

Comparative effects of two multi-species direct-fed microbial products on rumen fermentation, bacterial community and metabolome of beef steers

INTRODUCTION

In the last decade, efforts to improve ruminant productivity ha primarily focused on manipulating the ruminal microbial commun and fermentation (DiLorenzo, 2011). One of such efforts include use of direct-fed microbials (DFM). In recent years, most commerce DFM products are formulated to contain several microbial species a their fermentation products in order to ensure multi-factorial respon (McAllister et al., 2011). The objective of this study was to compare the effects of two DFM products containing multiple microbial spec and their fermentation products on ruminal fermentation, bacter community and metabolome of beef steer during a three 21-d period receiving period.

MATERIALS AND METHODS

- Nine rumen-cannulated Holstein steers (mean ± SD body weight (BW): 243 ± 12.4 kg) were assigned to 3 treatments arranged in a 3×3 Latin square design with three 21-d periods and 10-d wash-c between periods.
- Dietary treatments were (1) Control (CON; basal diet without additive), (2) Commence (PROB; basal diet plus 19 g/d of Commence), and (3) RX3 (SYNB; basal diet plus 28 g/d of RX3).
- Commence is a blend of active S. cerevisiae, Enterococcus lactis, Bacillus subtilis, Enterococcus faecium, and L. casei, and their fermentation products ((PMI, Arden Hills, MN).
- RX3 is a blend of active S. *cerevisiae* and the fermentation produce of S. cerevisiae, Enterococcus lactis, Bacillus licheniformis, and Bacillus subtilis (PMI, Arden Hills, MN).
- On d 21 of each period, 150 mL of the ruminal contents were collected via the cannula at 5 different sites within the rumen at approximately 1, 3, 6, 9, 12, and 18 h after feeding.
- Ruminal fluid samples collected at different time points were used analyze the following;
- VFA, lactate, and ammonia nitrogen concentrations
- Untargeted metabolome profile using a chemical isotope labeling (CIL)/liquid chromatography-mass spectrometry (LC-MS)based technique to target the amine/phenol-containing submetabolome
- Bacterial community composition using 16S rRNA sequencing

Ibukun M. Ogunade, Andres Pech Cervantes, D. M. Paulus Compart College of Agriculture, Food Science, Communities, and the Environment, Kentucky State University, Frankfort, KY 40601 Agricultural Research Station, Fort Valley State University, Fort Valley, GA 31030, USA $I = 1 \cap I = 1$

ermentation products or Item	CON	PROB	SYNB	SEM	P-v	value
Acetate, mM	25.8	27.8	30.5	2.02	0	.32
Propionate, mM	8.32 ^b	11.1 ^a	12.3ª	0.95	0	.03
Valerate, mM	0.99 ^b	1.76 ^a	1.55 ^a	0.18	0	.01
Isovalerate, mM	0.43 ^y	0.55 ^x	0.58 ^x	0.05	0	.10
Butyrate, mM	6.89	7.99	7.90	0.77	0	.83
Isobutyrate, mM	0.32	0.35	0.39	0.04	0	.58
Total VFA, mM	52.5 ^b	59.9 ^a	61.6 ^a	2.71	0	.01
Lactate, mM	0.63	0.69	0.53	0.20	0	.63
Ammonia-N, mM	3.72 ^a	2.13 ^b	3.21 ^a	0.42	0	.05
рН	6.67	6.54	6.71	0.12	0	.52
CON = control; PROB = a and their fermentation prod fermentation products of S. PMI, Arden Hills, MN). SEM = standard error of me A,b Within a row, treatment n A,y Within a row, treatment n	lucts fed at 19 g/ste cerevisiae, Entero ean; neans with differen neans with differen	eer/day (PMI, Arden Lococcus lactis, Bacillu nt superscripts differ, nt superscripts tend to	Hills, MN); SYN as licheniformis, a $P \le 0.05$. differ, $0.05 < P$	$B = a blend of and Bacillus su \leq 0.10.$	live S. cerevisia abtilis fed at 28 g	<i>d L</i> . ne a g/ste
CON = control; PROB = a and their fermentation prod fermentation products of S. PMI, Arden Hills, MN). SEM = standard error of me ,b Within a row, treatment n	lucts fed at 19 g/ste cerevisiae, Entero ean; neans with differen neans with differen tary supplement	eer/day (PMI, Arden Lococcus lactis, Bacillu at superscripts differ, at superscripts tend to ation of direct-fed	Hills, MN); SYN as licheniformis, a $P \le 0.05$. differ, $0.05 < P$ microbial cont	$B = a blend of and Bacillus su \leq 0.10.$	live S. cerevisia btilis fed at 28 g	<i>d L</i> . <i>ne</i> a g/ste
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SEM = standard error of mean;

Only bacterial genera ($\geq 0.1\%$ of total sequences) that were different in any of the treatment groups ($P \leq 0.05$) are shown. ^{a,b}Within a row, treatment means with different superscripts differ, $P \le 0.05$.

Table 3. Id Item

Taurine

Glutamyl-pi

Valine

Phenylalani

Cystathioni

Norleucine

Histidinyl-p

Tyrosine

Para-creso

Isomer of (F

3-(2,3-dihyd

Normalized RT (retention time) FC: fold change relative to Control; Tier 1 - Positive Identification (CIL Library); Tier 2 - High Confidence Putative Identification (LI Library). No identified metabolites were affected by supplemental SYNB. PROB = a blend of S. cerevisiae, Enterococcus lactis, Bacillus subtilis, Enterococcus faecium, and L. casei, and their fermentation products fed at 19 g/steer/day (PMI, Arden Hills, MN); SYNB = a blend of live S. cerevisiae and the fermentation products of S. cerevisiae, Enterococcus lactis, Bacillus licheniformis, and Bacillus subtilis fed at 28 g/steer/day PMI, Arden Hills, MN). Only metabolites with both fold-change ≥ 1.2 or ≤ 0.83 , relative to Control and $P \leq 0.05$ are shown.

• This study demonstrated that supplementation of PROB or SYNB differentially altered the ruminal bacterial community towards increased relative abundance of bacteria (Succinivibrio, Rikenellaceae, and Succinivibrionaceae) that can produce succinate and that of *Succiniclasticum* that can ferment succinate to propionate.

• The differential bacterial community and metabolome shift caused by the additives resulted into similar concentrations of ruminal fermentation acids

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RES Identified metabolites that w	ULTS were affected b	v sunnle	mental P R	OR
	Normalized RT	FC	P-value	Identification level
	157.6	1.50	0.04	Tier 1
oroline	389.6	1.42	0.03	Tier 1
	662.4	1.26	0.04	Tier 1
ine	783.2	1.73	0.01	Tier 1
ine	830.8	0.70	0.04	Tier 1
	837.8	1.63	0.01	Tier 1
proline	1098.2	1.25	0.03	Tier 1
	1363.3	1.62	0.04	Tier 1
	1482.6	1.42	0.01	Tier 1
R)-1-aminopropan-2-ol	413	0.51	0.04	Tier 2
droxyphenyl)propanoate	756.9	0.72	0.02	Tier 2

CONCLUSIONS

REFERENCES

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