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RESEARCH ARTICLE

Sodium propionate supplementation improves the negative energy balance in postpartum dairy cattle through regulation of glycolipid metabolism

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Highlights

- The milk proportions of 4:0, 8:0, 10:0, 13:0, 16:0, total odd-chain fatty acid (FA), and *de novo* FA increased in cows fed sodium propionate (NaPr).
- Supplementing the diet with NaPr significantly increased the concentrations of triglycerides (TG), glucose, and insulin in the plasma of postpartum dairy cattle.
- Supplementing the diet with NaPr alleviates negative energy balance (NEB) in postpartum dairy cattle by increasing plasma glucose levels and promoting lipid deposition in adipose tissue.

Abstract

The study goal was to determine the effects of sodium propionate (NaPr) during the postpartum period on lactation performance, milk fatty acid (FA) profile, blood metabolites, and fat mobilization. This study selected 24 cows with the same parity (3), similar due date, and physical condition in the postpartum period and randomly allocated them into two groups. The constituents of the two treatments were (1) a normal diet for the Control group and (2) a normal diet containing 246 g d^{-1} of NaPr for the NaPr group. This study demonstrated that supplementation of NaPr to dairy cows in the postpartum period had no significant impact on dry matter intake (DMI) and milk composition. The milk proportions of 4:0, 8:0, 10:0, 13:0, 16:0, *cis*-10 15:1, *cis*-13, *cis*-16 22:2, total odd-chain FA, and *de novo* FA increased, and those of all remaining individual saturated fatty acids (SFA) and preformed FA decreased in cows fed NaPr *vs.* the Control diet. Supplementing with NaPr significantly increased the concentrations of triglycerides (TG), glucose, and insulin in the plasma of cows. This indicates that NaPr supplementation in the postpartum period provides energy for cows, which is beneficial for the body's glucose and lipid balance. The TG content, Perilipin-1 (PLIN1) protein expression, and orange-red lipid droplet deposition in adipose tissue were increased in cows fed NaPr *vs.* the Control diet. The mRNA expression of *ACCα* and *ACSS2* in adipose tissue was up-regulated, and the expression of *CPT1A* and *CPT2* genes was down-regulated. This indicates that dietary NaPr supplementation promotes the generation of acetyl CoA, thereby up-regulating the FA synthesis pathway and reducing FA oxidation. In conclusion, dietary supplementation with NaPr promotes energy deposition, improves milk quality and fat accumulation, and alleviates negative energy balance (NEB) in postpartum dairy cattle.

Keywords: sodium propionate, negative energy balance, fat mobilization, fatty acid profile, postpartum

1. Introduction

During the perinatal period, cows may experience a series of physiological disorders after delivery, such as decreased dry matter intake (DMI), hormonal imbalances, and decreased reproductive performance (Silva *et al.* 2022). Previous studies have shown that cows' feed intake sharply

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decreases postpartum, leading to insufficient energy intake to meet lactation needs and resulting in a negative energy balance (NEB) in the body (Gross *et al.* 2011). Meeting the energy needs of lactating cows and mobilizing their own fat to generate non-esterified fatty acids for energy supply also leads to metabolic diseases such as ketosis and fatty liver in cows (Zhang *et al.* 2022b). Ketosis and fatty liver disease affect dairy cows' milk production and reproductive performance, causing severe economic losses. Therefore, reducing fat mobilization and alleviating the negative energy balance in postpartum cows is particularly important.

Intravenous glucose injection is often used to treat excessive fat mobilization in postpartum cows (Herdt and Emery 1992). Large amounts of glucose infusion can increase blood glucose and insulin concentrations in a short period of time. The increase in glucose and insulin can limit fat breakdown in adipose tissue and reduce free fatty acid levels, but the therapeutic effect is transient (Schoenberg *et al.* 2012). As a precursor of glucose, the advantage of propionate feeding to dairy cows is that it can improve milk production and feed intake (Zhang *et al.* 2022b). Propionate plays a vital role as the primary precursor for gluconeogenesis in the liver of lactating cows, accounting for approximately 45% to 60% of overall glucose production (Xue *et al.* 2018). Previous studies have shown that propionic acid can directly regulate glucose and lipid metabolism in bovine liver cells by up-regulating the mRNA expression of key enzymes involved in gluconeogenesis, such as cytoplasmic phosphoenolpyruvate carboxykinase, mitochondrial phosphoenolpyruvate carboxykinase, and pyruvate carboxylase (Zhang *et al.* 2016). In the human white adipocyte model, propionate has been shown to reduce plasma free fatty acids by activating fatty acid oxidation and inhibiting fat breakdown in adipose tissue (Jocken *et al.* 2017). Lukovac *et al.* (2014) found that host epithelial genes involved in lipid metabolism and lipolysis are strongly regulated by propionate produced by microbial metabolism.

Similarly, previous studies have found a significant association between circulating short-chain fatty acids (especially acetate and propionate) and peripheral insulin sensitivity, systemic lipolysis, and glucagon-like peptide-1 concentration (Muller *et al.* 2019). Spielman *et al.* (2018) found that propionate can regulate body energy balance and appetite and enhance systemic glucose homeostasis by reducing fat breakdown in adipocytes and improving leptin secretion (Spielman *et al.* 2018). Multiple studies have shown that propionate can effectively inhibit the rate of fat breakdown and participate in the balance control of fat breakdown and fat generation (Lu *et al.* 2016). This indicates that propionate supplementation may be one of the effective measures to alleviate fat mobilization and compensate for energy deficiency. Most studies on postpartum cows mainly focus on lipid metabolism disorders. However, there are few reports on the molecular mechanisms related to fat mobilization in postpartum cows.

We hypothesized that postpartum supplementation of sodium propionate (NaPr) would increase glucose content in plasma and lipid deposition in adipose tissue, thereby alleviating NEB in cows. With this in mind, this study addresses two primary objectives: 1) to determine the effects of postpartum NaPr supplementation on the production performance and milk quality of dairy cows; and 2) to determine the effect of postpartum NaPr supplementation on fat synthesis in cow adipose tissue by detecting indicators related to lipid metabolism in adipose tissue.

2. Materials and methods

2.1. Animals, experimental design, and diets

The study involved 24 Holstein cows that were selected based on similar physical condition, parity (3), and expected calving date. The experimental design of this study was a completely randomized design. Cows were randomly assigned to either the control group (Control, n=12 per group), which was fed a total mixed ration (TMR), or the NaPr group (n=12 per group), where 246 g d^{-1} of NaPr additive was added to the TMR. The dosage of sodium propionate added in this experiment was based on Maldini's experimental protocol (Maldini *et al.* 2019). Uncoated NaPr (food grade) was purchased from Jiangsu Runpu Food Technology Co., Ltd. (purity≥99%, Lianyungang, Jiangsu, China). NaPr was added to the diet thrice daily (4:3:3 according to three diet times). The experimental diet was fed to cows from the day of delivery (d 0) to d 21 postpartum. All diets were isonitrogenous and isoenergetic and met the nutritional requirements of the National Research Council (NRC), USA (Table 1; NRC 2021). The average ambient temperature and relative humidity during the trials were (17.4±2)°C and (58±5)%, respectively. Cows were housed in individual stalls with unrestricted access to water and fed thrice daily (at 07:00, 13:00, and 20:00) at 105% of their *ad libitum* intake.

2.2. Feed sample collection and analysis

Daily feed intake data were recorded to determine DMI. TMR samples were collected on two consecutive days per week following the method described by Jiang *et al.* (2022). TMR samples collected were mixed, and a representative sample was finally obtained using the quartering method. In short, all samples were dried in an oven at 65°C for 48 h, ground using a Wiley mill (1188Y, Thomas Wiley, USA) with a 2 mm mesh size, and then stored for further analysis. Feed was milled and analyzed for dry matter (DM), ash, crude protein (CP), neutral detergent fiber (NDF), acid detergent fiber (ADF), and ether extract (EE). According to the method of the AOAC (2006), the nutrient content in feed samples was determined, including DM (method 930.15), crude fat (EE; method 981.10), crude ash (ash; method 942.05), CP (method 990.03), neutral detergent fiber and acid detergent fiber (NDF and ADF; method 973.18), calcium (Ca; method 985.35), and phosphorus (P; method 986.24).

2.3. Milk sample collection and analysis

Milk production was recorded daily, and 50 mL was sampled from the three consecutive milkings (06:00, 13:00, and 20:00; 4:3:3) during d 8, 15, and 21 after parturition. Each group consisted of 12 cows, and milk samples were collected from each cow in each group at each time point.

Each sample was placed in tubes with a preservative (0.05%

Table 1 Basal diet formulations and nutritional contents (dry matter (DM) basis %)

Ingredients	Content
Alfalfa	3.76
Oat	12.83
Whole corn silage	39.39
Corn	14.18
Soya bean meal	18.91
Cotton seed meal	1.75
DDGS	4.80
Calcium carbonate	0.6
Salt	0.5
Dicalcium phosphate	0.5
Premix[1)]	2.78
Total	100.00
Nutrient levels[2)]	
CP	16.07
EE	3.84
NDF	33.44
ADF	24.42
Ash	7.28
Ca	0.74
P	0.48
NE_L[3)], MJ kg^{-1}	3.25

[1)] Provided per kilogram of total mixed ration (TMR): 1.53 mg of Mn as $MnSO_4$; 17.38 mg of Zn as $ZnSO_4$; 0.12 mg of Se as Na_2SeO_3; 20.85 mg of Fe as $FeSO_4$; 8.90 mg of Cu as $CuSO_4$; 0.28 mg of I as KI; 0.15 mg of Se as Na_2SeO_3; 0.11 mg of Co as $CoCl_2$; 305,800 IU of vitamin A; 75,060 IU of vitamin D; 55,600 mg of vitamin E; and 1,167.6 mg of vitamin K3.
[2)] CP, crude protein; EE, ether extract; NDF, neutral detergent fiber; ADF, acid detergent fiber; Ca, calcium; P, phosphorus. Among them, CP, EE, NDF, ADF, Ash, Ca, and P are measured values; based on triplicate determinations.
[3)] Estimated based on the NRC (2021).

benzoic acid). The detection of indicators such as protein, fat, lactose, total solids (TS), and somatic cell count (SCC) in milk was carried out by Dairy One Cooperative Co., Ltd. (Shanghai, China). Milk urea nitrogen (MUN) levels were measured with a Beckman BUN Analyzer 2 (Beckman Instruments Inc., Fullerton, CA, USA). Preservatives (0.05% benzoic acid) were added to milk for sample preservation. The determination of fatty acids in milk followed the experimental method of Razzaghi *et al.* (2021). In short, 1 mL of liquid milk was placed in a centrifuge tube and equilibrated. The tube was centrifuged at 8,000 r min^{-1} for 30 min at 8°C. Afterwards, 1.8 mL of n-hexane/isopropanol (3:2, v/v) was added and vigorously vortexed for 30 s. Subsequently, 1.2 mL of sodium sulfate solution was added, and the resulting solution was vortexed for 30 s. The tube was centrifuged at 2,600 r min^{-1} for 5 min at 5°C. The upper layer or supernatant was transferred into a new tube, and 300 mg of ground anhydrous sodium sulfate was added. The tube was left to stand at room temperature for 30 min, filtered, and flushed with nitrogen at 40°C. The methylation process was accomplished by adding 2 mL of n-hexane and 40 μL of methyl acetate and then vortexing for 30 s. Then 40 μL of methylation reagent was added to the solution, vortexed for 2 min, and left to stand undisturbed for 8 min. Subsequently, 60 μL of termination reagent was added and vortexed for 30 s. Next, 100 mg of ground calcium chloride was added, then vortexed and left to stand for 1 h. The tube containing the solution was centrifuged at 2,600 r min^{-1} for 5 min at 5°C. The upper solution (supernatant) was transferred to a new extraction tube and flushed with nitrogen at 40°C. Finally, 500 μL of n-hexane was added to the tube, and the precipitate was stirred up by shaking for 30 s. It was then filtered using a 0.22 μm nylon filter membrane and placed in the inner tube of the gas chromatography (GC) analysis-specific vial. The sample was then stored at –20°C until used for detection analysis.

2.4. Blood sample collection and analysis

Each group consisted of 12 cows, and blood samples were collected on the d 7, 14, and 21 after calving. At each time point, blood samples were collected from the tail vein of each cow in each group before morning feeding. Blood samples were collected into tubes containing sodium heparin anticoagulant and kept on ice until centrifugation (4°C, 3,000 r min^{-1}, 20 min) to isolate plasma. Plasma biochemical indicators were detected using an automatic biochemical analyzer. According to the manufacturer's instructions, plasma non-esterified fatty acids (NEFA) concentrations were measured using a NEFA Kit (Wako Pure Chemicals Industries, Osaka, Japan). Plasma β-hydroxybutyrate (BHBA) concentrations were measured using a commercial kit (Stanbio, Boerne, TX, USA). TG and glucose concentrations were measured by enzymatic methods using commercial kits (Wiener Lab, Rosario, Argentina). Immunoglobulin A (IgA), immunoglobulin M (IgM), and immunoglobulin G (IgG) were measured using a commercial kit (Hengyi, Yangzhou, China).

2.5. Adipose tissue collection and analysis

On the d 21 after delivery, local anesthesia was administered to the cows. Six cows were randomly selected from each group of 12 cows, and subcutaneous adipose tissue was collected from the buttocks and immediately stored in liquid nitrogen to detect gene and protein expression levels. In accordance with the manufacturer's instructions, RNA extraction was performed using the TaKaRa RNA Extraction Kit (TaKaRa, code: RR036A, China) on collected adipose tissue. RNA concentration was then measured using an OD1000 instrument (1,000 drops, Nanjing, China). Subsequently, RNA was reverse transcribed into cDNA using Takara Reverse Transcription Kits (Tokyo, Japan). Real-time PCR detection was performed using the SYBR® Premix Ex TaqTM II Kit (Dalian, China). *β-actin*, *RPS9*, and *GAPDH* were identified as suitable reference genes for cow tissue using the $2^{-\Delta\Delta C_T}$ method (Hosseini *et al.* 2012), and their geometric mean was utilized for calculation consistency. Primers used in the experiment were synthesized by GENEWIZ Bioscience Co., Ltd. (Suzhou, China) and are listed in Appendix A.

NEFA concentrations in adipose tissue were measured using a NEFA C Kit (Wako Pure Chemicals Industries, Osaka, Japan). TG concentrations were measured by enzymatic methods using commercial kits (Wiener Lab, Rosario, Argentina).

The collected adipose tissue was placed into phosphate

buffer saline (PBS). The adipose tissue was gently incubated in a hypotonic extraction buffer (30 mmol L^{-1} sucrose, 17 mmol L^{-1} trisodium citrate dihydrate, 5 mmol L^{-1} EDTA, 0.5 mmol L^{-1} DTT, and 0.5 mmol L^{-1} PMSF, pH=8.2) at room temperature for 1 h. The adipose tissue was placed into 100 mmol L^{-1} sucrose (pH=8.2) droplets (approximately 40 μL), and the remaining suspension was added to the edge of a glass slide coated with fresh fixative solution (1% PFA, 0.15% Triton X-100, 10 mmol L^{-1} sodium borate, pH=9.2). The prepared glass slide was placed overnight in a humidified chamber at 4°C. The slide was then washed with 0.4% Photo-Flo (Kodak, 1464510), dried thoroughly, and stored at –20°C until use. Protein (Perilipin-1) immunofluorescence assay was performed as described in Jiang *et al.* (2023). Each group processed six samples and prepared six slices, and the average fluorescence intensity was calculated based on the fluorescence intensity obtained from the six slices.

Oil red O staining was performed to detect lipid droplets using the Lipid (Oil red O) Staining Kit from BioVision (Germany) according to the manufacturer's instructions. Finally, these slides were sealed with glycerin gelatin. The calculation of adipose tissue area was similar to the calculation of protein fluorescence intensity. Fat deposition in the adipose tissue was assessed through direct observation using an optical microscope (Olympus, Olympus Optical Ltd., Tokyo, Japan). The adipocyte area was measured using image analysis software (ImageJ 1.47, National Institutes of Health, USA).

2.6. Statistical analysis

For all data analyses, an individual cow was used as an experimental unit. The data from the current study were analyzed using the SPSS Statistical Software Package (v. 20.0, IBM Corp., Armonk, NY, USA) (Jiang *et al.* 2023). Specifically, feed intake, lactation performance, and plasma NEFA and BHBA data were analyzed using two-way ANOVA, while other indicators were analyzed using the Student's *t*-tests. A *P*-value less than 0.05 was considered statistically significant.

3. Results

3.1. Lactation performance

No treatment or treatment×day interactions were observed for DMI, milk yield, milk protein, milk fat, and milk lactose ($P>0.05$; Table 2), but an effect of day was observed ($P<0.01$; Table 2).

3.2. Milk FA profile

The milk profiles of saturated fatty acids (SFA) and *de novo* synthesis of FA (<16-carbon), mixed FA (16-carbon), and preformed FA (18-carbon) are presented in Table 3. The proportions of 6:0, 14:0, 17:0, 20:0, 21:0, and total odd-chain FA in milk fat were unaffected by treatments. However, the milk proportions of 4:0, 8:0, 10:0, 13:0, 16:0, total odd-chain FA ($P=0.02$), and *de novo* FA ($P=0.04$) increased, and those of all remaining individual SFA and preformed FA decreased ($P=0.01$) in cows fed NaPr *versus* the Control diet.

The milk proportions of individual unsaturated fatty acid (UFA), total *n*-6, and total *n*-3 FA are presented in Table 4. The milk proportions of *cis*-10 15:1 and *cis*-13, *cis*-16 22:2 increased ($P<0.01$), and those of all remaining individual UFA, total *n*-6 FA, and total *n*-3 FA were unaffected ($P>0.05$) in cows fed NaPr *vs.* the Control diet.

3.3. Changes in blood metabolites

The plasma concentrations of lipids metabolism, glucose metabolism, immunology, and antioxidants are presented in Table 5. Treatments did not affect the concentrations of lipids metabolism, immunology, and antioxidants in plasma. However, the plasma concentrations of Glu and INS increased ($P<0.01$ and $P=0.04$), and the TG concentration showed an increasing trend ($P=0.05$) in cows fed NaPr *vs.* the Control diet.

There were no interactions between treatment and time for NEFA and BHBA in plasma (Fig. 1). Compared with the Control group, the NaPr group decreased plasma NEFA concentration in cows ($P<0.05$, Fig. 1-A). Compared with d 0, 14, and 21 increased plasma NEFA concentration in cows ($P<0.05$, Fig. 1-B). However, d 7 had no significant effect on plasma NEFA concentration compared with d 0 ($P>0.05$, Fig. 1-B). Compared with d 14, plasma NEFA concentration was significantly increased on d 21 ($P<0.05$, Fig. 1-B).

Table 2 Effects of dietary sodium propionate (NaPr) on feed intake and lactation performance of dairy cows in postpartum

Item[1)]	Treatment		SEM[2)]	*P*-value		
	Control	NaPr		Treatment	Day[4)]	Treatment×Day
DMI (kg d^{-1})	17.61	17.25	0.64	0.32	<0.01	0.99
Milk yield (kg d^{-1})	33.52	33.87	0.45	0.64	<0.01	0.99
FCM (kg d^{-1})	33.78	34.97	0.74	0.42	<0.01	0.80
Milk protein (%)	3.38	3.37	0.03	0.73	<0.01	0.83
Milk fat (%)	4.11	4.22	0.05	0.16	<0.01	0.24
Milk lactose (%)	4.82	4.93	0.03	0.07	<0.01	0.87
SCC (10^3 mL^{-1})	220.81	212.03	99.16	0.21	0.68	0.89
MUN (mg dL^{-1})	8.21	8.12	1.19	0.52	0.27	0.54

[1)] DMI (dry matter intake): each value represents the mean of 108 replicates ($n=108$); milk yield: each value represents the mean of 84 replicates ($n=84$); other indicators: each value represents the mean of 36 replicates ($n=36$); FCM (fat corrected milk)=Milk yield×(0.4+0.15×Milk fat); SCC, somatic cell count; MUN, milk urea nitrogen.
[2)] SEM, standard error of mean.
[3)] Data collection of DMI was recorded at d 2, 4, 6, 8, 11, 13, 15, 18 and 21 after calving; data collection of milk yield was recorded at d 8, 10, 12, 15, 16, 18 and 20 after calving; milk samples for metabolites were tested at d 7, 14, and 21 after calving.

Table 3 Milk saturated fatty acids (SFA) profile and sum of FA in dairy cows fed experimental diets containing Control or sodium propionate (NaPr)

Item	Treatment		SEM[3]	*P*-value
	Control	NaPr		
SFA (g 100 g^{-1} of total FA)[1]				
$C_{4:0}$	2.04	2.28	0.046	<0.01
$C_{6:0}$	0.77	0.77	0.10	0.99
$C_{8:0}$	0.20	0.33	0.032	<0.01
$C_{10:0}$	1.45	2.20	0.26	0.01
$C_{12:0}$	2.56	3.41	0.46	0.08
$C_{13:0}$	0.22	0.34	0.033	<0.01
$C_{14:0}$	9.76	11.69	1.15	0.11
$C_{15:0}$	0.89	1.16	0.13	0.06
$C_{16:0}$	22.47	26.37	1.78	0.04
$C_{17:0}$	0.91	0.95	0.09	0.65
$C_{18:0}$	14.37	9.71	2.65	0.09
$C_{20:0}$	0.21	0.20	0.02	0.68
$C_{21:0}$	0.49	0.49	0.04	0.93
$C_{23:0}$	0.25	0.17	0.02	<0.01
Sum of FA[2]				
Total odd-chain FA	3.25	3.89	0.14	0.02
Total <16-carbon FA	17.54	21.58	1.02	0.04
Total 16-carbon FA	25.34	29.23	1.09	0.08
Total 18-carbon FA	52.84	44.11	1.80	0.01

[1] Each value represents the mean of 6 replicates (*n*=6).
[2] Total odd-chain FA=13:0+15:0+17:0+*cis*-10 15:1 (see Table 4) +*cis*-10 17:1 (see Table 4)+21:0+23:0; total<16-carbon FA (originated from *de novo* synthesis in the mammary gland) =4:0+6:0+8:0+10:0+12:0+14:0+10:1 (see Table 4)+*cis*-9 14:1 (see Table 4); total 16-carbon FA (mixed)=16:0+*cis*-9 16:1 (see Table 4); total 18-carbon FA (preformed FA)=18:0+all 18-carbon UFA (see Table 4, Razzaghi *et al.* 2021).
[3] SEM, standard error of mean.

Table 4 Milk unsaturated fatty acid (UFA) profile and sum of fatty acids (FA) in dairy cows fed experimental diets containing Control or sodium propionate (NaPr)

Item	Treatment		SEM[3]	*P*-value
	Control	NaPr		
UFA (g 100 g^{-1} of total FA)[1]				
cis-9 14:1	0.73	0.86	0.09	0.16
cis-10 15:1	0.00	0.30	0.01	<0.01
cis-9 16:1	2.86	2.85	0.44	0.97
cis-10 17:1	0.46	0.44	0.06	0.79
trans-9 18:1	4.07	2.28	2.24	0.43
cis-9 18:1	30.33	28.29	3.43	0.56
trans-9, *trans*-12 18:2	0.21	0.20	0.02	0.53
cis-9, *cis*-12 18:2	3.31	3.10	0.27	0.47
cis-6, *cis*-9, *cis*-12 18:3	0.19	0.18	0.02	0.74
cis-11, 20:1	0.27	0.26	0.03	0.84
cis-9, *cis*-12, *cis*-15 18:3	0.32	0.32	0.02	0.93
cis-8, *cis*-11, *cis*-14 20:3	0.22	0.16	0.03	0.08
cis-13, *cis*-16 22:2	0.00	0.23	0.01	<0.01
cis-5, *cis*-8, *cis*-11, *cis*-14, *cis*-17 20:5	0.32	0.32	0.04	0.97
Sum and ratio of FA[2]				
Total *n*-6 FA	3.53	3.27	0.14	0.36
Total *n*-3 FA	0.52	0.51	0.36	0.88

[1] Each value represents the mean of 6 replicates (*n*=6).
[2] Total *n*-6 FA=*cis*-9, *cis*-12 18:2+*cis*-8, *cis*-11, *cis*-14 20:3; total *n*-3 FA=*cis*-9, *cis*-12, *cis*-15 18:3+*cis*-5, *cis*-8, *cis*-11, *cis*-14, *cis*-17 20:5.
[3] SEM, standard error of mean.

Table 5 Plasma glucose and lipid metabolism, immunoglobulin, and antioxidant activity in dairy cows fed experimental diets containing Control or sodium propionate (NaPr)

Item[1]	Treatment		SEM[2]	*P*-value
	Control	NaPr		
Lipids metabolism				
TC (mmol L^{-1})	3.15	3.35	0.26	0.45
TG (μmol g^{-1})	121.82	167.78	22.23	0.05
HDL-C (mmol L^{-1})	2.27	2.47	0.18	0.27
LDL-C (mmol L^{-1})	0.50	0.43	0.06	0.32
Glucose metabolism				
Glu (mmol L^{-1})	2.93	3.54	0.18	<0.01
INS (μIU mL^{-1})	3.47	12.67	4.42	0.04
Immunology				
IgA (mg L^{-1})	14.92	15.42	1.60	0.76
IgG (mg L^{-1})	90.58	105.33	11.86	0.23
IgM (mg L^{-1})	40.50	39.92	3.85	0.88
Antioxidant				
SOD (U mL^{-1})	107.59	112.02	7.68	0.57
TAOC (mmol L^{-1})	6.33	6.27	0.30	0.84

[1] Each value represents the mean of 36 replicates (*n*=36); TC, total cholesterol; TG, triglyceride; HDL-C, high-density lipoprotein cholesterol; LDL-C, low-density lipoprotein cholesterol; Glu, glucose; INS, insulin; IgA, immunoglobulin A; IgM, immunoglobulin M; IgG, immunoglobulin G; SOD, super oxide dismutase; TAOC, total antioxidant capacity.
[2] SEM, standard error of mean.

3.4. Energy metabolism and mobilization of adipose tissue

The adipose tissue involved in lipid metabolism is presented in Fig. 2. Body weight was not affected by treatments. However, the adipose tissue levels of TG, Perilipin-1 (PLIN1) protein expression, and orange-red lipid droplet deposition were increased in cows fed NaPr *versus* the control diet (P<0.05). The NEFA content in the adipose tissue of the NaPr group showed an increasing trend (0.05<P<0.1).

3.5. mRNA expression of lipid metabolism genes in adipose tissue

The mRNA expression of lipid metabolism genes in the adipose tissue is presented in Table 6. The mRNA expression of *HK2*, *GLUT8*, *ACCα*, and *ACSS2* increased (P<0.05),

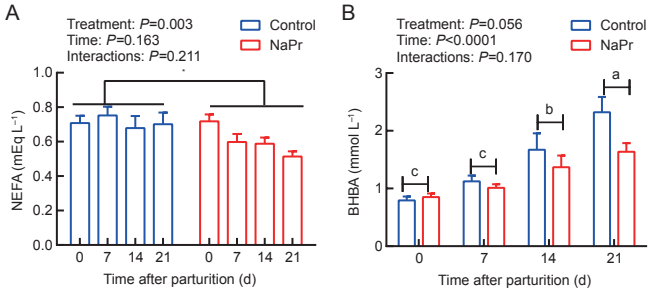


Fig. 1 Effect of dietary sodium propionate (NaPr) on energy metabolism at d 0, 7, 14 and 21 after parturition (*n*=12). A, the concentration of non-esterified fatty acids (NEFA) in plasma. B, the concentration of β-hydroxybutyrate (BHBA) in plasma. Data are mean±SE. Different letters indicate significant differences among treatments (P<0.05). *, P<0.05.

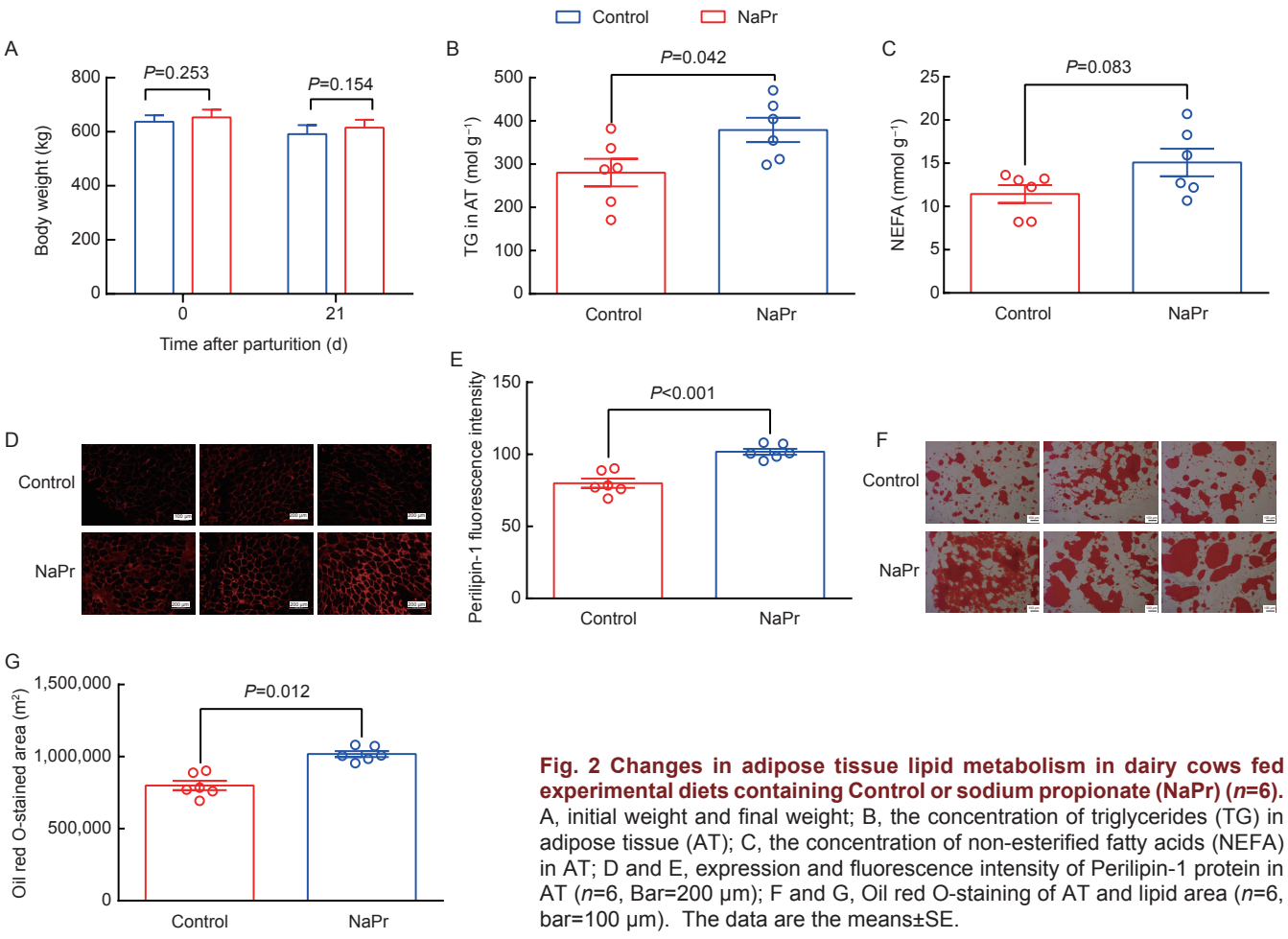


Fig. 2 Changes in adipose tissue lipid metabolism in dairy cows fed experimental diets containing Control or sodium propionate (NaPr) (*n*=6). A, initial weight and final weight; B, the concentration of triglycerides (TG) in adipose tissue (AT); C, the concentration of non-esterified fatty acids (NEFA) in AT; D and E, expression and fluorescence intensity of Perilipin-1 protein in AT (*n*=6, Bar=200 μm); F and G, Oil red O-staining of AT and lipid area (*n*=6, bar=100 μm). The data are the means±SE.

and that of *CPT1A*, *CPT2*, and *PPARα* decreased ($P<0.01$, $P=0.05$, and $P=0.02$) in cows fed NaPr *vs.* the control diet.

4. Discussion

The results of this experiment supported the hypothesis that postpartum supplementation of NaPr could improve NEB in cows by increasing plasma glucose levels and promoting lipid deposition in adipose tissue. Supplementing with sodium propionate increased the levels of glucose and TG in plasma. Propionate, as a precursor of hepatic gluconeogenesis, has a positive impact on glucose synthesis. NaPr promotes lipid synthesis in adipose tissue, reducing fat breakdown, as evidenced by decreased plasma NEFA levels. In addition, BHBA is one of the essential indicators reflecting the energy metabolism of dairy cows (Sun *et al.* 2020). On d 21, the sodium propionate treatment group significantly reduced the content of BHBA in plasma. Therefore, these results highlight the key role of NaPr in postpartum relief of negative energy balance in cows, and its rational use provides a strategy for alleviating NEB during the perinatal period.

During the transition period, cows are affected by various physiological stresses, resulting in lower DMI, hormone changes, and a high incidence rate (Contreras *et al.* 2018). Especially in high-performance cows, the reduction in DMI makes it unable to meet the nutritional needs of large-scale milk production, leading to the occurrence of NEB (Lopreiato *et al.* 2020). Therefore, increasing energy metabolism and reducing excessive fat mobilization to generate NEFA in transitional period cows are some of the most effective measures to alleviate NEB in cows.

Compared to acetate and butyrate in the liver, propionate is crucial for the oxidation of the tricarboxylic acid cycle (Allen and Piantoni 2013). Gluconeogenesis is one of the primary energy sources for ruminants, as the potential glucose in the ingested carbohydrates is usually metabolized by microorganisms in the rumen. However, after metabolism in the rumen, the glucose absorbed by the small intestine is insufficient to meet the body's needs (Yost *et al.* 1977). Therefore, the propionate produced by rumen microbial fermentation is usually the primary source of liver gluconeogenesis (Aschenbach *et al.* 2010). Evidence suggests that plasma NEFA was reduced in lactating cows supplemented with calcium propionate, which may be due to increased energy provided by calcium propionate, thereby reducing the need for fat mobilization (DiCostanzo *et al.* 1999). Generally speaking, a decrease in DMI often leads to an increase in plasma NEFA. However, the increased energy demand of lactating cows leads to a rise in NEFA, and the addition of propionate reduces fat mobilization, which may reduce the content of NEFA. Our experimental results found that supplementing the diet with NaPr did not lessen the DMI of cows in the postpartum period compared to the control group. McNamara and Valdez (2005)

Table 6 Adipose tissue mRNA expression of glucose and lipid metabolism genes in dairy cows fed experimental diets containing Control or sodium propionate (NaPr)

Gene[1)]	Treatment		SEM[2)]	*P*-value
	Control	NaPr		
Glucose metabolism				
HK1	1.04	0.64	0.15	0.02
HK2	1.23	5.47	1.12	<0.01
GLUT1	1.28	3.54	1.17	0.08
GLUT8	1.45	6.87	2.48	0.05
Lipid metabolism				
GPR41	2.15	3.06	1.52	0.56
ACCα	1.07	2.27	0.38	0.01
CPT1A	1.04	0.46	0.13	<0.01
CPT2	1.09	0.62	0.21	0.05
DGAT1	1.27	1.35	0.61	0.89
DGAT2	1.26	1.19	0.61	0.91
FAS	1.14	2.10	0.78	0.25
SCD1	1.59	1.30	0.78	0.72
SREBP1	1.12	0.85	0.38	0.51
ACSL1	1.24	0.72	0.47	0.29
ACSL3	1.11	0.64	0.26	0.09
ACSS1	1.12	0.64	0.13	0.47
ACSS2	1.05	2.27	0.33	<0.01
HSL	1.01	1.92	0.82	0.29
PLIN1	1.04	2.15	0.75	0.17
PPARα	1.17	0.36	0.30	0.02
PPARγ	1.43	0.38	0.62	0.12

[1)] Each value represents the mean of 6 replicates (n=6).
[2)] SEM, standard error of mean.

found that feeding propionate during the perinatal period did not affect the DMI of transitional cows, which is consistent with our results. Interestingly, compared to the control group, cows fed NaPr showed a trend of increased milk lactose concentration on d 15 and 20. As a precursor for lactose synthesis, the increase of propionate may promote milk lactose synthesis.

Nutrition is the main factor affecting milk fat secretion in cows. The adjustment of diet will affect milk production and the composition of milk components. The synthesis of fatty acids in milk and milk fat content are extensively studied in relation to human dietary needs. Manipulating the diets of dairy cows to modify these factors has become a popular area of research due to its potential impact on human well-being (Liu *et al.* 2016). Therefore, we also observed the changes in FA content in the milk of lactating cows during the postpartum period after supplementing the diet with NaPr. Maxin *et al.* (2010) showed that the infusion of propionic acid at a dose of 500 g d^{-1} in Holstein cows during late lactation did not alter milk fat percentage and milk fat production but altered the FA profile in milk. Perfusion of propionic acid resulted in decreased production and percentage of $C_{4:0}$ and $C_{18:0}$ fatty acids in milk. At the same time, all odd-chain FA increased from $C_{7:0}$ to $C_{17:0}$. However, in this experiment, although a decrease in the proportion of $C_{18:0}$ fatty acids was observed in milk, the addition of NaPr up-regulated the content of $C_{4:0}$ and $C_{8:0}$ fatty acids in milk. This may be because the cows were in the postpartum period in this experiment, and some of the milk fat energy came from fat mobilization. Although we did not detect $C_{7:0}$, $C_{9:0}$, and $C_{11:0}$ fatty acids, experiments have shown that NaPr increases the content of $C_{13:0}$, $C_{15:0}$, and $C_{15:1}$ fatty acid in milk. The increase in these odd-chain FA may be due to the rise in propionic acid that can be used as a precursor for *de novo* synthesis in the mammary gland. Other studies have also shown that increased odd-chain FA can be manipulated to improve insulin sensitivity and reduce the risk of diabetes (Sun *et al.* 2018). The oxidative capacity of mitochondria is improved mainly through partial oxidation of odd-chain FA to form succinyl CoA (Lean *et al.* 2013). Dietary supplementation with NaPr does not change the milk fat rate and milk production of dairy cows but changes the FA profile in milk, increases the content of odd-chain FA in milk, and improves milk quality.

Lipids (especially TG) provide more energy storage than the body's protein and glycogen reserves. When the energy intake exceeds the animal's needs, it can lead to fat being stored in the form of TG. The body can mobilize these reserves when the energy demand exceeds the current supply. In cow production, the concentration of NEFA in the plasma increases when fat mobilization occurs. During the transition from late pregnancy to early lactation, a large amount of adipose tissue is mobilized, increasing plasma NEFA to support the increased energy demand of cows. After delivery in cows, an increase in plasma NEFA concentration can be observed to compensate for the energy required for NEB. However, fat mobilization after delivery in cows typically exceeds the liver's ability to oxidize NEFA (McNamara and Valdez 2005), leading to the formation of ketone bodies (such as BHBA) and the re-esterification of TG (Brickner *et al.* 2009). In this study, we found that the concentration of BHBA in the plasma of cows during the postpartum period consistently increased, which is consistent with Gross's results. Under the influence of time, adding NaPr to the diet reduced the concentration of BHBA and the incidence of ketosis (Gross *et al.* 2011).

In this study, glucose and lipid metabolism-related indicators were detected in the plasma of cows on the 21st day of the postnatal period. It was found that supplementing with NaPr significantly increased the concentrations of TG, glucose, and insulin in the plasma of cows. This indicates that supplementing with NaPr in the postpartum period can provide energy for cows, alleviate NEB, and thereby reduce body fat mobilization, which is beneficial for the body's glucose and lipid balance. McCarthy *et al.* (2015) found that feeding cows with high starch and *Monensin* during the postpartum period increased rumen propionic acid production, increased plasma glucose and insulin concentrations, and reduced plasma NEFA and BHBA concentrations, thereby alleviating weight loss in cows and reducing the incidence of subclinical ketosis. This is consistent with our initial hypothesis that the body can absorb NaPr as a precursor of gluconeogenesis to maintain energy metabolism balance. However, the mechanism of changes in glucose concentration in plasma involves multiple factors such as gluconeogenesis, glycogen breakdown, and glucose absorption. Therefore, further experiments are needed to verify the mechanism by which NaPr up-regulates plasma glucose and insulin concentrations in cows during the postpartum period.

During the postpartum period, cows need an appropriate level of fat mobilization to adapt to the transition from pregnancy to lactation. TG is an efficient and inert intermediate for storing and transporting FA deposited in adipose tissue. Compared to cows' protein and glycogen reserves, TG provides high-calorie energy storage. In this study, supplementing with NaPr in the diet increased the concentration of TG in adipose tissue, and most of the TG stored in adipocytes was taken up by circulating blood TG, which is consistent with the increase in TG concentration in the plasma of cows in the NaPr group. It is worth noting that fat mobilization initiates the breakdown of TG into glycerol and NEFA. However, the concentration of NEFA in adipose tissue in this experiment was not affected by NaPr.

TG in plasma is hydrolyzed by lipoprotein lipase (LPL) to form fatty acids, which then enter adipocytes with the assistance of transport proteins (Kersten 2014). Finally, it is stored in lipid droplets in the form of TG under the action of lipase. Glucose produces glycerol-3-phosphate through glycolysis, which can further undergo esterification with fatty acids to promote lipid production. In this experiment, although the mRNA expression of related fatty acid synthase *DGAT1*, *DGAT2*, *FAS*, and *SCD1* did not change, the mRNA expression of glucose transporter 8 (*GLUT8*) and glycolytic gene (*HK2*) was up-regulated. This indicates that inhibiting lipid breakdown by NaPr may be mediated by promoting liver gluconeogenesis and up-regulating plasma glucose concentration, activating insulin to promote glycolysis and increase lipid synthesis. Due to the poor specificity of the relevant antibodies currently available on the market for these genes, these proteins have not been successfully captured by Western blot. As an adipocyte-specific lipid-coated protein, PLIN1 has been shown to regulate adipose lipid synthesis (Sun *et al.* 2013), cows (Zhang *et al.* 2018), and humans (Natori *et al.* 2017). Under basic conditions, PLIN1 can prevent lipolysis but increase lipid synthesis and lipid droplet formation (Straub *et al.* 2008). In this study, TG determination, PLIN1 fluorescent protein expression, and Oil red O staining of adipose tissue showed that PLIN1 expression increased in cows fed NaPr *versus* the control diet. This result also indicates that NaPr promotes lipid synthesis in the adipose tissue of cows during the postpartum period.

In addition, propionic acid can also regulate lipid metabolism through the acetyl-coenzyme A pathway. Acetyl CoA participates in synthesis in the form of malonyl CoA. Acetyl CoA carboxylates to form malonyl CoA, which ACC catalyzes. This reaction is a rate-limiting step in FA synthesis, activated by citric acid isomerization and inhibited by palmitoyl CoA (McGlory *et al.* 2019). However, there are two subtypes of ACC, namely *ACCα* and *ACC2*. The former is located in the cytoplasm, and the generated malonyl CoA is used for fatty acid synthesis. *ACC2* is located on the cytoplasmic surface of mitochondria, and the product is used to inhibit *CPT1* to reduce FA oxidation (Zhao *et al.* 2016). In this experiment, after supplementing with NaPr, the mRNA expression of *ACCα* and *ACSS2* in adipose tissue was up-regulated, and the expression of *CPT1A* and *CPT2* genes was down-regulated. This indicates that supplementing with NaPr in the diet promotes the generation of acetyl CoA, thereby up-regulating the FA synthesis pathway and reducing FA oxidation.

5. Conclusion

This study has demonstrated that the diet with NaPr in the postpartum period can provide energy for cows, which is beneficial for the body's glucose and lipid balance. In summary, postpartum supplementation of NaPr alleviates negative energy balance in cows by increasing plasma glucose levels and promoting lipid deposition in adipose tissue. The effectiveness of prenatal NaPr treatment should be evaluated in large-scale ranch environments to better assess productivity and health responses.

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Declaration of competing interest

The authors declare that they have no conflict of interest.

Declaration of generative AI and AI-assisted technologies in the writing process

The authors declare that they did not use AI in the preparation and writing of this manuscript.

Ethical approval

The experiment was conducted at the experimental farm of Yangzhou University (Yangzhou, China), and the Yangzhou University Institution Animal Care and Use Committee (IACUC) approved all procedures involving animals (SYXK (Su) 2016-0019).

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References

Allen M S, Piantoni P. 2013. Metabolic control of feed intake: Implications for metabolic disease of fresh cows. *Veterinary Clinics of North America: Food Animal Practice*, **29**, 279–297.

AOAC (Association of Official Analytical Chemists). 20026. *Official Methods of Analysis.* 18th ed. Association of Official Analytical Chemists, Gaithersburg, MD.

Aschenbach J R, Kristensen N B, Donkin S S, Hammon H M, Penner G B. 2010. Gluconeogenesis in dairy cows: The secret of making sweet milk from sour dough. *International Union of Biochemistry and Molecular Biology Life*, **62**, 869–877.

Brickner A E, Pires J A, Gressley T F, Grummer R R. 2009. Effects of abomasal lipid infusion on liver triglyceride accumulation and adipose lipolysis during fatty liver induction in dairy cows. *Journal of Dairy Science*, **92**, 4954–4961.

Contreras G A, Strieder-Barboza C, De Koster J. 2018. Symposium review: Modulating adipose tissue lipolysis and remodeling to improve immune function during the transition period and early lactation of dairy cows. *Journal of Dairy Science*, **101**, 2737–2752.

DiCostanzo A, Williams J E, Keisler D H. 1999. Effects of short- or long-term infusions of acetate or propionate on luteinizing hormone, insulin, and metabolite concentrations in beef heifers. *Journal of Animal Science*, **77**, 3050–3056.

Gross J, van Dorland H A, Bruckmaier R M, Schwarz F J. 2011. Performance and metabolic profile of dairy cows during a lactational and deliberately induced negative energy balance with subsequent realimentation. *Journal of Dairy Science*, **94**, 1820–1830.

Herdt T H, Emery R S. 1992. Therapy of diseases of ruminant intermediary metabolism. *Veterinary Clinics of North America*: *Food Animal Practice*, **8**, 91–106.

Hosseini A, Behrendt C, Regenhard P, Sauerwein H, Mielenz M. 2012. Differential effects of propionate or beta-hydroxybutyrate on genes related to energy balance and insulin sensitivity in bovine white adipose tissue explants from a subcutaneous and a visceral depot. *Journal of Animal Physiology and Animal Nutrition*, **96**, 570–580.

Jiang M, Hu Z, Wang K, Yang T, Lin M, Zhan K, Zhao G. 2023. CRISPR/Cas9-mediated knockout of *SLC15A4* gene involved in the immune response in bovine rumen epithelial cells. *Journal of Integrative Agriculture*, **22**, 3148–3158.

Jiang M C, Datsomor O, Cheng Z Q, Meng Z T, Zhan K, Yang T Y, Huang Y H, Yan Q, Zhao G Q. 2022. Partial substitution of alfalfa hay by Stevia (*Stevia rebaudiana*) hay can improve lactation performance, rumen fermentation, and nitrogen utilization of dairy cows. *Frontiers in Veterinary Science*, **9**, 899148.

Jocken J W E, Gonzalez Hernandez M A, Hoebers N T H, van der Beek C M, Essers Y P G, Blaak E E, Canfora E E. 2017. Short-chain fatty acids differentially affect intracellular lipolysis in a human white adipocyte model. *Front Endocrinol* (Lausanne), **8**, 372.

Kersten S. 2014. Physiological regulation of lipoprotein lipase. *Biochimica et Biophysica Acta*, **1841**, 919–933.

Lean I J, Van Saun R, Degaris P J. 2013. Energy and protein nutrition management of transition dairy cows. *Veterinary Clinics of North America: Food Animal Practice*, **29**, 337–366.

Liu S, Zhang R, Kang R, Meng J, Ao C. 2016. Milk fatty acids profiles and milk production from dairy cows fed different forage quality diets. *Animal Nutrition*, **2**, 329–333.

Lopreiato V, Mezzetti M, Cattaneo L, Ferronato G, Minuti A, Trevisi E. 2020. Role of nutraceuticals during the transition period of dairy cows: A review. *Journal of Animal Science and Biotechnology*, **11**, 96.

Lu Y, Fan C, Li P, Lu Y, Chang X, Qi K. 2016. Short chain fatty acids prevent high-fat-diet-induced obesity in mice by regulating G protein-coupled receptors and gut microbiota. *Scientific Reports*, **6**, 37589.

Lukovac S, Belzer C, Pellis L, Keijser B J, de Vos W M, Montijn R C, Roeselers G. 2014. Differential modulation by *Akkermansia muciniphila* and *Faecalibacterium prausnitzii* of host peripheral lipid metabolism and histone acetylation in mouse gut organoids. *mBio*, **5**, e01438-14.

Maldini G, Kennedy K M, Allen M S. 2019. Temporal effects of ruminal infusion of propionic acid on hepatic metabolism in cows in the postpartum period. *Journal of Dairy Science*, **102**, 9781–9790.

Maxin G, Glasser F, Rulquin H. 2010. Additive effects of *trans*-10, *cis*-12 conjugated linoleic acid and propionic acid on milk fat content and composition in dairy cows. *Journal of Dairy Research*, **77**, 295–301.

McCarthy M M, Yasui T, Ryan C M, Pelton S H, Mechor G D, Overton T R. 2015. Metabolism of early-lactation dairy cows as affected by dietary starch and monensin supplementation. *Journal of Dairy Science*, **98**, 3351–3365.

McGlory C, Calder P C, Nunes E A. 2019. The influence of omega-3 fatty acids on skeletal muscle protein turnover in health, disuse, and disease. *Frontiers in Nutrition*, **6**, 144.

McNamara J P, Valdez F. 2005. Adipose tissue metabolism and production responses to calcium propionate and chromium propionate. *Journal of Dairy Science*, **88**, 2498–2507.

Muller M, Hernandez M A G, Goossens G H, Reijnders D, Holst J J, Jocken J W E, van Eijk H, Canfora E E, Blaak E E. 2019. Circulating but not faecal short-chain fatty acids are related to insulin sensitivity, lipolysis and GLP-1 concentrations in humans. *Scientific Reports*, **9**, 12515.

Natori Y, Nasui M, Kihara-Negishi F. 2017. Neu1 sialidase interacts with perilipin 1 on lipid droplets and inhibits lipolysis in 3T3-L1 adipocytes. *Genes Cells*, **22**, 485–492.

NRC (National Research Council). 2021. *Nutrient Requirements of Dairy Cattle10.17226/25806*. National Academies Press, USA.

Razzaghi A, Drackley J K, Malekkhahi M. 2021. Concentrate allowance and corn grain processing influence milk production, body reserves, milk fatty acid profile, and blood metabolites of dairy cows in the early postpartum period. *Journal of Dairy Science*, **104**, 5479–5492.

Schoenberg K M, Ehrhardt R M, Overton T R. 2012. Effects of plane of nutrition and feed deprivation on insulin responses in dairy cattle during late gestation. *Journal of Dairy Science*, **95**, 670–682.

Silva A S, Cortinhas C S, Acedo T S, Morenz M J F, Lopes F C F, Arrigoni M B, Ferreira M H, Jaguaribe T L, Ferreira L D, Gouvea V N, Pereira L G R. 2022. Effects of feeding 25-hydroxyvitamin D_3 with an acidogenic diet during the prepartum period in dairy cows: Mineral metabolism, energy balance, and lactation performance of Holstein dairy cows. *Journal of Dairy Science*, **105**, 5796–5812.

Spielman L J, Gibson D L, Klegeris A. 2018. Unhealthy gut, unhealthy brain: The role of the intestinal microbiota in neurodegenerative diseases. *Neurochemistry International*, **120**, 149–163.

Straub B K, Stoeffel P, Heid H, Zimbelmann R, Schirmacher P. 2008. Differential pattern of lipid droplet-associated proteins and *de novo* perilipin expression in hepatocyte steatogenesis. *Hepatology*, **47**, 1936–1946.

Sun B, Cao Y, Cai C, Chao Y, Xiang L, Yao J. 2020. Temporal dynamics of nutrient balance, plasma biochemical and immune traits, and liver function in transition dairy cows. *Journal of Integrative Agriculture*, **19**, 820–837.

Sun Y, Liu B, Du Y, Snetselaar L G, Sun Q, Hu F B, Bao W. 2018. Inverse association between organic food purchase and diabetes mellitus in US adults. *Nutrients*, **10**, 1877.

Sun Z, Gong J, Wu H, Xu W, Wu L, Xu D, Gao J, Wu J W, Yang H, Yang M, Li P. 2013. Perilipin1 promotes unilocular lipid droplet formation through the activation of Fsp27 in adipocytes. *Nature Communications*, **4**, 1594.

Xue M, Sun H, Wu X, Guan L L, Liu J. 2018. Assessment of rumen microbiota from a large dairy cattle cohort reveals the pan and core bacteriomes contributing to varied phenotypes. *Applied and Environmental Microbiology*, **84**, 0978.

Yost W M, Young J W, Schmidt S P, McGilliard A D. 1977. Gluconeogenesis in ruminants: Propionic acid production from a high-grain diet fed to cattle. *Journal of Nutrition*, **107**, 2036–2043.

Zhang F, Wang Y, Wang H, Nan X, Guo Y, Xiong B. 2022a. Calcium propionate supplementation has minor effects on major ruminal bacterial community composition of early lactation dairy cows. *Frontiers in Microbiology*, **13**, 847488.

Zhang F, Zhao Y, Wang H, Nan X, Wang Y, Guo Y, Xiong B. 2022b. Alterations in the milk metabolome of dairy cows supplemented with different levels of calcium propionate in early lactation. *Metabolites*, **12**, 699.

Zhang Q, Koser S L, Donkin S S. 2016. Propionate induces mRNA expression of gluconeogenic genes in bovine calf hepatocytes. *Journal of Dairy Science*, **99**, 3908–3915.

Zhang S, Liu G, Xu C, Liu L, Zhang Q, Xu Q, Jia H, Li X, Li X. 2018. Perilipin 1 mediates lipid metabolism homeostasis and inhibits inflammatory cytokine synthesis in bovine adipocytes. *Frontiers in Immunology*, **9**, 467.

Zhao S, Torres A, Henry R A, Trefely S, Wallace M, Lee J V, Carrer A, Sengupta A, Campbell S L, Kuo Y M, Frey A J, Meurs N, Viola J M, Blair I A, Weljie A M, Metallo C M, Snyder N W, Andrews A J, Wellen K E. 2016. ATP-citrate lyase controls a glucose-to-acetate metabolic switch. *Cell Reports*, **17**, 1037–1052.

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