MiSACbriefings

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Fungi at work 2: Feeding farm animals

David Moore: *World of Fungi* www.davidmoore.org.uk/index.htm

Did you know that your beef, milk and dairy products all depend on fungi? Well, they do.

There are many animals, known as **ruminants**, that have stomachs with four compartments and depend on fungi in the gut to digest the grass they eat. The group includes cows, sheep, goats, deer, and even bison, yak, water buffalo, wildebeest, giraffes and various antelopes. There's even a group of 'pseudo-ruminants' that have three-chambered stomachs; this group includes the camel, llama, alpaca and vicuña.

Ruminant digestion

Such animals have a specialised multi-chambered stomach for the digestion of their exclusively vegetarian diet (Fig. 1). The first chamber the food enters is called the rumen, hence the name ruminant. This drastic adaptation of the alimentary canal is the 'evolutionary investment' that the animals have made to provide a population of microbes with a steady supply of freshly-cropped plant material and a warm safe habitat in which they can digest the supplied food matter. In return the animal gets a high-efficiency 'plant cell wall digester'.

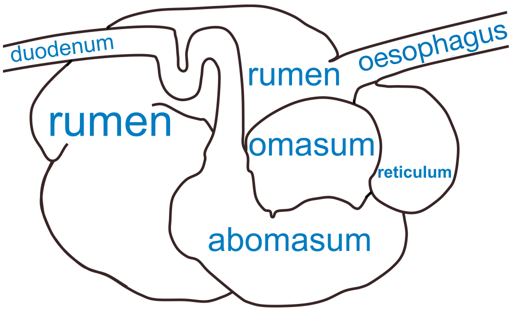

Fig. 1. The ruminant stomach is composed of 4 separate compartments. Food first passes into the rumen, then the reticulum, omasum and finally into the abomasum before entering the duodenum. In structure and function, the abomasum is most similar to the human 'monogastric' stomach.

Image © David Moore.

The ruminant we're most familiar with is the cow, and we all know that cows spend most of their time eating grass and hay. Plant cell walls contain cellulose, which is an excellent source of fibre in the diet of most animals. Fibre is important as it provides roughage which keeps the egestion of waste products regular. However, cows, like all animals, are not capable of producing enzymes to digest the cellulose. So, without help, they can't extract most of the nutrients the grass contains.

Anaerobic chytrids

Ruminants overcome this problem by having special fungi in the rumen called ***chytrids***, more generally referred to as 'rumen fungi' (Fig. 2). These fungi are **anaerobic**; they can survive without oxygen, digesting plant cell walls by making specific enzymes called cellulases.

The rumen acts like a large fermenter; the grass is stored there whilst the fungal enzymes from the chytrids break down the cellulose. The chytrid fungi also produce a wide range of digestive enzymes with broad substrate specificity. These transform the core structural polymers of plant cell walls into a variety of simple molecules: oligosaccharides, disaccharides, monosaccharides, amino acids, carboxylic acids and fatty acids. The microbes ferment these metabolites to release energy and other products that contribute to their cell growth, reproduction and microbial population growth.

Fig. 2. A phase-contrast microphotograph of a thallus of the rumen chytrid, *Neocallimastix* sp., showing the general morphology of chytrids. Scale bar = 40 µm.

The most obvious morphological feature is the thallus, in which most of the cytoplasm resides and from which branching rhizoids emerge. These anchor the fungus in its substratum and secrete digestive enzymes; they do not contain nuclei. The sac-like thallus is converted into a sporangium during reproduction, its protoplasm becoming internally divided to produce zoospores.

Image © Tony Trinci; reproduced with permission from the textbook *21st Century Guidebook to Fungi* (Cambridge University Press, 2020).

Anaerobic fungi don't have mitochondria; instead, they possess **hydrogenosomes.** These convert carboxylic acids into CO_2 and hydrogen with the release of energy in the form of ATP, using the enzymes pyruvate oxidoreductase and hydrogenase. The carboxylic acid substrates (formate, acetate, lactate, with some ethanol) are produced from hexose and pentose sugars in what is known as a **mixed-acid fermentation**.

After the plant material is processed in the rumen, it is brought back up into the mouth of the cow. This material is now called 'cud' and the cow chews it again to grind it further.

When it is swallowed for the second time, it passes through the next three chambers of the stomach where further digestion occurs.

Mutual benefits

The relationship between chytrids and ruminants is said to be **symbiotic** (or mutualistic). This means that both the fungi and the cow benefit from having the other present. In this case the cow benefits because plant material the animal can't degrade is digested and turned into microbial biomass which the cow can absorb. In return, the microbes reproduce using nutrients from the cow's food and live out their lives in the protective cow's rumen. All this microbial growth is eventually passed through the host's stomach and intestines where digestion by the cow allows it to become the animal's source of nutrients and energy (Fig. 3).

Microorganisms within the rumen form a complex ecosystem. This consists of archaea (a domain of single-celled organisms), many bacteria, some of which compete with fungi to digest xylan in cereal straw and ciliate protozoa. The ciliates ingest fungal zoospores as well as bacteria. Overall degradation of plant material is greater when the fungi, archaea and bacteria are working together than when they are working individually.

The relationship between the archaeal methanogenic organisms and chytrid fungi in the rumen is most important. Together they carry out a more-efficient fermentation process, which releases a higher biomass yield from the food, in the form of a larger microbial community, giving greater benefit to the host animal. Methanogenic archaea produce ATP for growth by using CO_2 as an electron acceptor to convert the excess H_2 into methane.

Hydrogen is released by the activity of hydrogenase in the fungal hydrogenosome, but even low levels of free hydrogen inhibit the action of hydrogenase. So a crucial contribution of the synergistic methanogenic archaea is to remove the hydrogen from the rumen and prevent hydrogenase inhibition. The result of archaeal methanogenic activity is an increase in carbon flow through the chytrids and, therefore, an overall increase in microbe productivity in the animal.

But the activity of these archaeal organisms also results in the release of enormous quantities of the greenhouse-gas methane into the atmosphere from the global herd of 1.8×10^9 cattle and the global sheep flock of about 10^9 sheep. This is one reason for the recent '*eat less meat to save the planet*' campaign, although another reason is that our farm livestock provides just 18% of calories for human nutrition but takes up 83% of farmland.

We now have a tripartite mutualism: ruminant-chytrid-methanogen. If we add the farmer who manages the pastureland for his/her cattle, the butcher who prepares the meat, the restaurateur who turns the meat into a meal and the diner who eats that meal, the range of mutual dependencies increases even further.

True grazing animals appeared late in the Miocene epoch (around 20 million years ago) following the emergence of grassland-forming grasses of the plant family Poaceae. Before that, plant-eating mammals during the late Cretaceous and early Palaeocene (say, 80 million years ago) were physically-small fruit-eaters (frugivores, also called fructivores). Mammals did not become herbivore browsers until the middle Palaeocene (60 million years ago) and did not become major components of the fauna until the late Eocene (40 million years ago). The Eocene climate was humid and tropical, and its forests probably favoured browsers and frugivores, so it is thought that the earliest herbivores were large, ground-dwelling mammals that evolved from large frugivores. It is also argued that the first to develop were animals with a simple, single-chambered stomach; plant wall cellulose was digested with the aid of symbiotic microbes in the large intestine and caecum ('hind-gut fermentation'). Present-day animals of this sort are the horse, zebra, rhinoceros, elephant, rabbit and all rodents.

Mutual evolution

Ruminants (with 'fore-gut fermentation') emerged after this initial adaptation of the hind-gut, when grass-dominated ecosystems, including steppes, temperate grasslands, and tropical/subtropical savannas, began to play a more central role. They occupy about a quarter of the Earth's land surface today. These ecosystems evolved during the Cenozoic era, which extends from 66 million years ago to the present day. Grasses initially evolved 60 million years ago; the first to appear used the C3 photosynthetic pathway, which is the typical one used by most plants.

Fig. 3. One cow; containing an average of about 1.5×10^{14} anaerobic chytrid fungi. Image © David Moore.

Grasses that dominate the semi-arid savanna are C4 plants that photosynthesise more efficiently in the higher temperatures and brighter sunlight because they use water more effectively and have adaptations to reduce photorespiration.

There is a good argument for the evolution of ruminants being driven by the development and expansion of savanna and steppe grasslands in Africa and Eurasia during the Oligocene and throughout the Tertiary. As they became the dominant vegetation, the emphasis in herbivore evolution would be to increase the efficiency of the fermentation of their more-fibrous plant material.

Changes in the environment drive major evolutionary events and, in this case, major changes in abundance and diversity of the entire family of ruminant mammals were caused by dramatic climatic changes affecting the entire ecosystem. Emergence of the genus *Homo* in the Pliocene (5 to 2.5 million years ago) of East Africa also appears to be broadly correlated with these same climatic changes.

The Poaceae family of plants that makes up the grasslands that currently represent 25% of the vegetation cover of planet Earth is the most important of all plant families to humans as it includes our staple **food grains**.

Ruminant mammals became the most abundant and successful order of current and fossil herbivores, with about 190 species living today. Humans found their main food animals and work animals among the ruminants and their relatives.

And all these evolutionary successes are owed to the symbiotic relationship of ruminant mammals with their rumen chytrids.

Something else for which we can thank fungi!

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