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Review Article

The Role of Direct-Fed Microbes to Ruminants: A Review

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ABSTRACT
Feed additives are used in livestock feed and feeding to increase feed quality, the utility of feed derived from animals, and the performance and health of the animals. Digestibility gravies, rumen flora stabilizers, and microbial are some of the zoo's technological additions. Direct feed microbial are characterized as microbial-based feed additives, with a tighter definition than probiotics. It improves feed use by boosting energy usage per unit of feed and enhancing fiber digestibility. The term direct-fed microbial (DFM) was coined by the Food and Drug Administration and the American Feed Regulator Representatives Associations to describe a feed product that contains live, naturally occurring microorganisms such as bacteria, fungi, and yeast; the bacteria can produce or consume lactic acid. Microbial feed additives have traditionally been given to animals during stressful times in the hopes of establishing a beneficial microbe population in the digestive tract, which would reduce or prevent harmful organism development. DFM has several mechanisms of action, some of which affect the rumen and others which affect the gastrointestinal system. Lactic acid-generating bacteria (LAB) have a favorable impact on the rumen by reducing ruminal acidosis, encouraging the proliferation of ruminal microorganisms that have adapted to the presence of lactic acid in the rumen, and boosting lactic acid-using bacteria (LUB). LUB has been presented as a DFM that can lower lactate levels while maintaining ruminal pH. Through hydrophobic interactions, DFM can block or

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prevent pathogens like Escherichia coli from attaching to the intestinal mucosa. DFM medication helps dairy calves adapt quickly to solid feed by speeding up the formation of ruminal and intestinal microbes and preventing the spread of enteric pathogens, which can cause diarrhea. DFM was utilized to improve dairy cow performance by improving dry matter intake, milk output and protein content, as well as blood glucose and insulin levels before and after delivery. DFM is critical in beef cattle to prevent ruminal acidosis induced by highly fermentable diets, as well as to promote growth, meat output, and feed efficiency. Powders, pastes, boluses, and capsules are only some of the direct-fed microbial products available. It can be added to feed or ingested by drinking water. According to one study, feeding more than 107 CFU per head per day may cause lower nutrient absorption due to overpopulation in the gastrointestinal tract.

Keywords: Direct feed microbial, lactic acid-producing bacteria, lactic acid utilizing bacteria, mode of action.

INTRODUCTION

Direct feed microbial are characterized as microbial-based feed additives, with a tighter definition than probiotics. It improves feed use by increasing fiber digestibility, boosting energy use per unit of feed, and lowering feed costs (Beauchemin *et al*., 2008). overcrowding of the gastrointestinal tract has the power to influence the immunological system of the host

Ruminant animals and bacteria have developed a symbiotic connection in which microorganisms may ferment plant cell wall polysaccharides that are resistant to mammalian enzymatic degradation. The symbiotic connection fills a need in the ecosystem, and the conversion of complex plant sugars to energy benefits both the host animal and microbial symbionts (Knapp *et al*., 2014).

The reticulum, rumen, omasum, and abomasum make up the ruminant digestive system. The principal fermentation activities in the ruminant's digestive tract take place mostly in the rumen (Tharwat *et al*., 2012). Microorganisms create the enzymes found in the rumen. These enzymes help ruminants digest and ferment their food, hence the Rumen is thought of as a fermentation vat (Aschenbach *et al*., 2011).

Even with intensely concentrated feeding systems, forages remain the most significant component of ruminant animals' diets (Beauchemin *et al*., 2003). However, huge amounts of fiber components creating plant cell walls limit energy accessibility from forages, limiting feed intake and animal performance (Jung and Allen, 1995). Producers have been encouraged to provide greater starch diets due to fiber digestion issues. However, dietary starch content can be difficult to control and can have negative repercussions on the rumen environment, putting cows at risk for subacute ruminal acidosis and a frequent digestive problem (Enermark, 2008).

Rumen microbial research aim to increase feed utilization, animal productivity and health, and animal food safety for the reasons stated above. These objectives can be met by utilizing feed additives to facilitate optimal fermentation, reduce ruminal diseases, and eliminate pathogens. Antibiotics, probiotics, and prebiotics have been researched in the rumens and digestive tracts of cattle animals to modify the microbial environment and fermentation properties (Hong *et al*., 2005).

Poppy (2008) defines feed additives as "items used in animal nutrition to increase the quality of feed, the quality of food from animals, or the performance and health of animals." They are divided into the following categories: Technological supplements (e.g. preservatives, antioxidants, emulsifiers, stabilizing agents, acidity regulators, silage additives), Sensory enhancers (e.g. flavors, colorants), Vitamins, minerals, amino acids, and trace elements are examples of nutritional additions (e.g. digestibility enhancers, gut flora stabilizers) Histomonostats and coccidiostats (additives used in poultry diets for health reasons).

Feed costs account for 40 to 60% of the overall cost of production in farm animals (Bozic *et al*, 2012), hence nutritionists are always looking for ways to improve feed utilization, which may be done by improving feedstuff dietary digestibility. As a result, the goal of this study is to evaluate the microorganisms employed as DFM, their mechanism of action and effects, as well as practical considerations and a path ahead.

LITERATURE REVIEW

Direct-fed microbes (DFM)

Sub-therapeutic dosages of antibiotics have been used in feed to stimulate growth and preserve health in farm animals, but feeding antibiotics to animals was forbidden in the European Union (EU) in 2006 owing to worries about rising bacterial antibiotic resistance in people (Prieto *et al*., 2014). Concerns over the use of antibiotics in animal agriculture have sparked interest in finding alternatives to antimicrobial feed additives (Martin *et al*., 1999).

The potential use of probiotics in feeding operations has been emphasized by societal concerns about the use of antibiotics and other growth stimulants in agricultural production, as well as the necessity for farmers to adopt preventative measures against pathogen outbreaks in the food supply (Elam *et al*., 2003; Krehbiel *et al*., 2003). Ruminal probiotics are "live cultures of microorganisms that are intentionally introduced into the rumen to enhance animal health or nutrition," according to the definition. Probiotic is a broad phrase that refers to a variety of microbial cultures, extracts, and enzyme preparations (Elam *et al*., 2003).

The term Direct-Fed Microbials (DFM) is used to describe feed products that contain a source of life, naturally occurring microorganisms, such as bacteria, fungi, and yeast, according to the Food and Drug Administration's Office of Regulatory Affairs (2003) and the Association of American Feed Control Officials (1999).

DFM is a wide term that encompasses a variety of conditions. They may be divided into three categories: bacterial, fungal, and a mix of both. Lactic acid-generating bacteria (LAB), lactic acid-using bacteria (LUB), and other microorganisms are all possible classifications for bacterial DFM strains. Lactobacillus, Propionibacterium, Bifidobacterium, Enterococcus, Streptococcus, and Bacillus are all frequent bacteria found in bacterial DFM for ruminants, as well as Megasphaera elsdenii and Prevotella bryantii (Kung, 2006; Seo *et al*., 2010). DFM grows in the rumen and changes the microbial environment and fermentation properties for the better. DFM may potentially find a home in the digestive system (Seo *et al*., 2010).

To be successful, microbial feed supplement solutions must meet the following criteria: The microbial strain must be non-pathogenic and non-toxic to the host animal, capable of producing antimicrobial agents, antagonistic toward pathogenic (Kullen and Klaenhammer, 1999), able to adhere to and colonize the epithelial cells of the rumen and gut, capable of competing with normal microflora and metabolizing in the gut environment (e.g., resistant to low pH, organic acids, bile salts, and digestive enzymes), genetically stable (Parvez *et al*., 2006).

Microorganisms Used as Direct-Fed Microbes

Supplementation of fungal cultures (Aspergillus oryzae, Saccharomyces cerevisiae), lactate producing (Enterococcus) and lactate-utilizing (Propionibacterium) bacterial species, as well as Bifidobacterium spp., and Bacillus spp., are the most common DFM interventions of ruminal fermentation to promote desirable intestinal microflora, improve nutrient utilization, and stabilize pH to promote (NRC, 2001; Beauchemin, 2003 and FAO, 2013)

Based on the Food and Drug Administration (FDA, 2003) and the Association of American Feed Control Officials (AAFC, 1999), Seo *et al*., (2010) identified the microorganisms that are employed as direct feed microbial for ruminants. Those microorganisms are listed in Table 1.

Modes of Action of DFM

According to Azzaz *et al*., (2016), microbial feed additives have traditionally been given to animals during stressful times in the hopes of establishing a beneficial microorganism population in the digestive system, which would reduce or prevent harmful organism establishment. When an animal is stressed, however, the gut microflora changes. An increase in the quantity of coliform and other enterotoxigenic bacteria is frequently seen.

Any of the following factors might cause an animal to become stressed: Environmental stress (thermal, moisture, crowding, and sanitary conditions), Emotional stress (handling or shipping, changes in pen-mates, and weaning), and Disease stress (nutrient shortage or excess, and antagonism between levels of two or more nutrients) (infectious and metabolic). To boost production performance, change ruminal fermentation, or improve nutrient usage, microbial feed additives should be supplied continually (Azzaz *et al*., 2016).

Table 2: Microorganisms used as DFM for ruminants

Genus		Species
Lactic acid-producing bacteria (LAB)		
	Lacto bacillus	*Lactobacillusplantarum*
		Lactobacillus casei
		Lactobacillus gallinarum
		Lactobacillus salivarius
		Lactobacillus reuteri
		Lactobacillus bulgaricus
	Bifidobacterium	*Bifidobacterium pseudolongum*
		Bifidobacterium thermophilium
		Bifidobacterium longum
		Bifidobacterium lactis
	Streptococcus	*Streptococcus bovis*
		Streptococcus faecium
	Enterococcus	*Enterococcus faecium*
		Enterococcus faecal
Lactic acid utilizing bacteria (LUB)		
	Megasphaera	*Megasphaera elsdenii*
	Propionibacterium	*Propionibacterium shermanii*
		Propionibacterium freudenreichii
		Propionibacterium acidipropionici
		Propionibacterium jensenii
Other bacteria		
	Prevotella	*Prevotella bryantii*
	Bacillus	*Bacillus subtilis*
		Bacillus licheniformis
		Bacillus coagulans
Yeast		
	Saccharomyces	*Saccharomyces cerevisiae*
		Saccharomyces boulardii
Fungi		
	Aspergillus	*Aspergillus oryzae*
		Aspergillus niger

Source: Seo *et al*., (2010)

Bacterial DFM

Many factors influence the manner of action of bacterial DFM, including doses, feeding periods and frequencies, and DFM strains. DFM affects the rumen and the gastrointestinal system in different ways (Puniya *et al*., 2015).

(1) within the rumen: LAB and LUB have a major role in the mode of action of various DFM sources in the rumen. LAB may have a favorable effect on the rumen by avoiding ruminal acidosis in dairy cows (Nocek *et al*., 2002). By activating LUB and facilitating the proliferation of ruminal bacteria suited to the presence of lactic acid in the rumen. LUB has been presented as a DFM that can lower lactate levels while maintaining ruminal pH. (Yoon and Stern, 1995). When fed a highly fermentable diet, *Megasphaera elsdenii,* the predominant lactate-utilizing bacteria in the rumen, inhibits the severe pH reductions induced by lactate buildup in the rumen (Yang *et al*., 2004 and Kung, 2006). Another bacterial species observed in large numbers in the rumen of animals fed forage and medium concentration diets is the Propioni bacterium (Kung, 2006). Among the volatile fatty acids, propionate is the most essential single precursor of glucose production (VFA). Stein *et al*. (2006) discovered that specific species of Propioni bacteria can alter rumen fermentation and increase the molar fraction of ruminal propionate. In early lactation in dairy cows, it can convert lactate to propionate, resulting in enhanced hepatic glucose production, more substrates for lactose synthesis, improved energy efficiency, and reduced ketosis (Weiss *et al*., 2008). Propionate is thought to account for 61 to 67 percent of glucose release in developing ruminants and nursing cows (Huntington, 2000). According to the stoichiometric rules of chemical balance and its equation, increasing propionate has been followed by a reduction in methane (CH_4) generation (Van Soest, 1994).

(2) In the post-ruminal gastrointestinal tract, DFM can restrict or prevent pathogens like Escherichia coli from sticking to the intestinal mucosa via hydrophobic interactions, as well as limit pathogens from binding to the enterocytic receptor or creating enterotoxins that cause diarrhoea (Lee *et al*., 2003 and Kung 2006). LAB was able to stick to the intestine and protect the mice against Salmonella (Frizzo *et al*., 2010). LAB has important functions in infiltrating microbial cells and interfering with fundamental cell function, in addition to creating lactate and acetate as key metabolic end-products (Holzapfel *et al*., 1998).

Another mechanism is that DFM and LAB can create antibacterial chemicals with competitive exclusion and probiotic properties, such as bacteriocin and hydrogen peroxide. The sulfhydryl groups of metabolic enzymes such as glucose transport enzymes, hexokinase, and glycerol aldehyde-3-phosphate dehydrogenase can be oxidized by hydrogen peroxide, leading glycolysis to be blocked (Dicks and Botes, 2010). LAB bacteriocins, on the other hand, can prevent substrates from binding to the rib nucleotide reductase subunit, therefore interfering with target microorganism DNA synthesis (Dicks and Botes, 2010).

DFM have the power to influence the immunological system of the host. Dendritic cells, natural killer cells, macrophages, neutrophils, and T and B lymphocytes are among the immune cells found in the GIT's Peyer's patches, lamina propria, and intraepithelial areas (Krehbiel *et al*., 2003). DFM are immediately taken up by intestinal epithelial cells by transcytosis, then engulfed by antigen-presenting cells, macrophages, or dendritic cells, ultimately triggering an immunological response (Dicks and Botes, 2010).

Fungal DFM

In ruminants, fungal DFMs are commonly utilized to improve performance and regulate rumen fermentation. The most commonly employed species are Saccharomyces Cerevisiae and Aspergillus Oryzae (Elghandour *et al*., 2014a and Puniya *et al*., 2015).

When ruminants are given fungal-based DFM, several mechanisms have been proposed to explain changes in ruminal fermentation and improvements in performance. Fungal cultures may help the ruminal bacteria Selenomonas ruminantium utilize lactate more effectively by supplying dicarboxylic acids and other growth factors. When ruminants are fed high concentration diets, yeast may aid to buffer excess lactic acid generation by mediating the abrupt dips in rumen pH. (Kung, 2006). Furthermore, yeasts may remove oxygen from the surfaces of recently eaten feed, allowing the rumen to retain metabolic activity while remaining anaerobic. Another process relies on yeast's capacity to lower the rumen's redox potential, which allows strict anaerobic cellulolytic bacteria to grow more easily, accelerates their adhesion to fodder particles, and boosts the initial rate of cellulolysis (Newbold *et al*., 1996).

Furthermore, S. cerevisiae was able to compete with other starch-using bacteria for starch fermentation, limiting lactate buildup in the rumen, supplying growth factors such as organic acids or vitamins, and stimulating ruminal cellulolytic bacteria and LUB (Lynch and Martin, 2002).

Effect of DFM on Ruminant Performance

Pre ruminant calves (Young Calves)

Young calves vary from adult ruminants in that they may digest a large number of diet nutrients in their gut, but this comes with the danger of intestinal proliferation of harmful organisms, which can lead to diarrhoea and weight loss. The major purpose of DFM administration for dairy calves, according to Kung (2001), is to aid quick adaptation to solid feed by increasing the creation of ruminal and intestinal microbes while preventing the establishment of enteropathogens, which typically results in diarrhea.

According to Dicks and Botes (2010), Bifidobacteria generates acetic and lactic acids in a 3:1 ratio, and these acids are more efficient in the GIT for controlling Gram-negative infections and yeasts. Young calves were also infected with LAB to help them develop faster (Adams *et al*., 2008 & Frizzo *et al*., 2010). Adams *et al*. (2008) discovered that giving Propionibacterium jensenii 702 (PJ702) to calves improves weight increase throughout both the pre-weaning and weaning periods, with heavier calves' final weight. To create an intestinal imbalance, LAB fed newborn calves milk replacer and a huge amount of spray-dried whey powder. Calves fed LAB showed better daily growth, total feed intake, and starting diet consumption, as well as a lower fecal consistency score, indicating that diarrhea incidence was reduced under these conditions (Frizzo *et al*., 2010).

Feeding L. acidophilus 27SC to calves considerably reduced the incidence of diarrhoea, according to Abu-Tarboush *et al*. (1996). Lactobacillus and Streptococcus species are the most prevalent DFM species in young calves. There have been several reports of a reduction in diarrhea.

Adult ruminants

Early lactation high-producing cows would be the greatest candidates for such goods since they are in a negative energy balance and have diets high in fermentable carbs, which can lead to acidosis (Kung, 2006).

Cows may be subjected to many metabolic disorders during the three weeks prior to calving to three weeks after calving (i.e., transition periods) as a result of calving stress, changing diets to rapidly fermented carbohydrate sources, and lactation, according to the findings of Oetzel *et al*., (2007) and Chiquette *et al*., (2008). In this scenario, DFM was employed to improve dairy cow performance by boosting dry matter intake, milk output, milk protein content, and pre-and post-partum blood glucose and insulin levels (Nocek *et al*., 2003; Nocek and Kautz, 2006; and Oetzel *et al*.,2007).

Weiss *et al*., (2008) supplemented dairy cows with Propionibacterium P169 from 2 weeks before expected calving to 119 days in milk, finding reduced quantities of acetate and higher concentrations of propionate and butyrate. DFM did not affect plasma glucose or plasma-hydroxybutyrate levels, but it did result in greater plasma non-esterified fatty acid concentrations. Cows given Propionibacterium P169 consumed less dry matter, increasing their energy efficiency by 4.4 percent.

P. bryantii 25A treatment did not affect milk supply, but it did seem to increase milk fat due to increased acetate and butyrate concentrations in the rumen, according to Chiquette *et al*., (2008). When compared to control treatments, P. bryantii 25A lowered lactate concentration after 2–3 hours of feeding, indicating that it can prevent acidosis.

The use of exogenous cellulolytic bacteria as DFM to promote ruminal fermentation has been investigated. Ruminococcus flavefaciens NJ was given to non-lactating dairy cows who were fed either a high concentrate or a high forage diet regularly. When administered as part of a high concentration diet, R. flavefaciens NJ altered the abundance of other cellulolytic bacterial communities and enhanced sacco digestibility of hay in the rumen (Chiquette *et al.*, 2008).

DFM is critical in finishing beef cattle to minimize ruminal acidosis induced by widely used highly fermentable diets. DFM-fed beef cattle demonstrated enhanced growth, meat output, and feed efficiency (Ghorbani et al., 2002; Krehbiel et al., 2003). According to Krehbiel et al. (2003), giving bacterial DFM to feedlot cattle leads to a 2.5 to 5% increase in daily gain and a 2% improvement in feed efficiency, whereas DMI proved inconclusive.

DFM Practical Considerations

Powders, pastes, boluses, and capsules are only some of the direct-fed microbial products available. DFM can be blended with feed or injected into drinking water in specific situations. However, because interactions with chlorine, water temperature, minerals, flow rate, and antibiotics can alter the survival of DFM organisms, their usage in water must be carefully monitored (Kung, 2011).

Non-hydroscopic whey is frequently employed as a carrier for bacterial DFM and is an excellent growth medium. Bacterial DFM pastes are made using vegetable oil and inert gelling agents, whereas fungal DFM pastes are made with grain by-products as carriers. Some DFM is intended for one-time use, while others are intended to be fed regularly. Furthermore, bacterial DFM dosage levels have been reported to vary in investigations where L. acidophilus was fed at levels ranging from 106 to 1010 colony forming units (CFU) per animal per day (Kung, 2011). According to research by Hutchenson *et al.*, (1980), feeding more than 107 CFU per head per day may result in decreased nutritional absorption due to overpopulation in the gastrointestinal tract (GIT). Feeding a continuous high dosage of L.acidophilus to feeder calves (1010 CFU/head/day) did not affect body weight increase but lowered feed efficiency when compared to feeding a lower dose of 106 CFU/head/day, according to Orr *et al.* (1988).

Because many feeds are pelleted, the heat tolerance of DFM bacteria is critical. Heat kills the majority of yeast, Lactobacillus, Bifidobacterium, and Streptococcus. Bacilli are now employed in a variety of applications that require pelleting (Kung, 2011). Increased DFM inclusion can compensate for microbial loss during pelleting, however, this is not a recommended technique. According to Kung (2011), the viability of DFM goods has increased in recent years, although it is still important to follow storage guidelines. Products should be maintained free from dampness, excessive heat, and light, for example. If oxygen-sensitive microorganisms are to be created for commercial uses, further research on novel DFM products will need to address survivability.

CONCLUSIONS

In general, several feed additives are utilized in ruminant nutrition to increase feed quality (feed utilization), improve animal performance, and improve animal health. Microbes are a type of zootechnical feed additive used in the diet of ruminants. Ruminants and microorganisms have a symbiotic connection; the animals consume fodder to feed the bacteria, and the microbes eat to feed the ruminant. When ruminants ingest high fiber content feed, this aids them in obtaining efficient energy.

Direct-fed microbial refers to the methods of adding microorganisms to ruminant rations. DFM was not used by all microorganisms. DFM's routes of action in the rumen and gastrointestinal tract include the production of organic acids, antimicrobial production, competitive exclusion, immunological stimulation, enzyme activity, and toxic amine reduction. DFM influences the performance of both pre-ruminant and ruminant animals as a result of this. DFM can be given in a variety of ways, including as a supplement, in feed, or drinking water.

CONFLICTS OF INTEREST

The authors declare no conflict of interest.

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