



Special rumen microbiology

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ABSTRACT

Bacteria make up about half of the living organisms inside of the rumen. However, they do more than half of the work in the rumen. The bacteria work together. Some breakdown certain carbohydrates and proteins which are then used by others. Some require certain growth factors, such as B-vitamins, which are made by others. Some bacteria help to clean up the rumen of others' end products, such as hydrogen ions, which could otherwise accumulate and become toxic to other organisms. This is called "cross-feeding". Rumen microbiological studies based on laboratory culture have contributed greatly to our understanding of how feeds are transformed into the VFA and microbial cells that provide nutrients to the cow. Over the last 20 years, the limitations of culture-based microbiological studies of almost all microbial habitats become apparent.

Key words: microbiology, Rumen, Bacteria, pH, Fungi

Classification of Rumen Bacteria

- **Fiber-Digesting (or Cellulolytic) Bacteria**

The fiber-digesters are some of the "fussiest" bacteria in the rumen. They are very sensitive to acid. When a cow has acidosis (pH<6.0), the rumen produces a lower proportion of acetate to propionate because the fiber-digesters who primarily make acetate are not working well. Also, high levels of rumen available fat (generally over 5% of the diet) reduce the growth of the fiber-digesters. The exact reason for fat's negative effect on the fiber-digesters is not known. Some think that it reduces the microbe's ability to move nutrients into and out of its body. Others think that the fat coats fiber particles making it difficult for the fiber-digesting microbes to get in to do their work.

- **Starch and Sugar-Digesting (or Amylolytic) Bacteria**

Starch and sugar-digesters make up a significant part of the rumen's bacterial population. Generally, high-producing dairy cows are fed diets containing more than 30% starches and sugars, so these bacteria are greatly needed. Even if a cow is on an all-straw diet, the fiber-digesters still never account for more than 25% of the rumen bacterial population. Starch and sugar-digesters are still present, cross-feeding off of the fiber-digesters' byproducts.

- **Lactate-Using Bacteria:**

Includes: *Megasphaera elsdenii*, "The Rumen Maid". As mentioned above, some bacteria, such as *Streptococcus bovis*, produce a strong acid called lactic acid. *Megasphaera elsdenii* uses lactic acid to grow. This helps to clean up the rumen a bit and raise rumen pH, aiding the growth of the acid-intolerant fiber-digesters.

- **Protozoa**

As much as 50% of the microbial mass in the rumen can be made up of protozoa. However, their role, as compared to the rumen bacteria, is not as significant. The protozoa are actually predators to the bacteria in the rumen, they eat the bacteria for dinner! Protozoa are about 40 times the size of rumen bacteria. The rumen protozoa produce fermentation end-products similar those made by the bacteria, particularly acetate, butyrate, and hydrogen. Rumen methane bacteria actually attach and live on the surface of rumen protozoa for immediate access to hydrogen. Rumen protozoa eat large amounts of starch at one time and can store it in their bodies. This may help to slow down the production of acids that lower rumen pH, benefiting the rumen. Rumen protozoa multiply very slowly in the rumen over 15-24 hours – as opposed to the bacteria that may take as little as 13 minutes to multiply. For this reason, the rumen protozoa hide out in the slower moving fiber mat of the rumen so that they aren't washed out before they have a chance to multiply (Church et al., 1988). Low roughage diets reduce the retention of fiber in the rumen and may decrease the number of protozoa in a cow's rumen.

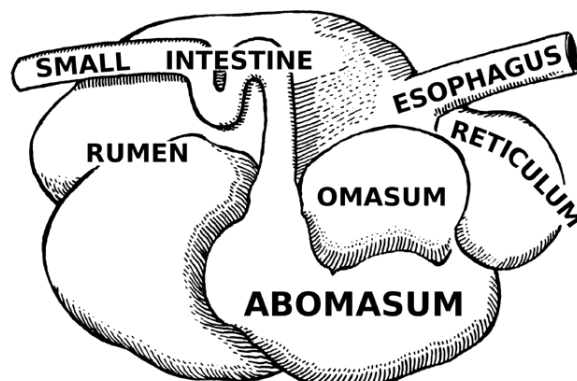
- **Rumen Fungi**

Fungi are known to exist in the rumen (up to 8% of the total mass) but they are poorly understood. They attach to feed particles and they reproduce very slowly. They may help out the fiber-digesting bacteria by doing some of the initial work of splitting fibrous material apart and making it more accessible for the bacteria. Higher numbers of fungi have been found in the rumens of cows fed very poorly digestible sub-tropical forages. Vertebrates lack the ability to hydrolyse the beta (Annison et al, 1998) glycosidic bond of plant cellulose due to the lack of the enzyme cellulase. Thus ruminants must completely depend on the microbial flora, present in the rumen or hindgut, to digest cellulose. Digestion of food in the rumen is primarily carried out by the rumen microflora, which contains dense populations of several species of bacteria, protozoa, sometimes yeasts and other fungi - it is estimated that 1 mL of rumen contains 10-50 billion bacteria, 1 million protozoa as well as several yeasts and fungi. As the environment inside a rumen is anaerobic, most of these microbial species are obligate or facultative anaerobes that can decompose complex plant material, such as cellulose, hemicellulose, starch, proteins. The hydrolysis of cellulose results in sugars, which are further fermented to acetate, lactate, propionate, butyrate, carbon dioxide and methane. During grazing, ruminants produce large amounts of saliva - estimates range from 100 to 150 litres of saliva per day for an adult cow (Huffman et al, 1949). The role of saliva is to provide ample fluid

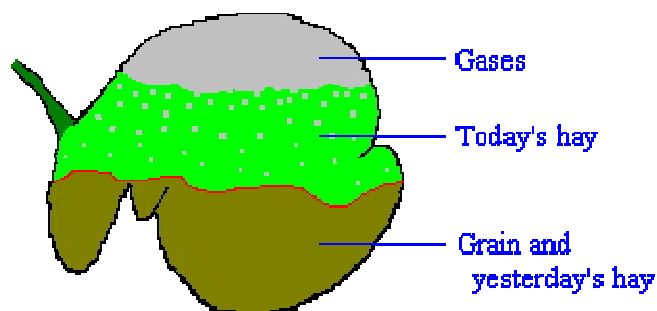
for rumen fermentation and to act as a buffering agent. Rumen fermentation produces large amounts of organic acids and thus maintaining the appropriate pH of rumen fluids is a critical factor in rumen fermentation.

- **Dynamics of Cranial Digestion**

Feed, water and saliva are delivered to the reticulorumen through the esophageal orifice. Heavy objects (grain, rocks, nails) fall into the reticulum, while lighter material (grass, hay) enters the rumen proper. Added to this mixture are voluminous quantities of gas produced during fermentation.



Ruminants produce prodigious quantities of saliva. Published estimates for adult cows are in the range of 100 to 150 liters of saliva per day! Aside from its normal lubricating qualities, saliva serves at least two very important functions in the ruminant: provision of fluid for the fermentation and alkaline buffering - saliva is rich in bicarbonate, which buffers the large quantity of acid produced in the rumen and is probably critical for maintenance of rumen pH (Tajima et al, 1999). All these materials within the rumen partition into three primary zones based on their specific gravity. Gas rises to fill the upper regions, grain and fluid-saturated roughage ("yesterday's hay") sink to the bottom, and newly arrived roughage floats in a middle layer.



The rate of flow of solid material through the rumen is quite slow and dependent on its size and density. Water flows through the rumen rapidly and appears to be critical in flushing particulate matter downstream. As fermentation proceeds, feedstuffs are reduced to smaller and smaller sizes and microbes constantly proliferate (Russell et al., 1988). Ruminal contractions constantly flush lighter solids back into the rumen. The smaller and more dense material tends to be pushed into the reticulum and cranial sac of the rumen, from which it is ejected with microbe-laden liquid through the reticulo-omasal orifice into the omasum. The function of the omasum is rather poorly understood. It may function to absorb residual

volatile fatty acids and bicarbonate(Hespell et al., 1987). The tendency is for fluid to pass rapidly through the omasal canal, but for particulate matter to be retained between omasal leaves. Periodic contractions of the omasum knocks flakes of material out of the leaves for passage into the abomasum. The abomasum is a true, glandular stomach which secretes acid and otherwise functions very similarly to the stomach of a monogastric. One fascinating specialization of this organ relates to its need to process large masses of bacteria. In contrast to the stomach of non-ruminants, the abomasum secretes lysozyme, an enzyme that efficiently breaks down bacterial cell walls. The processes described above apply to adult ruminants. For the first month or so of life, the ruminant is functionally a monogastric. The forestomachs are formed, but are not yet fully developed. If milk is introduced into such a rumen, it basically rots rather than being fermented. To avoid this problem in such young ruminants, suckling causes a reflex closure of muscular folds that form a channel from the esophageal orifice toward the omasum (the esophageal groove), shunting milk away from the rumen and straight toward the stomach where it can be curdled by rennin and eventually digested enzymatically(Hungate et al ., 1975). Fermentation is supported by a rich and dense collection of microbes. Each milliliter of rumen content contains roughly 10 to 50 billion bacteria, 1 million protozoa and variable numbers of yeasts and fungi(Thain et al 2001). The micrograph below, of sheep rumenal fluid, shows a Gulliver-like ciliated protozoon in the midst of thousands of bacteria (the small specks).



The environment of the rumen and large intestine is anaerobic and, as expected, almost all these microbes are anaerobes or facultative anaerobes. Fermentative microbes interact and support one another in a complex food web, with the waste products of some species serving as nutrients for other species. Fermentative bacteria representing many genera provide a comprehensive battery of digestive capabilities. These organisms are often classified by their substrate preferences or the end products they produce(Bauchop et al ., 1977). Although there is some specialization, many bacteria utilize multiple substrates. Some of the major groups, each of which contain multiple genera and species, are:

Cellulolytic (digest cellulose)

Hemicellulolytic (digest hemicellulose)

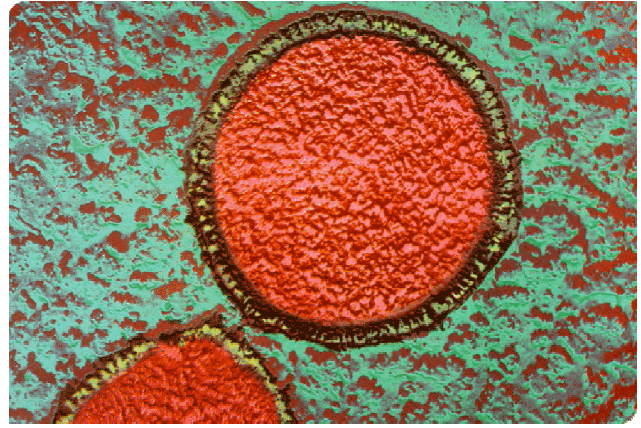
Amylolytic (digest starch)

Proteolytic (digest proteins)

Sugar utilizing (utilize monosaccharides and disaccharides)

Acid utilizing (utilize such substrates as lactic, succinic and malic acids)

Ammonia producers
Vitamin synthesizers
Methane producers



Protozoa, predominantly ciliates, appear to contribute substantially to the fermentation process (Macy et al., 1979). Several experiments have demonstrated that lambs and calves deprived of their ruminal protozoa show depressed growth rates and are relative "poor-doers" compared to controls with both bacteria and protozoa. In general, protozoa utilize the same set of substrates as bacteria and, as with bacteria, different populations of protozoa show distinctive substrate preferences. Many utilize simple sugars and some store ingested carbohydrate as glycogen. An interesting feature of some protozoa is their inability to regulate glycogen synthesis: when soluble carbohydrates are in abundance, they continue to store glycogen until they burst. An additional feature of protozoa is that many species consume bacteria, which is thought to perhaps play a role in limiting bacterial overgrowth (Prescott et al., 2005). The distribution of microbial species varies with diet. Some of this appears to reflect substrate availability; for example, populations of cellulolytic bugs are depressed in animals fed diets rich in grain. Environmental conditions in the fermentation vat also can have profound effects on the microbial flora. Rumen fluid normally has a pH between 6 and 7, but may fall if large amounts of soluble carbohydrate are consumed. If pH drops to about 5.5, protozoal populations become markedly depressed due to acid intolerance. More drastic lowering of rumen pH, as can occur with grain engorgement, can destroy many species and have serious consequences to the animal.

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