor process. In trial 2, sows were again monitored during the birthing process, and if no birthing for 45–60 min, oxytocin was administered as was normal practice at that farm. In trial 1, farm practices included equalizing piglet numbers across sows 24 h after the sow finished farrowing. Piglets were equalized as determined by the viable number of teats on each sow and were equalized within treatments. However, excess piglets were equalized on sows not in the study, and during this process, the early weight advantage was nullified. Because of this, no differences were observed at weaning (see *Results*). In trial 2, piglets were tagged at birth, and no cross-fostering was performed.

In trial 2 conducted at the Bloomkest, MN, USA, site, it was noticed that there appeared to be a difference in performance of the piglets between group 1 and group 2. As group 1 was studied in late winter and group 2 in early summer, the ambient outdoor air temperatures were determined at weather.gov and are footnoted in Table 6 below (USA).

In trial 2, sow body condition score (BCS) was evaluated using a scale of 1 (thin) to 5 (fat) by the use of calipers (Knauer and Baitinger, 2015). In trial 2, sow colostrum was collected as soon as practical after farrowing (24–36 h). Briefly, colostrum from the distal part of the teat was collected by closing off the proximal teat cistern with one's fingers and pressurizing the teat cistern to extract colostrum from the distal teat sphincter. Colostrum samples were stored at -20°C until shipment to the laboratory for analysis. A blood sample was also taken from the caudal vena cava and allowed to clot. The sample was centrifuged in a serum separator centrifuge, and the serum was decanted and frozen at -20°C until shipment to the laboratory for analysis. Colostrum and serum HMB were then analyzed (Heartland Assays, Ames, IA, USA) (Nissen et al., 1990b).

2.4 Statistical analysis

Data were analyzed using Proc GLM in SAS (SAS for Windows 9.4, SAS Institute, Inc., Cary, NC, USA). A twoway analysis of variance (ANOVA) model was used with the main effects of group, treatment, and group by treatment interaction. Means reported are least squares means with the standard error of the mean. In trial 2, weaning weights were adjusted to 21 days of age and analyzed. Weights were corrected using the average daily gain over the measurement period to adjust to a 21-day weaning weight. For discrete variables, parity, BCS at farrowing, and BCS at weaning, Proc Freq in SAS was used to perform a chi-square test of the data. A P-value ≤0.05 was determined to indicate significance, while trends were identified as 0.05 ≤ P ≤ 0.10.

3 Results

3.1. Trial 1

3.1.1 Trial 1 sow characteristics

As shown in Table 1, sows averaged fourth parity, and the range in each group was from third to fifth parity. No group, treatment, or group by treatment effects were measured on sow characteristics shown in Table 1 that included weights at 103 days of gestation, before farrowing, and at weaning and for observed weight changes pre-farrowing and during lactation.

3.1.2 Litter characteristics

There were no group or group by treatment effects on litter and piglet characteristics measured (Table 2). Piglet numbers tended to be greater in the control sow litters at birth and at 24 h. No treatment effects were seen on stillborn or mummified piglets at birth. Birth weights and 24-h piglet weights were increased with HMB/D supplementation (P < 0.03 and P < 0.02, respectively). At 24 h, farm practice was to normalize the number of piglets per sow, and the numbers after normalization are also shown in Table 2, with no difference between treatments for number of piglets per sow (P = 0.78) after equalization. Sow piglet distribution by number is shown in Supplementary Table S4. As seen in Table 2, litter sizes decreased by 2–3 piglets per litter, meaning some piglets were transferred to non-study sows. After equalization, there was no weight advantage of the piglets supplemented with HMB/D (P = 0.21). Weaning weights are also shown in Table 2, and there was no difference in weaning weights between the treatment groups.

3.2 Trial 2

3.2.1 Sow characteristics

Sow characteristics are shown in Table 3. Overall, the sows studied averaged 3.83 ± 0.087 and 3.74 ± 0.089 for control and HMB/D sows, respectively. There was a group effect (P < 0.0001) but no group by treatment interaction effect (P = 0.93). Sows in group 1 averaged parity 4.79 ± 0.09, and sows in group 2 averaged parity 2.78 ± 0.09 (P < 0.001). There was a group by treatment interaction for sow weaning weight. No effect of treatment on weaning weight was observed in group 1, the older sows, whereas in group 2, the younger sows treated with HMB/D were heavier at weaning than control-treated sows (P < 0.03).

No treatment effects were observed on BCSs pre-farrowing or at weaning. However, group effects on BCSs were observed pre-farrowing (4.58 ± 0.13 and 3.91 ± 0.13 in groups 1 and 2, respectively, P < 0.0008) and at weaning (4.00 ± 0.16 and 2.86 ± 0.17 in groups 1 and 2, respectively, P < 0.0001). BCSs were greater in the older sows in group 1 (P < 0.008 and P < 0.0001 for pre-farrowing and weaning, respectively), but no group by treatment interactions were observed (P = 0.67 and P = 0.34 for pre-farrowing and weaning, respectively).

3.2.2 Litter characteristics and performance

The number of piglets born alive, stillborn, mummified, weaned, and that died per litter is shown in Table 4. There were no treatment or group by treatment effects on any of these

TABLE 1 The effect of supplementing with calcium beta-hydroxy-beta-methylbutyrate and vitamin D (HMB/D) during late gestation and lactation on sow weights and condition in trial 1[a].

	Control	HMB/D	P-Value		
			Group	Treatment	Group by treatment
Parity[b]	4.16 ± 0.14	4.07 ± 0.14	0.93	0.82	-
103-Day Weight, kg	269.0 ± 4.6	269.4 ± 4.8	0.32	0.96	0.83
Farrowing Weight, kg	276.3 ± 4.3	277.6 ± 4.5	0.33	0.83	0.88
Weaning Weight, kg	254.0 ± 4.6	254.1 ± 4.9	0.47	0.99	0.59
Pre-Farrowing BW Change, kg	7.32 ± 0.76	8.28 ± 0.80	0.60	0.39	0.66
Lactational BW Change, kg	-22.3 ± 2.72	-23.6 ± 2.84	0.77	0.75	0.49

[a]Treatments were top-dressed to the rations in the morning. The control contained $CaCO_3$, and HMB/D contained 3 g CaHMB and 500 IU vitamin D_3. Results are means ± SEM. n = 46 control sows and n = 41 HMB/D-treated sows.
[b]Parity analysis was performed using chi-square test of significance for group and treatment.

TABLE 2 The effect of supplementing calcium beta-hydroxy-beta-methylbutyrate and vitamin D (HMB/D) during late gestation and lactation on number of piglets per sow in trial 1.[a]

	Control	HMB/D	P-Value		
			Group	Treatment	Group by Treatment
Born Alive, n	16.87 ± 0.38	15.83 ± 0.39	0.36	0.06	0.97
Stillborn, n	2.60 ± 0.34	2.16 ± 0.36	0.87	0.38	0.51
Mummified, n	0.49 ± 0.14	0.57 ± 0.15	0.42	0.71	0.58
24-Hour, n	16.59 ± 0.37	15.63 ± 0.39	0.36	0.08	0.93
After equalization, n	13.72 ± 0.13	13.67 ± 0.14	0.09	0.78	0.81
Birth Weight, g	1320 ± 26.7	1400 ± 27.9	0.83	0.03	0.38
24-Hour Weight, g	1390 ± 28.8	1490 ± 30.1	0.39	0.02	0.42
24-Hour Weight after equalization, g	1460 ± 40.7	1530 ± 42.6	0.39	0.21	0.94
Weaning Weight, g	6250 ± 115.2	6200 ± 120.6	0.13	0.78	0.87

[a]Treatments were top-dressed to the rations in the morning. Control contained $CaCO_3$, and HMB/D contained 3 g CaHMB and 500 IU vitamin D_3. Results are means ± SEM. n = 46 control sows and n = 41 HMB/D-treated sows.

measurements. Sow piglet distribution by number is shown in Supplementary Table S5. There was, however, a group effect on the number of stillborn piglets (P < 0.04). The numbers of stillborn piglets per sow in group 1 were 3.58 ± 0.64 and 2.83 ± 0.64 and in group 2 were 2.08 ± 0.64 and 1.64 ± 0.67 for Control and HMB/D treatments, respectively. Group 1 had a greater number of stillborn piglets than that of group 2, but there was no group by treatment interaction (P = 0.82).

3.2.3. Piglet performance

As shown in Table 5, birth and 24-h piglet weights were increased by 9.7% and 9.2% (P < 0.0001) in piglets from HMB/D-supplemented sows, respectively. A group effect was also observed for birth and 24-h weights likely due to the higher parity sows in group 1 vs. group 2 (P < 0.0002 and P < 0.001, respectively). However, no group by treatment interaction was observed for either birth or 24-h weights (P = 0.21 and P =0.28, respectively). Weaning weights were not different between the treatments (P = 0.36). Age to weaning was determined by using the actual farrowing and weaning dates. Sows were weaned on the same day; however, farrowing occurred on multiple days following transfer to the farrowing room. Group and treatment differences in age at weaning were observed (P < 0.0001). Therefore, 21-day adjusted weaning weights were calculated and are also shown in Table 5. Adjusted weaning weights were not different between groups (P = 0.12); however, there was a numerical 4.2% increase. As noted in the footnote to Table 6, group 1, studied in late winter, had a lower average ambient outdoor air temperature (6.1°C) than that of group 2, studied in early summer, which had a higher average ambient outdoor air temperature (21.0°C).

Because there were significant group effects observed in early piglet weights, birth, 24-h, weaning, and adjusted weaning weights are shown by group in Table 6. Treatment with HMB/D had the greatest effect on birth and 24-h piglet weights in the older fourth and fifth parity sows, about double what was

TABLE 3 The effect of supplementing calcium beta-hydroxy-beta-methylbutyrate and vitamin D (HMB/D) during late gestation and lactation on sow weights and condition.[a]

	Control	HMB/D	P-Value		
			Group	Treatment	Group by treatment
Parity[b]	3.83 ± 0.087	3.74 ± 0.089	0.0001	0.88	-
103-Day Weight, kg	275 ± 3.92	281 ± 4.00	0.003	0.27	0.17
Farrowing Weight, kg	281 ± 4.00	287 ± 4.09	0.004	0.29	0.93
Weaning Weight, kg	240 ± 4.04	247 ± 4.13	0.02	0.25	0.04
Pre-Farrowing BW Change, kg	6.23 ± 0.45	6.14 ± 0.46	0.76	0.89	0.005
Lactational BW Change, kg	-40.8 ± 3.72	-40.2 ± 3.80	0.50	0.91	0.64
Farrow Body Condition score	4.29 ± 0.13	4.20 ± 0.13	0.0008	0.64	0.67
Wean Body Condition Score	3.54 ± 0.16	3.32 ± 0.17	0.0001	0.34	0.34

[a]Treatments were top-dressed to the rations in the morning. Control contained $CaCO_3$, and HMB/D contained 3 g CaHMB and 500 IU vitamin D_3. Results are means ± SEM. n = 24 control sows and n = 23 HMB/D-treated sows.
[b]Parity analysis was performed using chi-square test of significance for group and treatment.

TABLE 4 The effect of supplementing calcium beta-hydroxy-beta-methylbutyrate and vitamin D (HMB/D) during late gestation and lactation on number of piglets per sow.[a]

	Control	HMB/D	P-Value		
			Group	Treatment	Group by treatment
Born Alive, n	15.00 ± 0.58	14.68 ± 0.60	0.48	0.70	0.48
Stillborn, n	2.83 ± 0.46	2.23 ± 0.47	0.04	0.36	0.82
Mummified, n	0.50 ± 0.21	0.57 ± 0.22	0.29	0.81	0.97
24-Hour, n	14.83 ± 0.60	14.50 ± 0.61	0.78	0.70	0.50
Weaned, n	12.25 ± 0.57	11.83 ± 0.58	1.00	0.61	0.84
Died, n	2.75 ± 0.37	2.85 ± 0.38	0.27	0.85	0.42

[a]Treatments were top-dressed to the rations in the morning. Control contained $CaCO_3$, and HMB/D contained 3 g CaHMB and 500 IU vitamin D_3. Results are means ± SEM. n = 24 control sows and n = 23 HMB/D-treated sows.

TABLE 5 The effect of supplementing calcium beta-hydroxy-beta-methylbutyrate and vitamin D (HMB/D) during late gestation and lactation on piglet weights.[a]

	Control[b]	HMB/D[c]	P-Value		
			Group	Treatment	Group by treatment
Birth Weight, g	1333 ± 19.1	1462 ± 19.8	0.0002	0.0001	0.21
24-Hour Weight, g	1419 ± 21.2	1549 ± 22.0	0.0001	0.0001	0.28
Weaning Weight, g	5117 ± 97.3	5246 ± 101.2	0.49	0.36	0.58
Weaning Age, d	20.50 ± 0.07	20.05 ± 0.07	0.0001	0.0001	0.0001
21-Day Adjusted Weaning Wt., g	5205 ± 99.5	5426 ± 103.5	0.38	0.12	0.34

[a]Treatments were top-dressed to the rations in the morning. Control contained $CaCO_3$, and HMB/D contained 3 g CaHMB and 500 IU vitamin D_3. Results are means ± SEM.
[b]n = 360 for birth, n = 356 for 24-h weights, and n = 294 for weaning, weaning age, and 21-day adjusted weaning weights.
[c]n = 337 for birth, n = 333 for 24-h weights, and n = 272 for weaning, weaning age, and 21-day adjusted weaning weights.

TABLE 6 The effect of supplementing calcium beta-hydroxy-beta-methylbutyrate and vitamin D (HMB/D) during late gestation and lactation on piglet weights by group.[a]

	Group 1[b]		P-Value treatment	Group 2[c]		P-Value treatment
	Control	HMB/D		Control	HMB/D	
Birth Weight, g	1264 ± 24.5	1428 ± 25.2	0.0001	1402 ± 29.5	1496 ± 30.5	0.026
24-Hour Weight, g	1332 ± 26.9	1496 ± 27.8	0.0001	1505 ± 33.0	1602 ± 34.2	0.042
Weaning Weight, g	5108 ± 135.6	5158 ± 138.9	0.80	5126 ± 139.7	5333 ± 147.5	0.31
Weaning Age, d	19.65 ± 0.11	19.60 ± 0.12	0.78	21.35 ± 0.08	20.50 ± 0.08	0.0001
21-Day Adjusted Weaning Weight, g	5337 ± 138.8	5421 ± 142.2	0.67	5074 ± 142.7	5432 ± 150.7	0.086

[a]Treatments were top-dressed to the rations in the morning. Control contained $CaCO_3$, and HMB/D contained 3 g CaHMB and 500 IU vitamin D_3. Results are means ± SEM. Average outside ambient air temperature during the group 1 study was 6.1°C and during group 2 was 21.0°C.
[b]Group 1 Controls n = 180 for birth and 24-h weights and n = 148 for weaning, weaning age, and 21-day adjusted weaning weights. HMB/D n = 169 for birth and 24-h weights and n = 141 for weaning, weaning age, and 21-day adjusted weaning weights.
[c]Group 2 Controls n = 180 for birth, n = 176 for 24-h weights, and n = 146 for weaning, weaning age, and 21-day adjusted weaning weights. HMB/D n = 168 for birth, n = 164 for 24-h weights, and n = 131 for weaning, weaning age, and 21-day adjusted weaning weights.

observed in the second and third parity sows. When 21-day adjusted weights were analyzed, group 1 weights were not different; however, in group 2, adjusted weaning weights tended to be 7% greater ($P = 0.086$) with HMB/D supplementation.

3.2.4. Beta-hydroxy-beta-methylbutyrate measurement in serum and colostrum

Blood and colostrum samples were collected after farrowing (Table 7). The HMB levels in serum were increased with HMB supplementation ($P < 0.0007$); however, there was a group effect

TABLE 7 The effect of supplementing calcium beta-hydroxy-beta-methylbutyrate and vitamin D (HMB/D) during late gestation and lactation on sow serum and colostrum HMB levels.[a]

	Control	HMB/D	P-Value		
			Group	Treatment	Group by treatment
Serum HMB, nmol/ml	1.60 ± 1.75	11.15 ± 1.91	0.0005	0.0007	0.0005
Colostrum HMB, nmol/ml	2.48 ± 1.48	6.31 ± 1.48	0.20	0.07	0.54

[a]Treatments were top-dressed to the rations in the morning. Control contained $CaCO_3$, and HMB/D contained 3 g CaHMB and 500 IU vitamin D_3. Results are means ± SEM. For serum, n = 22 and n = 19 for control and HMB/D sows, respectively. For milk, n = 23 each for control and HMB/D sows.

($P < 0.0005$). Serum levels were increased in group 2 and not in group 1 likely due to the time of sampling in relation to time of supplementation. Colostrum HMB levels were increased with HMB/D supplementation ($P < 0.07$).

4 Discussion

These studies support the findings of earlier university-conducted trials that feeding HMB in late gestation and lactation improves early piglet weights. The present studies consisted of two trials at commercial farrowing operations, one in Brazil and the other in the United States. The genetics utilized by both farms were developed for increased offspring, which can lead to lower birthweight piglets. Improved early piglet weight should result in improved growth and weaning weight and possibly limit mortality of lower-weight piglets (Yuan et al., 2015). In both trials, supplementation with HMB/D resulted in increased early weight of the piglets, which in trial 2 tended to increase weaning weights in one group. In trial 1, the weight advantage was nullified after 24 h during the cross-fostering process used at the farm, and as the experimental unit was litter, the individual piglet weights were not able to be monitored. Thus, the early advantage of piglet weight was lost during piglet equalization, and no advantage was observed at weaning.

Cross-fostering is a common practice when the number of piglets exceeds the number of teats available (Vande Pol et al., 2021). Several methods can be used; however, allotting the piglets based upon teat number is likely common as was used in trial 1. While piglets were cross-fostered within treatments, a number of piglets were cross-fostered to sows not in the study. This brings up the question as to where the most benefit of HMB/D supplementation occurred, during later gestation, lactation, or during both. As discussed further below, studies have varied in the feeding time during gestation and whether HMB is fed during lactation as well and should likely be investigated in further commercial production settings.

During our trial 2 study, early birth and 24-h weight increases translated to a trend for increased weaning weight in the younger group of sows. The sows in this group were the younger sows, second and third parity, whereas in the other group, sows were fourth and fifth parity. Younger sows and gilts have more stress at farrowing time than multiparous sows (Blavi et al., 2021). Additionally, heat stress during intensive livestock operations has a dramatic effect on feed intakes, animal growth, and average daily gain of piglets (Renaudeau et al., 2012). During trial 2, there was a significant outside temperature difference between groups 1 and 2. During farrowing and lactation, the outside temperatures ranged from 0.2°C to 11.9°C with an average of 6.1°C for group 1 and a range from 13.9°C to 28.0°C with an average of 21.0°C for group 2 (United States). Temperature change in the farrowing room as little as 5°C, from 20°C to 25°C, decreases sow feed intake and 21-day piglet weights (Muns et al., 2016). Therefore, it is likely that group 2 sows had additional heat stress, and supplementing with HMB may have helped maintain piglet growth.

Similar to Nissen et al. (1994a) where sows studied were on average third parity sows, the current study also showed additional benefit in weaning weight of the piglets in the younger sows. Regardless of sow age, HMB/D supplementation increased birth and 24-h weights in older and younger sows when fed for 10 days pre-farrowing. Nissen et al. (1994a) failed to show an increase in birth weight when HMB was only fed for approximately 2 days pre-farrowing, and this study as well as others would indicate that HMB needs to be fed for at least 10 days pre-farrowing to observe an effect on early piglet weight (Tatara et al., 2007; Tatara et al., 2012; Wan et al., 2016b; Davis et al., 2021).

Hu et al. (2020) showed that 4–6 h after feeding HMB, net portal flux had returned to near baseline. This may mean that if piglets are to receive optimal benefit either late gestation or during lactation that the dose needs to be fed at least twice daily, which if mixed in the complete feed would be compatible with most commercial practices. In the study by Nissen et al., milk HMB levels were increased on day 10 of lactation and, in more recent studies, HMB in colostrum and milk has been shown to increase similarly to the study presented here (Wan et al., 2017; Hu et al., 2020). Additionally, Hu et al. (2020) found that 26% of the HMB dosage ended up in the colostrum. Wan et al. (2017) found that this lactational intake of HMB by the piglets resulted in improved growth characteristics that were carried through to slaughter, weight at 180 days, average daily gain, and a trend for improved feed efficiency.

A wide range of daily dosages and lengths of supplementation periods have been studied, dosages from 11 to 50 mg/kg body weight (BW) and feeding from as early as day 35 of gestation through parturition to supplementation during lactation only (Nissen et al., 1994b; Krakowski et al., 2002; Tatara et al., 2007; Wan et al., 2016a; Davis et al., 2021). Based upon this study and those of others, a dosage between 11 and 15 mg/kg BW would appear sufficient daily dosage to improve piglet performance, although this should likely be spread throughout the day. In particular, the length of supplementation should be optimized for the desired outcome. Davis et al. (2021) fed HMB 15 days prior to farrowing, and in the present studies, HMB/D was fed from 10 days pre-farrowing through lactation. This length of feeding would appear adequate to improve early piglet weights.

In the present trials, additional vitamin D_3 was top-dressed with HMB. While HMB had been studied over a range of dosages, the combination of HMB with vitamin D_3 has not. The present study focused on piglet performance, and we did not measure serum 25-OH-vitamin D levels in the sows or piglets. Future studies should look at the overall vitamin D status in relation to vitamin D_3 supplementation.

While no treatment effects were seen on the number of stillborn piglets, sows supplemented with HMB/D had numerically lower numbers of stillborn piglets in both trials. Future studies should be designed and powered to possibly detect if this difference is due to the HMB/D treatment. Also, additional studies should be conducted with HMB added directly to the creep feed and after weaning during transfer of the piglets to the nursery. Weaning and early nursery stresses are not unlike stresses shown in other species such as for young chicks (Nissen et al., 1994c; Qiao et al., 2013) or in calves stressed through shipping to feedlots (Van Koevering et al., 1993).

Some limitations of the current study include the farm practices such as cross-fostering that made it impossible to track piglet weights to weaning in trial 1. Additionally, the supplementation was supplied *via* a top-dress to the morning ration. Future studies should use the full feed incorporation method that Davis et al. (2021) used in experiments 3 and 4 reported by his group.

5 Conclusion

In conclusion, these trials are the first studies with HMB in commercial farrowing settings. These studies demonstrate that HMB supplementation to sows improves piglet birth weights and may improve weaning weights in commercial farrowing operations.

Data availability statement

The datasets presented in this article are not readily available because the raw data supporting the conclusions of this article will be made available by the authors, without undue reservation. Requests to access the datasets should be directed to Fuller@mti-hmb.com.

Ethics statement

Ethical review and approval was not required for the animal study because all animal procedures followed the Guide for the Care and Use of Agricultural Animals in Research and Teaching (4th edition, 2020).

Author contributions

Study Conceptualization, JF and JR. Methodology, JF and JR. Study Conduct and Oversight, MM, FC, and RC. Lab Analysis, JR. Data Analysis, JF. Original Draft Preparation, JF. Review and Editing of the Manuscript, JF, MM, FC, RC, and JR. All authors contributed to the article and approved the submitted version.

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Conflict of interest

JF is employed by Metabolic Technologies LLC, Missoula MT which markets CaHMB. JR is currently employed by MTI Biotech Inc., Ames IA and has been a principal investigator for clinical and animal studies with HMB and had previously worked for Metabolic Technologies, Inc. which had marketed HMB in North America.

The remaining authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

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Supplementary material

The Supplementary Material for this article can be found online at: https://www.frontiersin.org/articles/10.3389/fanim.2022.953854/full#supplementary-material

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