



J. Dairy Sci. 101:1–19  
<https://doi.org/10.3168/jds.2017-13858>  
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## Symposium review: Understanding diet–microbe interactions to enhance productivity of dairy cows<sup>1</sup>

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### ABSTRACT

Ruminants are dependent on the microbiota (bacteria, protozoa, archaea, and fungi) that inhabit the reticulo-rumen for digestion of feedstuffs. Nearly 70% of energy and 50% of protein requirements for dairy cows are met by microbial fermentation in the rumen, emphasizing the need to characterize the role of microbes in feed breakdown and nutrient utilization. Over the past 2 decades, next-generation sequencing technologies have allowed for rapid expansion of knowledge concerning microbial populations and alterations in response to forages, concentrates, supplements, and probiotics in the rumen. Advances in gene sequencing and emerging bioinformatic tools have allowed for increased throughput of data to aid in our understanding of the functional relevance of microbial genomes. In particular, metagenomics can identify specific genes involved in metabolic pathways, and metatranscriptomics can describe the transcriptional activity of microbial genes. These powerful approaches help untangle the complex interactions between microbes and dietary nutrients so that we can more fully understand the physiology of feed digestion in the rumen. Application of genomics-based approaches offers promise in unraveling microbial niches and respective gene repertoires to potentiate fiber and nonfiber carbohydrate digestion, microbial protein synthesis, and healthy biohydrogenation. New information on microbial genomics and interactions with dietary components will more clearly define pathways in the rumen to positively influence milk yield and components.

**Key words:** dairy cow, diet–microbe interaction, productivity, omics approach

### INTRODUCTION

As the global population continues to climb toward 9.7 billion in the next 30 yr and to 11.2 billion by 2100 (United Nations, 2017), the demand for high-quality animal protein (meat, milk, and eggs) will increase exponentially to meet the world's growing dietary needs. Advancements in the American livestock sector over the past 25 yr have generated significant improvements in animal genetics, nutrition, animal health, and herd management, which have led to increased production efficiencies of 54% in dairy and 45% in beef production (USDA-NASS, 2014). Additional increases in efficiency at the animal and herd levels will be required to meet global demands for animal protein (Gerber et al., 2013). Future endeavors will need to focus on the rumen environment to improve the efficiency of feed utilization by the microbiota (Weimer, 2015). Increased feed efficiency directs more nutrients to milk production, resulting in fewer nutrients wasted in urine and feces and reducing nutrient excretion into the environment. Improvements in animal production systems must address societal concerns and will need to recognize the impact of such systems on the environment (Hristov et al., 2013). Greenhouse gases (GHG) produced from ruminants have been an area of environmental concern (Morgavi et al., 2013; Knapp et al., 2014). Advances in feed efficiency to improve animal production will need to include rumen methane mitigation strategies to reduce GHG emissions including identification and modification of the microbes involved in methane production (Hristov et al., 2013; Connor, 2015). Innovative approaches to understanding digestive physiology and enhancing feed fermentation in the rumen will be necessary to ensure further increases in milk production efficiency.

Microbial fermentation in the rumen is fundamental to ruminant production and is driven by a diverse range of bacteria, protozoa, and fungi that work together to digest plant materials and contribute substantially to ruminant host metabolism (Hobson and Stewart, 2012; McCann et al., 2014; Kumar et al., 2015). The neutral

Received September 18, 2017.

Accepted April 30, 2018.

<sup>1</sup>Presented as part of the Ruminant Nutrition Symposium: Ruminant Metagenomics in Dairy Cattle—Beyond Microbial Diversity at the ADSA Annual Meeting in Pittsburgh, Pennsylvania, in June 2017.

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detergent insoluble fiber polysaccharides are cellulose and hemicellulose and the nonfiber carbohydrates include sugars, starch, and water-soluble carbohydrates (pectin,  $\beta$ -glucans, galactans). Fiber and nonfiber carbohydrates are fermented by microbes to VFA, and intermediary substrates of fermentation are utilized by microbes for cell growth. Absorbed VFA are used for host energy utilization and biosynthesis, signifying the symbiotic relationship of the rumen microbiome to the cow. Nearly 70% of energy (Annison and Armstrong, 1970) and 50% of the protein needs of dairy cows are met from rumen microbial fermentation (Smith, 1975). Other fermentation products include carbon dioxide and methane, which are not useful for the microbes or the host and are lost primarily through eructation (Hobson and Stewart, 2012). Strategies to enhance energy production and formation of microbial protein from rumen fermentation while reducing methane formation are needed to maximize rumen fermentation to relay increased energy and protein to the cow.

Traditional cultivation techniques have enabled us to characterize cellulolytic, amylolytic, proteolytic, and methanogenic microbes, which together contribute less than 11% of the total active microbes present in the rumen (Edwards et al., 2004). Over the past decade, next-generation sequencing (NGS) technologies coupled with “omics” approaches have become powerful tools to identify the microbial populations present and active in the rumen and how they respond to dietary components and time of exposure (Kumar and Pitta, 2015). This review describes the techniques and limitations of NGS technologies used to promote our knowledge and understanding of diet–microbe interactions required to improve rumen fermentation and products produced by dairy cows.

### SIGNIFICANCE OF THE RUMEN MICROBIOME TO DAIRY COWS

Dairy cows depend on rumen microbes to digest plant polymers to provide energy and protein for their growth and productivity. Forages alone can meet most of the nutrient requirements of dairy cows, but milk production in these systems is typically low. High-yielding dairy cows require grain supplementation to meet the energy required for increased milk production (Hills et al., 2015). Forages and concentrates are composed of varying proportions of carbohydrates (structural and nonstructural), nitrogenous substances, and lipids, all of which are partly or completely fermented in the rumen by microbes. Intermediary substrates of rumen fermentation can be utilized by microbes to provide energy (ATP) for assembling nitrogenous compounds, lipids,

and nucleic acids for microbial cell growth (Hobson and Stewart, 2012). The products of microbial fermentation in the rumen are VFA, carbon dioxide, methane, and ammonia (Hungate, 1984). The VFA are not utilized for energy by microbes but are readily absorbed and utilized as energy for host metabolism, making this relationship truly symbiotic. Strategies to manipulate rumen fermentation processes to optimize rumen performance should aim to benefit both microbes and host. A better understanding of microbial composition and the functional role of microbes in fermentation is first required to manipulate the rumen microbiota.

Broadly, microbial functions can be divided into 2 categories: symbiotic relationships between microbes and the host and nonsymbiotic microbial processes that benefit the microbial community (Figure 1). Most of the energy required for both microbes and the dairy cow is derived from fiber and nonfiber carbohydrate digestion, and the availability of rumen N (ammonia and peptides) to maximize microbial protein supply to the cow (Hoover and Stokes, 1991). In contrast, methane is an end product of microbial fermentation and is formed when methanogens capture hydrogen released from other microbes. The energy cost associated with methane production has approached 12% net energy loss to the dairy cow (Hristov et al., 2015), although microbes benefit through disposal of reducing potential. Biohydrogenation (BH) is a microbial process to escape the toxic effects of unsaturated fatty acids and, in the process, consumes reducing equivalents ( $H^+$ ). When dairy cows are fed high concentrations of starch and rumen-available fats concurrently, alternate microbial BH pathways trigger the formation of *trans*-fatty acid (*trans*-10, *cis*-12 C18:2 CLA; *trans*-10 C18:1) isomers. These *trans* fatty acids are potent inhibitors of milk fat de novo synthesis in the mammary gland (Bauman and Griinari, 2003). To maintain a stable rumen ecosystem, symbiotic relationships should be promoted. Future studies can offer opportunities to explore the manipulation of nonsymbiotic microbial processes on dairy cow performance.

### ADVANCES IN DAIRY COW NUTRITION TO IMPROVE MILK PRODUCTION

Nutritional advancements (Figure 1) have included better characterization of fiber (NDF) and NFC and development of RDP and RUP, a system to better describe the interactions between rumen-available carbohydrates and RDP for microbial growth. Advances in fat metabolism have centered on the balance of rumen-active and rumen-inert fats to maintain normal rumen BH pathways and to supply precursors for milk

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