Lecture 2

Rumen Physiology and Rumination

The rumen is a fermentation vat *par excellence*, providing an anaerobic environment, constant temperature and pH, and good mixing.

Well-masticated substrates are delivered through the oesophagus on a regular schedule, and fermentation products are either absorbed in the rumen itself or flow out for further digestion and absorption downstream.

Ruminants evolved to consume and subsist on roughages - grasses and shrubs built predominantly of cellulose. Although some ruminants, feedlot steers for example, are fed large quantities of grain.

Feed, water and saliva are delivered to the reticulo-rumen 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:

(a) Provision of fluid for the fermentation vat

(b) 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.

All these materials within the rumen are partitioned 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

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.

Reticuloruminal Motility

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 where it is ejected with microbe-laden liquid through the reticulo-omasal orifice into the omasum.

An orderly pattern of ruminal motility is initiated early in life and, except for temporary periods of disruption, persists for the lifetime of the animal. These movements serve to mix the ingesta, aid in eructation of gas, and propel fluid and fermented foodstuffs into the omasum.

If motility is suppressed for a significant length of time, ruminal impaction may result. A cycle of contractions occurs 1 to 3 times per minute.

The highest frequency is seen during feeding and the lowest when the animal is resting.

Two types of contractions are identified:

Primary contractions originate in the reticulum and pass caudally around the rumen. This process involves a wave of contraction followed by a wave of relaxation, so as parts of the rumen are contracting, other sacs are dilating.

Secondary contractions occur in only parts of the rumen and are usually associated with eructation.

Conditions inside the rumen can significantly affect motility. If, for example, ruminal contents become very acidic (as occurs in grain engorgement), motility will essentially cease. Also, the type of diet influences motility: animals on a high roughage diet have a higher frequency of contractions than those on a diet rich in concentrates.

Rumination and Eructation

Ruminants are well known for "cud chewing". Rumination is regurgitation of ingesta from the reticulum, followed by remastication and reswallowing. It provides for effective mechanical breakdown of roughage and thereby increases substrate surface area to fermentative microbes.

Regurgitation is initiated with a reticular contraction distinct from the primary contraction. This contraction, in conjunction with relaxation of the distal oesophageal sphincter, allows a bolus of ingesta to enter the esophagus. The bolus is carried into the mouth by reverse peristalsis.

Fermentation in the rumen generates enormous, even frightening, quantities of gas, about 30-50 liters per hour in adult cattle and about 5 liters per hour in a sheep or goat.

Eructation or belching is how ruminants continually get rid of fermentation gases.

The fluid in the bolus is squeezed out with the tongue and reswallowed, and the bolus itself is remasticated, then reswallowed.

Rumination occurs predominantly when the animal is resting and not eating, but that is a considerable fraction of the animal's lifespan. An eructation is associated with almost every secondary ruminal contraction.

Eructated gas travels up the oesophagus at 160 to 225 cm per second and, interestingly, a majority is actually first inspired into the lungs, then expired.

Anything that interferes with eructation is life threatening to the ruminant because the expanding rumen rapidly interferes with breathing. Animals suffering bloat die from asphyxiation

The processes described above apply to adult ruminants. For the first month or so of life, the ruminant is functionally a monogastric. The fore stomachs 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). This helps in shunting milk away from the rumen and straight toward the stomach where it can be curdled by rennin and eventually digested enzymatically.

Microbiology of the rumen

The rumen is an organ where microbial populations collaborate to digest cellulose and other polysaccharides producing carbon dioxide, methane and organic acids.

Microbial content of rumen comprises of:

Fungi - digest lignin and cellulose

Bacteria - a thick paste of 10^{10} - 10^{11} cells/mL (compare to stationary phase *E. coli* cultures that contain ~ 4 x 10^8 cells/mL)

Protozoa - ~ 10^{6} /mL, mostly ciliates that prey on bacteria and ferment some carbohydrates

Protozoa, predominantly ciliates, appear to contribute substantially to the fermentation process.

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.

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, protozoa 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 on the animal.

Fermentation is supported by a rich and dense collection of microbes. 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.

Although there is some specialization, many bacteria utilize multiple substrates.

Some of the major groups of bacteria, 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

Ingested food first enters the rumen (pH 6.5, temperature of 30° C) where it is microbially digested for ~ 9 hours. The gaseous products of the microbial degradation are expelled from the animal (eructation), and the material from the rumen, called the cud, is regurgitated.

This regurgitated mixture of microorganisms and partially digested materials then travels through the omasum (pH \sim 2), the abomasum, and the rest of the digestive tract, for further digestion.

Biochemistry of the rumen

Biochemistry is the study of the chemical processes in living organism. It deals with the structure and function of cellular components, such as proteins, carbohydrates, lipids, nucleic

acids, and other biomolecules. *Or Biochemistry* is the study of the chemical substances and vital processes occurring in living organisms.

Use of carbohydrates as an energy source

Glucose is the major energy source in most life forms. For instance, polysaccharides are broken down into their monomers (glycogen phosphorylase removes glucose residues from glycogen). Disaccharides like lactose or sucrose are cleaved into their two component monosaccharide.

Glycolysis (anaerobic)

Glucose is mainly metabolized by a very important and ancient ten-step pathway called glycolysis, the net result of which is to break down one molecule of glucose into two molecules of pyruvate; this also produces a net two molecules of ATP, the energy currency of cells, along with two reducing equivalents in the form of converting NAD+ to NADH.

This does not require oxygen; if no oxygen is available (or the cell cannot use oxygen), the NAD is restored by converting the pyruvate to lactate/lactic acid (e. g. in humans) or to ethanol plus carbon dioxide (e. g. in yeast).

Other monosaccharides like galactose and fructose can be converted into intermediates of the glycolytic pathway.

IN SUMMARY :

1. Cellulose and starch are broken down to glucose monomers.

2. Fermentative bacteria produce organic acids from glucose.

3. The non-volatile acids (e.g. lactate, succinate) produced from fermentation are converted into volatile acids (e.g. acetate). These enter the bloodstream of the host mammal from the rumen.

4. Methanogens utilize formate ($H_2C=O$) or carbon dioxide (CO_2), together with molecular hydrogen (H_2), to produce methane (CH_4).

5. The eructate consists of methane (35%) and carbon dioxide (65%). Daily production of gas from a cow is about 100 to 150 L; a well-fed dairy cow can generate as much as 500 L of gas daily!

Summary of protein utilization

Sources of Rumen Nitrogen

Feed

Protein nitrogen: Protein supplements (SBM, CSM, grains, forages, silages etc)

Non protein nitrogen (NPN): Usually means urea. However, from 5% of N in grains to 50% of N in silage and immature forages can be NPN

Endogenous (recycled) N

Saliva Rumen wall

Protein Leaving the Rumen

Microbial protein

Escape protein (also called "bypass" protein)

These enter abomasum & small intestine and are digested by proteolytic enzymes similar to non ruminants