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Effects of dietary neutral detergent fiber and starch ratio on rumen epithelial cell morphological structure and gene expression in dairy cows

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ABSTRACT

This study was designed to investigate the effect of dietary neutral detergent fiber to starch ratio on rumen epithelial morphological structure and gene expression. Eight primiparous dairy cows including 4 ruminally fistulated cows were assigned to 4 total mixed rations with neutral detergent fiber to starch ratios of 0.86, 1.18, 1.63, and 2.34 in a replicated 4 × 4 Latin square design. The duration of each period was 21 d including 14 d for adaptation and 7 d for sampling. Rumen epithelial papillae were collected from the ruminally fistulated cows for morphological structure examination and mRNA expression analysis using quantitative real-time PCR of several genes related to volatile fatty acid absorption and metabolism, and cellular growth. Increasing dietary neutral detergent fiber to starch ratio resulted in a linear increase in the thickness of the stratum spinosum and basale. In contrast, expression of *HMGCS2* (encoding the rate-limiting enzyme in the synthesis of ketone bodies) decreased linearly, whereas the expression of *MCT2* (encoding a transporter of volatile fatty acid) increased linearly with increasing dietary neutral detergent fiber to starch ratio. As dietary neutral detergent fiber to starch ratio increased, expression of *IGFBP5* (a gene related to the growth of rumen epithelial papillae) decreased, whereas *IGFBP6* expression increased. Both of these *IGFBP* genes are regulated by short-chain fatty acids. Overall, the data indicate that dietary neutral detergent fiber to starch ratio can alter the thickness of the rumen epithelial papillae partly through changes in expression of genes associated with regulating volatile fatty acid absorption, metabolism, and cell growth.

Key words: dairy cow, gene expression, neutral detergent fiber to starch ratio, rumen morphological

INTRODUCTION

Dietary NDF to starch ratio is a key determinant of carbohydrate composition of ruminant diets, and it could affect the composition and content of VFA in the rumen. Epithelial papillae lining the rumen increase the surface area for VFA absorption. Previous research indicated that dietary chemical composition and physical properties were the main factors affecting the morphological structure of the rumen epithelial papillae (Beharka et al., 1998; Steele et al., 2011; Liu et al., 2013). In addition, carbohydrate composition of the diet could influence the rumen environment and might affect the morphological structure of the rumen papillae (Steele et al., 2011).

The integrity of the rumen morphological structure is the key to ensuring absorption of the VFA. Classical studies indicated that 60 to 80% of the VFA were absorbed in the rumen, and VFA could supply 60 to 80% of the maintenance energy requirements of the dairy cow (Bergman, 1990; Baldwin, 1998). Although many factors affect the absorption rate of VFA by the rumen epithelium, the main factors include the carbon chain length, pH, rumen epithelium surface area, concentration of VFA, and the osmotic pressure of the rumen fluid (Gaebel et al., 1987; Odongo et al., 2006).

Previous research demonstrated that rumen epithelial papilla development was increased by feeding a high content of digestible carbohydrate, which in turn enhanced density and surface area of the rumen epithelium, and hence, overall VFA absorption capacity (Goodlad, 1981). Thus, compared with a high-fiber diet, the ability of the rumen epithelium to absorb increasing amounts of VFA could be improved by feeding highly digestible carbohydrate such as starch (Aschenbach et al., 2009).

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In dairy nutrition, dietary carbohydrate composition can be assessed through the content of NDF, NFC, and NDF to NFC ratio (Armentano and Pereira, 1997). Thus, the composition of rumen-fermentable carbohydrates and physically effective fiber, and their interaction, should be considered when formulating diets (Allen, 1997). The NDF to starch ratio has been successfully used to investigate the effect of carbohydrate composition on nutrient digestibility and milk production (Beckman and Weiss, 2005; Zhao et al., 2016). Whether or not the rumen epithelium responds to a change in NDF to starch ratio is unclear. Thus, the objective of this study was to investigate the effect of different dietary NDF to starch ratios on rumen morphological structure and expression of genes related to VFA adsorption, papilla cell growth, and metabolism in dairy cows.

MATERIALS AND METHODS

Animals, Experimental Design, and Diets

The experiment was performed at the Sino-farm, Beijing, China. All the experimental procedures were approved by the Animal Care and Use Committee for Livestock issued by the Institute of Animal Science, Chinese Academy of Agricultural Sciences. Eight primiparous dairy cows including 4 ruminally cannulated cows were assigned to 4 TMR with NDF to starch ratios of 0.86, 1.18, 1.63, and 2.34 in a replicated 4 × 4 Latin square design. The duration of each period was 21 d including a 14-d adaptation and 7 d for sampling. At the beginning of the study, milk production and DIM were 35.1 ± 0.5 kg/d and 146 ± 21.6, respectively. The dietary treatments were achieved by altering the proportions of forage (corn silage and oat hay) and concentrate (corn) inclusion to achieve the desired NDF to starch ratios, with the composition and inclusion proportion of other dietary ingredients in the TMR remaining the same (Table 1). Cows were housed in a free-stall barn and had free access to water. They were milked 3 times daily at 0700, 1400, and 2100 h. Feed was supplied 2 times daily at 0800 and 1600 h for ad libitum intake, with feed allocation adjusted to achieve approximately 5% refusals.

Sample Collection and Optical Microscope Scanning

Rumen epithelial papillae were collected on d 18 through the rumen cannula after partial emptying of

Table 1. Ingredient and composition of TMR with NDF to starch ratios of 0.86 (T1), 1.18 (T2), 1.63 (T3), and 2.34 (T4; 12 samples analyzed)

	Diet (NDF to starch ratios)			
Item	T1	T2	T3	T4
Ingredient, % of DM				
Alfalfa hay	15.0	15.0	15.0	15.0
Corn silage	20.0	25.0	30.0	35.0
Oat hay	0	5.0	10.0	15.0
Ground corn	35.0	25.0	15.0	5.0
Soybean meal	15.0	15.0	15.0	15.0
Extruded soybean	5.0	5.0	5.0	5.0
Whole cottonseed	3.0	3.0	3.0	3.0
Beet pulp	2.5	2.5	2.5	2.5
EB100[1]	2.0	2.0	2.0	2.0
Limestone	1.4	1.4	1.4	1.4
Sodium chloride	0.6	0.6	0.6	0.6
Premix[2]	0.5	0.5	0.5	0.5
DM, %, as-fed basis	47	50	51	54
Nutrition composition, % of DM				
OM	93.0	92.4	91.6	91.2
CP	17.5	17.6	17.6	17.6
NDF	29.8	34.0	37.7	41.2
Starch	34.4	28.8	23.2	17.6
Calcium	0.9	0.9	0.9	1.0
Phosphorus	0.4	0.4	0.4	0.4
NE_L,[3] Mcal/kg	1.83	1.76	1.69	1.62

[1]Mainly saturated free fatty acid fat supplement (EnergyBooster 100, Milk Specialties Global, Eden Prairie, MN).

[2]Contained (per kg of DM) 250,000 to 560,000 IU of vitamin A, 65,000 to 134,300 IU of vitamin D, 2,100 to 4,500 IU of vitamin E, 400 to 600 mg of Fe, 540 to 1,200 mg of Cu, 2,100 to 4,100 mg of Zn, 560 to 1,350 mg of Mn, 15 to 60 mg of Se, 35 to 70 mg of I, and 68 to 120 mg of Co.

[3]Calculated based on Ministry of Agriculture of P.R. China (People's Republic of China, 2004).