Effects of a blend of *Saccharomyces cerevisiae*-based direct-fed microbial and fermentation products on plasma carbonyl-metabolome and fecal bacterial community of beef steers

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INTRODUCTION

Direct-fed microbials (DFM) are commonly used in livestock production systems to improve the metabolic and energy status of animals especially during stress periods, thereby leading to improved animal productivity (Boyd et al., 2011; Broadway et al., 2015; Xu et al., 2017). Several studies have suggested that the effects of DFM on the metabolic and energy status of ruminants are attributed to the modulation of the rumen microbiota, improved gut integrity, and increased intestinal nutrient absorption (Sun et al., 2013; Qiao et al., 2010; Philippeau et al., 2017). The objective of this study was to apply a CIL/LC-MS-based quantitative untargeted metabolomics to evaluate the effects of PROB on the plasma concentrations of carbonyl-containing metabolites in beef steer during a 42-d receiving period.

MATERIALS AND METHODS

- Forty newly-weaned Angus crossbred steer calves (7 days post-weaning; 210 ± 12 kg of body weight (BW); 180 ± 17 d of age) were stratified by BW into 4 weight blocks.
- The steers were randomly assigned (within each weight block) to 1 of 2 treatments for a period of 42 d.
  - Diet with no additive (**CON**; n = 20)
  - CON + 19 g of Commence Additive (**PROB**; n = 20)
- The basal diet (corn silage-based) was fed daily as a total mixed ration at 08:00 h
- Commence™ Feed Additive (PMI, Arden Hills, MN) is a blend of *S. cerevisiae*, *Enterococcus lactis*, *Bacillus subtilis*, *Enterococcus faecium, and L. case*i, and their fermentation products.
- Body weights of steers were obtained before morning feeding on d 0, 21 and 42. The quantity of feed offered to each steer was recorded daily. Diet refused (as fed) was also measured daily.
- On d 42, blood samples were taken for plasma carbonyl-metabolome profiling using a chemical isotope labelling/liquid chromatograph/mass-spectrometric method.
- Rectal fecal samples were also collected approximately 4 hours after feeding on d 40 for bacterial community analysis according to the Illumina 16S Metagenomic Sequencing Library protocols.

RESULTS

Table 1. Identified peak pairs that were affected by dietary supplementation of a blend of *S. cerevisiae*-based direct-fed microbials and fermentation products.

Compound	Fold Change	FDR
Galactose	2.60	< 0.01
Lactose	0.46	< 0.01
Glucose	2.62	< 0.01
Fructose	2.31	< 0.01
Isomer of fructose	2.30	< 0.01
Isomer of glyceraldehyde	2.01	0.01
Glyceraldehyde	2.01	0.01
Hippuric acid	2.13	< 0.01
Phenylacetylglycine	1.98	0.01
5-hydroxykynurenamine	2.63	< 0.01
4-oxoglutaramate	1.82	< 0.01
2-dehydro-3-deoxy-D-glucarate	1.80	< 0.01
3-fumarylpyruvate	2.58	< 0.01
1-deoxy-D-xylulose 5-phosphate	2.36	< 0.01
Glycolaldehyde	1.63	0.01
Hydroxypyruvate	1.60	< 0.01
2-dehydro-3-deoxy-L-arabinonate	0.30	< 0.01
Acetoacetate	0.62	0.01
Dehydroascorbate - 2 tags	1.74	0.01
3-methylindolepyruvate	3.72	< 0.01
(S)-2-aceto-2-hydroxybutanoate	2.96	< 0.01
5-oxopentanoate	2.30	< 0.01
(R)-3-hydroxy-3-methyl-2-oxopentanoate	2.96	< 0.01
2-dehydropantoate	3.28	< 0.01
Isomer of (S)-2-aceto-2-hydroxybutanoate	3.51	< 0.01
Isomer of (S)-3-methyl-2-oxopentanoic acid	2.19	< 0.01

FC: fold change relative to control.
P-value was calculated from student's t-test.
Only metabolites with both fold-change ≥ 1.5 or ≤ 0.67 and FDR ≤ 0.01 are shown.

Table 2. Pearson correlations between plasma metabolites and performance indices of the beef steers.

	ADG		FE	
	r	P-value	r	P-value
3-(4-hydroxyphenyl)pyruvate	0.27	0.09	0.22	0.18
(S)-2-aceto-2-hydroxybutanoate	0.31	0.06	0.25	0.13
5-oxopentanoate	0.43	0.01	0.36	0.03
(R)-3-hydroxy-3-methyl-2-oxopentanoate	0.31	0.06	0.24	0.14
2-dehydropantoate	0.31	0.06	0.25	0.12
Isomer of (S)-2-aceto-2-hydroxybutanoate	0.33	0.04	0.27	0.09
Isomer of (S)-3-methyl-2-oxopentanoic acid	0.32	0.05	0.30	0.07

Only metabolites with correlation coefficient (r) of *P*-value ≤ 0.10 for either average daily gain (ADG) or feed efficiency (FE) are shown.

RESULTS

Table 3. Relative abundance of the dominant fecal bacterial genera (> 0.01% of total sequences) that were affected by dietary supplementation of a blend of *S. cerevisiae*-based direct-fed microbials and fermentation products.

Genus (% of total sequences)	CON	PROB	SE	P-value
Prevotellaceae UCG-003	1.91	4.15	0.48	0.03
p-2534-18B5 gut group*	0.81	0.00	0.60	0.01
Elusimicrobium	0.26	0.01	0.18	0.02
Megasphaera	0.00	0.07	0.00	0.01
Moheibacter	0.08	0.00	0.05	0.04
Comamonas	0.06	0.00	0.04	0.01
Dorea	0.07	0.15	0.01	0.02
Stenotrophomonas	0.04	0.00	0.02	0.01
Blautia	0.04	0.09	0.01	0.01
Acetitomaculum	0.01	0.04	0.00	0.01

*Uncultured bacterium belonging to the indicated family

CONCLUSIONS

- Supplementation of PROB improved the energy status of the beef steers by increasing the relative concentrations of plasma monosaccharides such as glucose, galactose, fructose, and glyceraldehyde, as well as others (hippuric acid, phenylacetylglycine, and 5-hydroxykynurenamine) with possible health benefits.
- Supplementation of PROB altered the fecal bacterial population towards increased relative abundance of *Prevotellaceae* UCG-003 and some lactate-utilizing bacteria.

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