Ruminant Digestion

14

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Abstract

This chapter reviews the ruminant digestion with a special emphasis on the mechanical factors, gastro-intestinal tract structure, and nutrient digestibility. Ruminants possess large compartmental gastro-intestinal tract viz. rumen, reticulum, omasum, abomasum, and intestine, which favors handling large amounts of fibrous plant materials. Among the four-rumen compartments, abomasum occupies large space in newborn ruminants; however, the growth rate of the rumen and reticulum will be faster compared to abomasum as the age advances. In adult ruminants, the rumen harbors vast range of microbes enabling microbial fermentation of ingesta before exposing to gastric juices of abomasum. Ruminant digestion involves mechanical processing of feed stuff. Among various mechanical factors, rumination aids in complete digestion of feed stuff and include regurgitation, remastication, reinsalivation, and redeglutition. The rumen microbiota, consisting of bacteria, protozoa, fungi, and archea degrade the ingested fiber-based diets and aids in nutrient fermentation. The fermentation of complex carbohydrates produces short-chain fatty acids (acetate, propionate, and butyrate), isoacids (valeric, isovaleric, isobutyric, and 2-methylbutyric acids), and gases such as CO_2, CH_4, and H_2. About 70% of the ruminant animal's energy supply will be met by the produced volatile fatty acids. High fiber diet induces the production of acetate while the starch and sugars yield propionate as end product. Milk fat synthesis requires acetate and hence, low fiber diets lead to milk fat depression. Similarly, propionate contributes to most of the energy required for weight gain and lactose production. Rumen pH is an important factor to be considered; low pH suppresses the growth of certain bacteria sensitive to pH-causing rumen dysfunction and subacute rumen acidosis. The protein metabolism in ruminants depends upon the ability of rumen microbes utilizing ammonia. More than 80% of the rumen bacteria utilizes ammonia as nitrogen source for growth and yields microbial protein. For every 1 kg organic matter digested, the microbial yield ranges from 90 to 230 g, which is sufficient for growth and production to certain extent. Fat digestion in ruminants is unique in that the ruminal bacteria split the fatty acids and sugars from glycerol backbone through lipolysis. The metabolism of lipids by rumen microbes involves a four-stepped process viz. hydrolysis of esterified fatty acids, biohydrogenation of unsaturated fatty acids, lipid biosynthesis in the rumen, and metabolism of phytal to phytanic acid. Further, incomplete biohydrogenation generally produces conjugated linoleic acids (CLA), which are proven to benefit human health.

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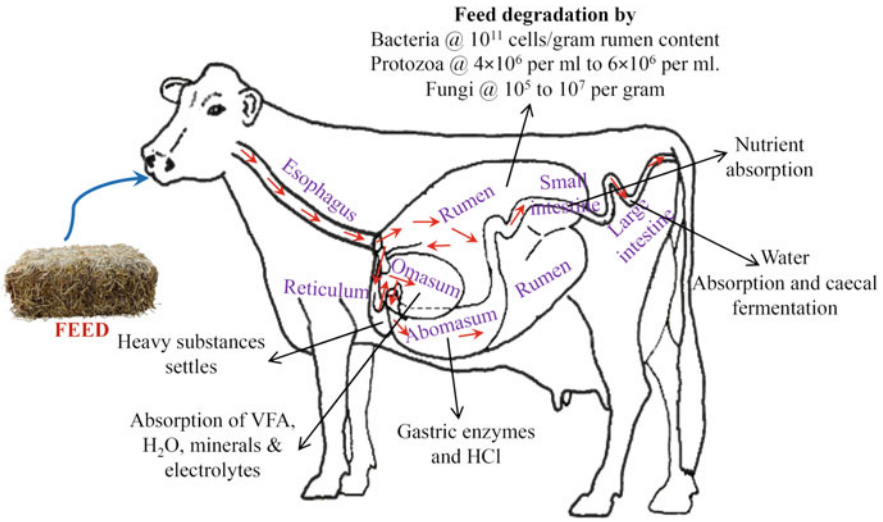
Graphical Abstract



Description of the graphic: The digestion in ruminants is fermentative, i.e., the nutrients are subjected to fermentation in a specialized compartment of stomach is called rumen. The specialized environments in the rumen favors the growth of protozoa, bacteria, and fungi required for fermentative digestion. The motility of the rumen facilitates continuous mixing of the ruminal content and eructation of gases. The partially degraded feed undergoes regurgitation and the cud reaches ventral rumen, followed by reticulum, omasum, and abomasum. The carbohydrates are hydrolyzed and converted to volatile fatty acids and utilized by the body after absorption. The dietary proteins are converted to microbial crude proteins in the rumen and digested in the abomasum. Abomasum acts as true stomach and favors enzymatic digestion. Further digestion takes place in small intestine, where absorption of nutrients occurs through villi. Ultimately, the undigested feed will be excreted as feces.

Keywords

Ruminant · Digestive system · Rumen fermentation · Subacute rumen acidosis

Learning Objectives

- The structure of rumen and its environment
- Mechanical factors involved in the ruminant digestion
- Significance of ruminal microbes in modulating nutrient digestibility
- Fermentative digestion of nutrients and utilization of fermentation end products

14.1 Overview of Ruminants' Stomach

Ruminants are the even-toed ungulate herbivorous mammals capable of regurgitating food from their stomach for remasticating. They possess large compartmental gastrointestinal tract (rumen, reticulum, omasum, abomasum, and intestine), which favors handling of large amounts of fibrous plant materials. The rumen harbors vast range of microbes enabling microbial fermentation of ingesta before exposing to gastric juices in abomasum. The rumen microbiota, consisting of bacteria, protozoa, fungi, and archea degrade the ingested fiber-based diets. The mechanical activity of rumen, reticulum, and omasum supports this degradation. Esophagus opens into the rumen through cardia. The rumen is composed of cranial sac, ventral sac, and ventral blind sac, which are freely communicated with one another. The rumen wall is covered with many finger-like projections known as papillae with 5 mm in length and 3 mm wide in cattle.

In ruminants, abomasum is analogous to monogastric stomach and hence called as true stomach. Similar to the monogastric's stomach, abomasum secretes gastric enzymes and HCl. Among the four-rumen compartments, abomasum occupies large space in newborn ruminants. However, the growth rate of the rumen and reticulum will be faster compared to abomasum as the age advances. After completing the growth, rumen and reticulum, omasum, and abomasum occupies 69%, 8%, and 23% of the stomach portion. The pictorial representation of bovine stomach development from birth to maturity is presented in Fig. 14.1.

The omasum component is not well developed in small ruminants and is completely absent in the animals belonging to the suborder tylopoda (Camel and Llama). Esophageal groove or reticular groove, a gutter like invagination, extends from the cardia to reticulo-omasal orifice. The stimulation of sensory receptors in the pharynx and mouth causes closure of reticular groove to bypass milk directly from esophagus into

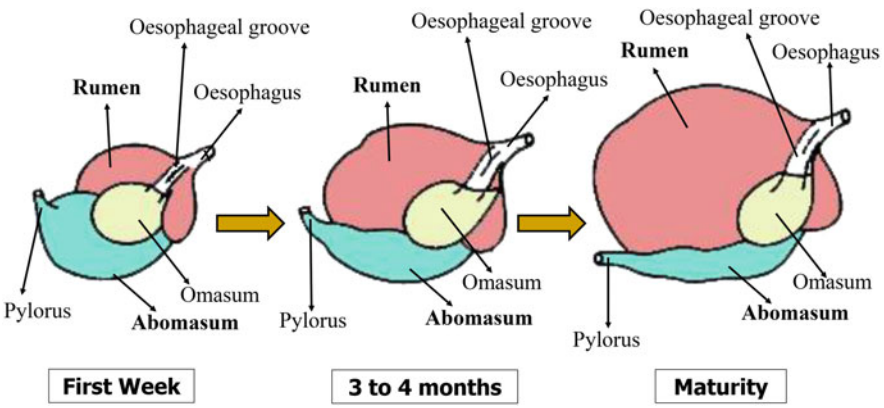


Fig. 14.1 Bovine stomach development from birth to maturity. [The size of rumen increases as the age advances and reaches maximum size at maturity]

reticulo-omasal orifice avoiding rumen and reticulum. Closure of esophageal groove is mediated by psychological responses, behavioral patterns, and chemicals such as sodium chloride, sodium bi-carbonate, copper sulfate, and sugar solutions. Among these chemicals, copper sulfate is less effective in calves and older ruminants and more effective in sheep.

14.2 Features of Digestion in Ruminants

Ruminants' gastro-intestinal tract holds numerous colonies of microorganisms. The type of microbes depends upon the diet and modifies accordingly as the age advances. A complex interaction exists between the host animal and variety of microbes. The gastro-intestinal tract of ruminants is unable to digest cellulose due to the lack of the degrading enzymes, hence completely relies on metabolic activities of gut microbes in utilizing the complex carbohydrate-based feed such as roughage. The fibrous materials retain in the gut for longer period to support the slow fermenting property of microbes. Among the fibrous particles, larger ones are retained at the reticulo-omasal orifice for mechanical digestion. Microbial fermentation produces volatile fatty acids, mainly acetate, propionate, and butyrate, which are of high value to the host ruminant system. Nearly 70% of the energy supply will be met by the produced volatile fatty acids. The ruminal microbes can also use non-protein nitrogen compounds such as ammonia to synthesize amino acids. The host proteolytic enzymes later digest the synthesized microbial protein. The gases produced by fermentation viz. CO_2 and CH_4 are expelled by eructation. In ruminant digestive system, saliva plays an indispensable role for buffering action against VFAs and monitoring the rumen pH. Therefore, physical effective NDF, a fraction of fiber that stimulates chewing activity and saliva production, is an important parameter to be considered while feeding the animals. Fermentation also allows detoxification of toxic substances before reaching small intestine.

14.3 Mechanical Factors Involved in Ruminants' Digestion

The mechanical factors involved in ruminant's digestion include mastication, deglutition, rumination, and eructation (Fig. 14.2). Rumination is a procedure of retrieving the food from upper part of rumen to the mouth for mastication. Rumination aids in complete digestion of feed stuff and include regurgitation, remastication, reinsalivation, and redeglutition.

14.3.1 Regurgitation

Heavy substances such as grains, rocks, or nails settle into the reticulum after ingestion, whereas lighter substances (roughage) enter the rumen. The saliva and fermentative gases accompany the lighter substances. Based on the specific gravity, the ruminal substances partitions into three zones viz. gas (upper), lighter roughage pieces (middle), and grain and fluid-saturated roughage (Bottom) (Fig. 14.3). Freshly eaten forages whose particle size is too great to be suspended in the rumen fluid for extensive maceration are not regurgitated immediately. The fermentation led constantly proliferating microbes reduce the feedstuff into micro-sized pieces. The continuous ruminal contractions push lighter roughage pieces into middle layer and denser substances into cranial sac of rumen and reticulum. The elevated soft palate closes glottis and the inspiratory effort with tongue drops intra-esphageal and intrathoracic pressure. The negative intrathoracic pressure opens cardia and caudal esophageal sphincters and forces the cud from middle layer of rumen into esophagus. The retrograde peristaltic wave originated from the terminal part of the thoracic esophagus carries cud to oral cavity. The lighter roughage pieces return to the mouth in cud form causing remastication. The regurgitated cud consists of small particulate matter highly mixed with liquid, which sinks to bottom layer within the rumen.

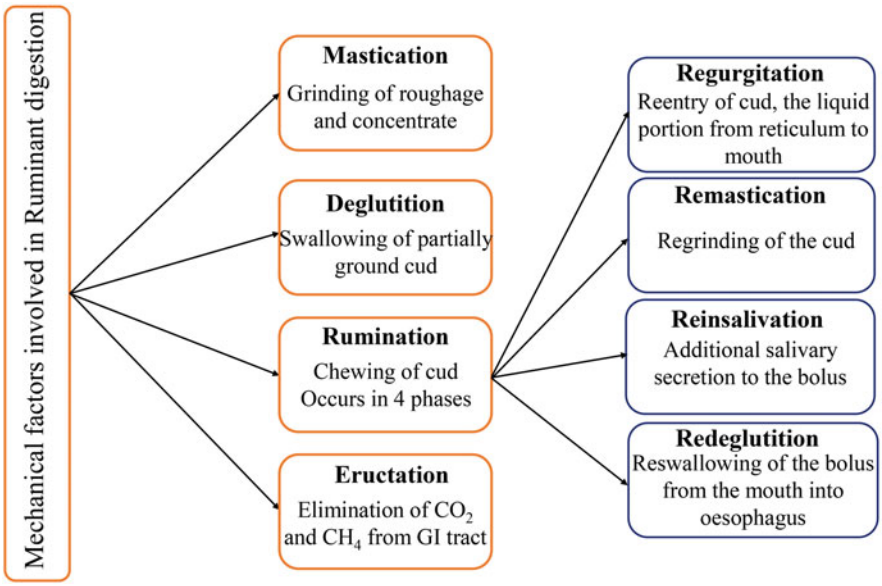


Fig. 14.2 Mechanical factors involved in ruminant digestion. [Mastication, deglutition, rumination, and eructation are the mechanical factors of ruminant digestion. Rumination in turn could be divided into four steps viz. regurgitation, remastication, reinsalivation, redeglutition]

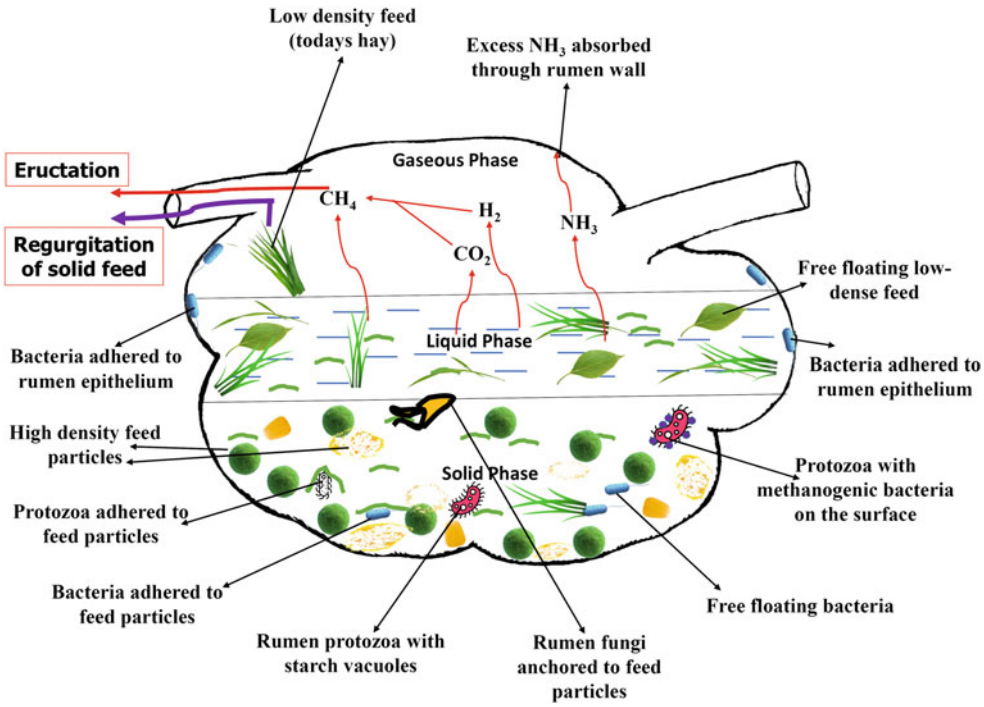


Fig. 14.3 Rumen ecology with the three partitions. [Rumen consists of complex ecosystem with diverse microorganisms such as bacteria, protozoa, and fungi. The gaseous phase of rumen contains CH_4 and NH_3, liquid phase comprises of free-floating low-dense feed, and the solid phase consists of high dense feed particles]

14.3.2 Remastication

After regurgitation, the rigid portion of cud is masticated for about 30–70 s. Entire cycle is repeated with an interval of 2–4 s between the remastication of two boluses. While chewing, the liquid portion is swallowed spontaneously. Remastication reduces the particle size and provides more surface area for the attack of microorganisms. Optimum remastication time is essential to lessen the risk of acidosis and improve the fiber degradation within the rumen. The chewing time is related to physically effective NDF portion of the diet.

14.3.3 Reinsalivation

As the cud is remasticated, parotid glands secrete saliva, facilitating reswallowing of the chewed cud. The remastication of the solid cud is accompanied by reinsalivation and redeglutition. Saliva is a significant

buffering agent for rumen and hence reinsalivation plays a pivot role in maintaining optimum rumen pH of 6–7.

14.3.4 Redeglutition

Redeglutition is an act of reswallowing the cud. The reswallowed cud directly reaches rumen for increased microbial action on the complex carbohydrates including cellulose and hemi-cellulose.

Several intrinsic factors such as sex, age, and body size, and extrinsic factors including diet, time, and season affect the time of rumination. On an average, the rumination time for cattle is 10 h per day on complete hay-based diet. Grinding the roughage may decrease rumination time to 3 h per day.

14.3.5 Eructation

Eructation is the expulsion of fermentation gases like carbon dioxide and methane accumulated in the rumen.

14.4 Rumen Fermentation

The distinctive feature of ruminant digestive system is the fermentative digestion of feed materials through microbes, which occurs in rumen and reticulum. Besides, the fermentative digestion of feed can also be seen in pseudo-ruminants such as llamas, camels, and hippopotamus. The major microbes in rumen include ciliate protozoa, non-spore forming anaerobic bacteria, and anaerobic fungi followed by few facultative anaerobic bacteria. About 3.6% of the strained rumen liquor is composed of microbes with equal weights of bacteria and ciliate protozoa. The amount of rumen fungi is insignificant, but their activity is of huge importance. Both the bacteria and protozoa grow on the substrates of structural and non-structural carbohydrates, which are hydrolyzed by microbial enzymes. The gases generated by fermentation (carbon dioxide, methane, and traces of hydrogen) maintains anaerobic environment. The little amount of oxygen released into rumen is utilized by facultative anaerobes to maintain anaerobic condition.

14.4.1 Rumen as Microbial Habitat

Rumen provides congenial environment for the growth and multiplication of microbes. The rumen maintains a constant temperature of 40 °C.The HCO_3 andHPO_4 buffers of saliva provide a constant pH of 6–7. The saliva secretion also provides aqueous environment, thereby supplying substrates for continuous microbial activity. The primary contractions of rumino-reticulum aids in proper mixing of ruminal contents and the secondary contractions cause eructation (Fig. 14.4).

Rumen microbes use the host ruminants' feed stuff constituting cellulose, hemi-cellulose, pectin, soluble sugars, and starch to synthesize their energy for growth. Consequently, the fermentation produces acetic, butyric, propionic, and lactic acids along with gases such as H_2, CO_2, and methane. The fermentative end products act as inhibitors of fermentation and are removed continuously from the rumen. Although the calves are devoid of rumen microbes, they later attain the microbial population because of the dams' rumination ability. Rumination aids in regurgitating feed and rumen contents back into the mouth thus salivating and contaminating the feed consumed by the young calves. The rumen microbes could also be passed directly to calves during grooming.

Abomasum is analogous to monogastric stomach and causes the hydrolysis of protein of both dietary and microbial

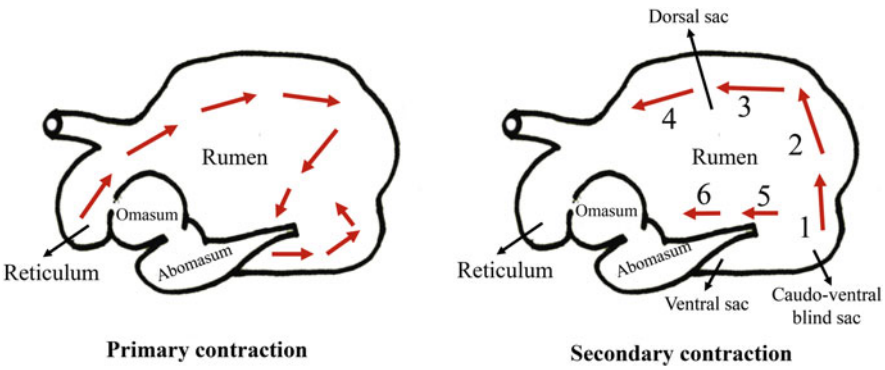


Fig. 14.4 Primary and secondary contractions. [The primary contraction occurs every 60 s and includes two contractions of the reticulum, which reaches the rumen. It leads to the ingesta flow from reticulum to cranial ruminal sac and later to ventral sac. The secondary contraction causes eructation. It leads to the ingesta flow from the caudo-ventral blind sac to the dorsal blind sac followed by dorsal sac (causes eructation) and ventral sac in sequence]

origin, which is later absorbed in small intestine. The HCl and gastrin secretion are stimulated by a rise in abomasal pH and short-chain fatty acid levels. Gastric secretion occurs both from fundic and pyloric glands with the former as a major secretory source. The secretion from fundus region contains pepsin and HCl with pH close to 1.0, while that from pyloric glands is slightly alkaline with slight peptic activity.

14.4.1.1 Rumen Bacteria

Among the diverse microorganisms of rumen, bacteria are the predominant microbes contributing to nitrogen and carbohydrate metabolism through fermentation (Table 14.1). The rumen content comprises as high as 10^{11} cells per gram of rumen content with more than 200 species. Although the total volume of small bacteria is same as ciliate protozoa, the metabolic activity of bacteria is far greater than protozoa, presumably because of the greater surface area. The rumen bacteria metabolize ingested feed material into volatile fatty acids, vitamins, and microbial biomass, which are later utilized by the host tissue. Based on the environmental existence, bacteria inhabiting the rumen have been classified into five groups. They include free-living bacteria associated with rumen liquid phase, bacteria loosely associated with feed particles, bacteria firmly adhered to feed particles, bacteria associated with rumen epithelium, bacteria attached to the

Table 14.1 The types, examples, substrates, and fermentative end products of rumen bacterial species

Type	Example	Substrate	Fermentative end product
Cellulolytic species	*Fibrobacter succinogenes*	Cellulose	Acetate, Formate, and Succinate
	Butyrivibrio fibrisolvens	Cellulose	Acetate, Butyrate, Formate, Lactate, H_2, CO_2
	Ruminococcus albus	Cellulose	Acetate, Formate, H_2, CO_2
	Clostridium lochheadii	Cellulose	Acetate, Formate, Butyrate, H_2, CO_2
Hemicellulolytic species	*Butyrivibrio fibrisolvens*	Xylans	Acetate, Butyrate, Formate, Lactate, H_2, CO_2
	Ruminococcus sp.	Xylans	Acetate, Formate, H_2, CO_2
	Bacteroides ruminicola	Xylans	Acetate, Formate, Succinate, CO_2
Pectinolytic Species	*Butyrivibrio fibrisolvens*	Pectin	Acetate, Butyrate, Formate, Lactate, H_2, CO_2
	Bacteroides ruminicola	Pectin	Acetate, Formate, Succinate, CO_2
	Succinivibrio dextrinosolvens	Pectin	Acetate, Succinate
Amylolytic species	*Bacteroides amylophilus*	Maltose	Formate, acetate, Succinate
	Selenomonas ruminantium	Oligosaccharides	Formate, acetate, Succinate
	Succinomonas amylolytica	Oligosaccharides	Acetate, Propionate, Succinate
	Streptococcus bovis	Starch substrates	Lactate at pH less than 5.5 Acetate, Formate, Ethanol at pH more than 6.0
Ureolytic species	*Succinivibrio dextrisolvens*	Urea with sugar or starch source	Acetate, Succinate
	Selenomonas sp.	Urea with sugar or starch source	Formate, acetate, Succinate
	Butyrivibrio sp.	Urea with sugar or starch source	Acetate, Butyrate, Formate, Lactate, H_2, CO_2
	Bacteroides ruminicola	Urea with sugar or starch source	Acetate, Formate, Succinate, CO_2
Lipolytic species	*Anaerovibrio lipolytica*	Triacylglycerols	Free fatty acids, Glycerol
	Micrococcus sp.	Triacylglycerols	Free fatty acids, Glycerol
Lactate utilizing sps.	*Selenomonas lactilytica* *Selenomonas ruminantium*	Lactic acid	Acetate, Succinate
	Megasphaera elsdenii	Lactic acid	Acetate, Propionate, Butyrate
Methane-producing species	*Methanobrevibacter ruminantium*	Cellulose, hemi-cellulose	H_2, CO_2, Formate, and the ultimate end product CH_4
	Methanobacterium formicicum	Cellulose or hemi-cellulose	H_2, CO_2, Formate, and the ultimate end product CH_4
	Methanomicrobium mobile	Cellulose or hemi-cellulose	H_2, CO_2, Formate, and the ultimate end product CH_4
Sugar-utilizing species	*Treponema bryantii*	Sugar	Acetate, Propionate
	Lactobacillus sp.	Sugar	Lactic acid
Proteolytic species	*Bacteroides amylophilus*	Proteins	Acetate, Propionate, Butyrate, CO_2, NH_3
	B. ruminicola	Proteins	Acetate, Propionate, Butyrate, CO_2, NH_3
	Butyrivibrio fibrisolvens	Proteins	Acetate, Propionate, Butyrate, CO_2, NH_3
	Streptococcus bovis	Proteins	Acetate, Propionate, Butyrate, CO_2, NH_3

surface of protozoa or fungal sporangia. Depending on the utilized substrates and end products, rumen bacteria are categorized into cellulolytic, hemi-cellulolytic, pectinolytic, amylolytic, ureolytic, methane producing, sugar utilizing, acid utilizing, proteolytic, ammonia producing, and lipid utilizing species.

The rumen bacteria and their activities are known to be influenced by several factors, revealing the possibility of their manipulation. These factors include, but not limited to, feeding regimen, diet changes, antibiotic usage, animal's age and health, season, stress level, geographic location, photoperiod, and environment.

14.4.1.2 Rumen Protozoa

Ciliates are the most abundant protozoa representing two physiologically and morphologically different groups viz. entodinomorphs and holotrichs, whereas flagellates occupy the niche to a very limited extent (Fig. 14.5).

The anaerobic rumen ciliates aid in digestion of plant material and ranges from 4×10^6 per mL to 6×10^6 per mL. On the basis of their substrates, the rumen protozoa were classified as starch degraders, soluble sugar utilizers, and lignocellulose hydrolyzers. The large quantities of reserve starch stored in protozoan vacuoles could be used on exhaustion of exogenous energy supply. Larger protozoa prefer structural polymers while smaller protozoa ingest sugars and storage polymers. Generally, holotrichs use soluble sugars and entodinomorphs utilize starch and other plant materials. The protozoal count is affected by ruminal pH, composition of diet, digestibility of diet, frequency of feeding, and season. Protozoa contribute 19–28% of cellulase activity of the total rumen fibrolytic activity. Protozoa are also a good source of lipids and roughly 27% of total lipids are thought to be contributed by holotrichs. The ruminal protozoa help in stabilizing the ruminal fermentation by ingesting feed particles and storing reserve polysaccharides. However, protozoa reduce the bacterial biomass by ingesting the ruminal bacteria. Because of the decreased bacterial protein availability, protozoa decrease the protein to energy ratio and increase the protein requirement by the host. Besides, the protozoa reduce the rate of bacterial colonization and feed degradation.

14.4.1.3 Rumen Fungi

The ruminal anaerobic fungal inhabitants range from10^5 to 10^7 per gram and include *Neocallimastix frontalis, Sphaeromonas communis*, and *Piromonas communis*. Rumen fungi degrade unlignified components of plant cell walls by producing cellulases, hemi-cellulases, and particularly xylanases. High roughage diet increases the fungal proportion, consequently increasing the adhesion and degradation of plant cell wall. The uniqueness in fungi lies in their ability to penetrate the cuticle.

14.4.2 Fermentation of Carbohydrates

The carbohydrate fermentation aids rumen microbial population to attain energy for growth and multiplication. About 75% of the plant tissue dry matter comprises carbohydrates. Microbial fermentation breaks carbohydrates into simple sugars. The end products of carbohydrate fermentation include volatile fatty acids (acetate, propionate, and butyrate) and gases (carbon dioxide and methane).

The speed of fermentation depends on the structure and solubility of carbohydrates. Glucose is a simple sugar with a molecular formula $C_6H_{12}O_6$. Starch contains amylose and amylopectin as polymer chains. Cellulose is beta 1,4 glucose linkage polysaccharide, and hemi-cellulose is composed of beta-linked xylose units and few hexoses. Pectin is beta-linked galacturonan (polysaccharide based on galactose with uronic acid). Lignin, a phenolic compound, is resistant even to microbial enzymatic digestion. Majority of the lignin is indigestible. However, rumen fungi are able to degrade the lignin to a certain extent. Based on the fermentation speed, the Cornell Net Carbohydrate and Protein System (CNCPS) classified soluble sugars as rapidly fermented, starch as less rapidly fermented, and cellulose and hemi-cellulose as slowly fermented carbohydrates. The carbohydrates in roughages are structural (cellulose, hemi-cellulose, lignin, and pectin)

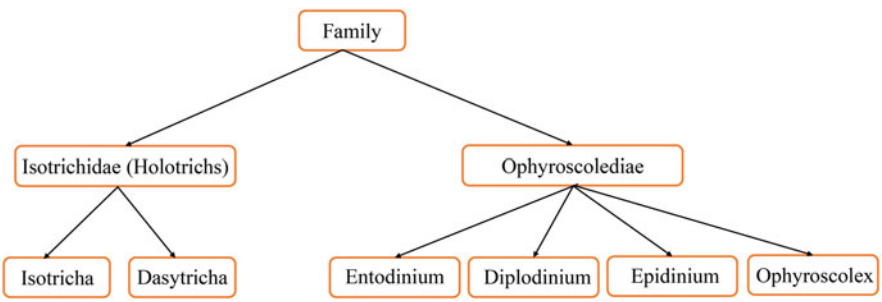


Fig. 14.5 Classification of rumen protozoa. [The Isotricha and Dasytricha genera belongs to Isotrichidae family while the genera Entodinium, Diplodinium, Epidinium, and Ophyroscolex belongs to Ophyroscolediae family]

while those in concentrates are non-structural (sugars and starch).

The extent of carbohydrate fermentation and the end products depends on the type of diet, maturity status, ruminal pH, anti-nutritional factors, and type of microbes. Matured forages are less digestible due to the higher proportion of lignin. Similarly, young grasses are more digestible due to the lower lignin quantity and higher fructosans fraction. Feeding roughage-rich diets leads to the production of acetate at higher proportion and concentrate-rich diet produces higher amount of propionate as end product.

Degradation of carbohydrates involves four steps.

14.4.2.1 Adherence

The bacteria adherence process plays a crucial role in fiber digestion. In the first phase, bacteria transport to fibrous substrate. Later the initial nonspecific adhesion of bacteria to substrates is followed by the specific adhesion of bacteria with digestible tissue. Finally, the attached bacteria proliferate to form colonies on specific sites of the plant tissue. Among various bacteria, coccoids prefer to attach plant cell wall. Attachment helps the bacteria to retain for a longer time and facilitates sustained action. Further, the adherence renders the produced enzymes to come into contact with the substrate and ensures that resulting degradation products are preferentially available. The adherence will be maximum at 40 °C, decreases at a pH below 5.0, and is facilitated at the pH of 5.5–7.8. Certain rumen fluid factors such as phenyl propanoic acid and phenylacetic acid stabilizes the bacterial adherence. The lignin and soluble cellulose derivatives like carboxy methyl cellulose are found to inhibit bacterial attachment.

14.4.2.2 Disaggregation

The fibrous feeds soak in the rumen fluid breaking them into small pieces. Disaggregation increases the degradable ability by rumen microbes. For instance, the starch granules are easily attacked on grounding.

14.4.2.2.1 Extracellular Degradation

The rumen liquor is the best source of bacterial and protozoal enzymes. The enzyme activities in rumen fluid are diverse. They include, cellulases, xylanases, β-glucanases, pectinases, amylases, proteases, phytases, and toxin-degrading enzymes such as tannases. Many of these microbial enzymes act on the soaked and disaggregated feed substances within the rumen, degrading them into short-chain oligosaccharides and sugars. Most of the crystalline cellulose is degraded through extracellular fungal cellulases.

14.4.2.2.2 Intracellular Degradation

The bacteria engulf simple sugars produced through the degradation of oligosaccharide and disaccharides. The intracellular enzymes of microbes metabolize mono- and disaccharides through phosphoroclastic cleavage forming pyruvate, phosphoenol pyruvate, volatile fatty acids, CO_2, and methane (CH_4). The bacterial enzymes degrade starch to maltose and glucose. Maltose is fermented to glucose, which gets converted to pyruvic acid through a metabolic pathway known as glycolysis. The anaerobic glycolysis yields two ATP molecules, contributing the energy source for rumen bacterial maintenance and growth. The degradation of amorphous form of cellulose occurs in anaerobic cellulolytic bacteria by producing enzymes such as endo-β-glucanohydrolase, glucosidase, and endo-xylanase. The hemi-cellulases are highly degradable compared to cellulose and require bacterial cellulases. The β-glucosidase hydrolyzes cellobiose and cellodextrins, producing hexose; however, the enzymatic degradation of hemi-cellulose yields pentoses. Pectin, a polymer of galacturonic acid, will be finally converted to short-chain fatty acids.

14.4.2.3 Formation of Volatile Fatty Acids

The pyruvate, an intermediate compound of carbohydrate fermentation, yields volatile fatty acids, CO_2 and CH_4. The metabolic pathways of pyruvate degradation are presented in Fig. 14.6. The yielded short-chain fatty acids act as major energy sources in ruminants.

14.4.2.3.1 Acetic acid formation

The acetic acid formation occurs in two pathways:

1. *Oxidative decarboxylation of pyruvic acid*
   The pyruvic acid formed during glycolysis enters into mitochondrial matrix and gets converted to acetyl CoAby removal ofCO_2 and H_2, in the presence of thiamine pyrophosphate (TPP) and lipomide. The reaction is catalyzed by pyruvic dehydrogenase. The Acetyl-CoA yields acetic acid by removal of thioester bond and coenzyme A.
2. *Phophoroclastic split*
   The phosphoroclastic reaction of pyruvate cause the formation of acetic acid and formic acid from two molecules of pyruvic acid yield. Being the simplest carboxylic acid, the formic acid (H_2CO_2) is dehydrogenated to CO_2 and H_2. A portion of the generated H_2 is utilized for the production of succinate, propionate, butyrate, and lactate and biohydrogenation of unsaturated fatty acids. Remaining portion will be utilized by methanogenic bacteria for methane production.

14.4.2.3.2 Propionic Acid Formation

The propionic acid formation occurs in two pathways:

1. *By carbon dioxide fixation:* The pyruvic acid combines with CO_2 forming oxalo-acetic acid, which is further

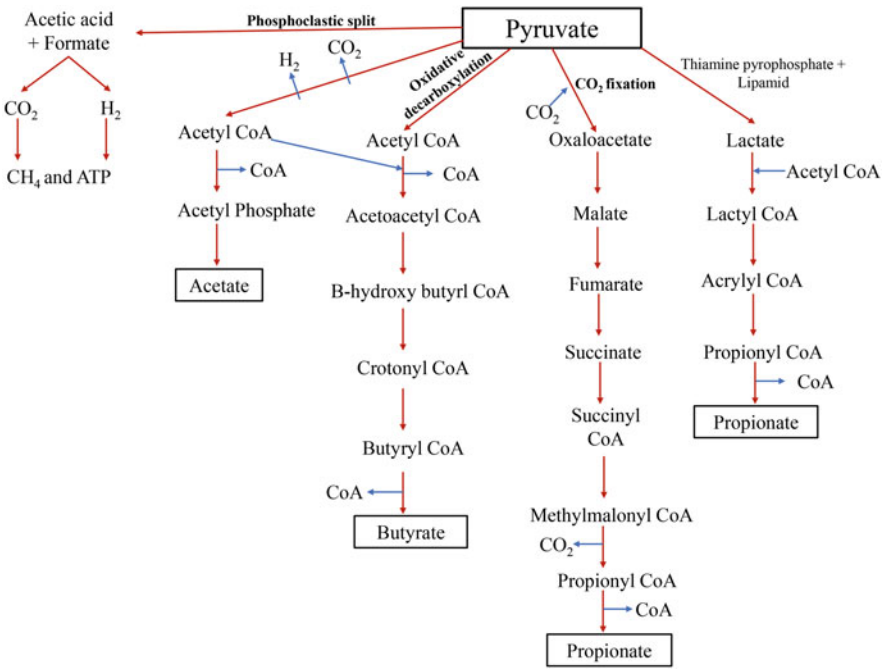


Fig. 14.6 The metabolic pathways of pyruvate degradation. [The phosphoclastic split of pyruvate yields acetic acid consequently producing CH_4. The propionates act as H sink and competes with CH_4, thereby indirectly regulating the CH_4 production whereas the acetate and butyrate formation releases hydrogen]

reduced to malic acid. The resultant malic acid is converted to fumaric acid on removal of one water molecule. The hydrogenation of fumaric acid produces succinic acid followed by its decarboxylation yielding propionic acid.

2. *By acrylate pathway*: The pyruvic acid produces lactic acid on hydrogenation and the resultant lactic acid is converted to acrylic acid on removing water. The hydrogenation of acrylic acid yields propionic acid.

14.4.2.3.3 Butyric Acid Formation

The different pathways of butyric acid formation are two molecules of acetyl-CoA condense to yield acetoacetyl-CoA and $2H_2$ by 3-ketoacyl-CoA thiolase. The acetyl-CoA is converted to beta hydroxybutyrl CoA by reduction. The resultant beta hydroxybutyrl CoA is converted to crotonyl CoA on removal of one H_2O molecule. Reduction of crotonyl CoA leads to formation of butyrl CoA along with one molecule of ATP. The butyrl CoA yields butyrate.

14.4.2.4 End Products of Carbohydrate Fermentation

The end products of carbohydrate fermentation include short-chain fatty acids (acetate, propionate, and butyrate), isoacids (valeric, isovaleric, isobutyric, and 2-methylbutyric acids), and gases such as CO_2, CH_4, and H_2. The CO_2 accounts for 40% of the total rumen gas, CH_4 accounts nearly 30–40% and hydrogen about 5%. The extra hydrogen should be removed from the rumen to maintain pH and rumen ecosystem. Methane acts as hydrogen sink and is considered as net energy loss as most of the CH_4 is lost as eructation. On an average, 4.5 g CH_4 is produced for every 100 g carbohydrate digested.

The total volatile fatty acid content of ruminants ranges from 60 to 120 mEq/L. The individual concentrations of VFA depends upon substrate composition, rumen ecosystem, and health status. The volatile fatty acids proportion changes according to the diet fed. The ratio of acetate, propionate, and butyrate ranges from 70:20:10 for high forage diets to 60:30:10 for high grain diets. The rumen liquor of ruminants fed with normal mixed diet contains 60–65% acetate, 15–20% propionate, and 10–15% butyrate.

14.4.2.5 Absorption of Volatile Fatty Acids

The VFA are directly absorbed from the rumen, reticulum, omasum, and large intestine. Undissociated acids absorb directly by simple diffusion in pH conditions of less than or equal to 6.7. The rate of absorption of VFA increases with the decreased pH, thereby regulating the rumen pH. The absorption of VFA results in accumulation of CO_2 and HCO_3 ion concentration. Lactacidemia is observed because of the lactate absorption on feeding high starch diets. Increased lactic acid formation in the rumen. Increased lactic acid formation in the rumen reduces rumen pH, leading to lactic acidosis and increased *Streptococcus bovis*. Low pH suppresses the growth of other types of bacteria sensitive to pH-causing rumen dysfunction and dehydration. The spiral flow chart of the rumen acidosis sequel is provided in Fig. 14.7.

14.4.2.6 Utilization of VFA in Ruminants

The fermentation of fiber yields acetate as main end product. Low energy and high fiber diets such as roughage leads to

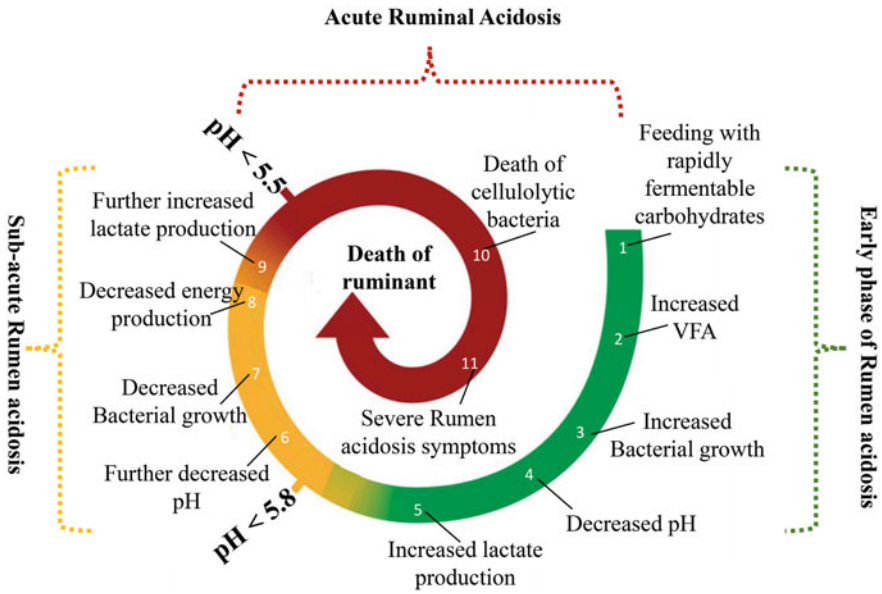


Fig. 14.7 Spiral flow chart of the rumen acidosis sequel. [Feeding rapidly fermentable carbohydrates lead to severe rumen acidosis and death of the animal]

increased ratio of acetate to propionate. Milk fat synthesis require acetate and hence low fiber diets lead to milk fat depression. Starch and sugars yield propionate as end product. The propionate converts to succinate and enters Krebs cycle producing glucose through gluconeogenesis. Propionate contributes to most of the energy required for weight gain and lactose production. Rapidly fermentable carbohydrates such as cereal grains lead to increased propionate proportion. Feeding inadequate amount of grain-based concentrate may decrease the lactose and overall milk production. As the propionate is glucogenic, the acetate and butyrate are ketogenic producing ketone bodies such as acetone, acetoacetic acid, and beta hydroxy butyric acid, ultimately contributing the energy needs of ruminant animals. The ketone bodies are used by skeletal muscles and other body tissues as a source of energy for fatty acid synthesis. Butyrate acts as energy source for rumen epithelium. It stimulates epithelial cell proliferation, consequently improving feed utilization. The concentration of butyrate significantly increases with increased concentrate feeding.

14.4.3 Protein Digestion in Ruminants

The rumen microbes utilize nitrogen and prepare their own sequence of amino acids for their growth and multiplication. The protein metabolism in ruminants depends upon the ability of rumen microbes utilizing ammonia. In ruminant nutrition, proteins can be divided into rumen degradable protein and non-degradable protein. The non-protein nitrogen substances are entirely degradable proteins. Of the protein consumed, depending upon the source, 20–100% will be degraded to ammonia. The rumen degradable protein fraction is hydrolyzed by extracellular proteolytic activities yielding short-chain peptides. Energy is a limiting factor determining the fate of absorbed peptides and amino acids. In the case of energy availability, the amino acids will be transaminated and used for microbial protein synthesis. In the event of energy deficit, the amino acids will be deaminated with the resulting carbon skeleton fermented into volatile fatty acids. Deamination causes the release of ammonia and carbon skeleton; the latter enters into various steps of VFA pathways, consequently producing acetic, propionic, and butyric acids. The rumen undegradable protein escapes ruminal microbial degradation reaching small intestine for enzymatic digestion.

14.4.3.1 Nitrogen Metabolism in Rumen

The protein requirement of ruminants met by the microbial protein. On total nitrogen basis, rumen bacteria contain about 65% protein. For every 1 kg organic matter digested, the microbial yield ranges from 90 to 230 g, which is sufficient for growth and production to certain extent. The peptides are generally absorbed by microbial cells. The efficiency of nitrogen incorporation into bacterial protein is higher for peptides. Whereas the individual amino acids will be subjected to rapid deamination producing NH_3 for bacterial growth along with CO_2 and volatile fatty acids. The pathways of digestion and metabolism of nitrogenous compounds in ruminants is presented in Fig. 14.8.

The bacteria synthesize protein by utilizing certain portion of true protein and entire non-protein nitrogen compounds such as urea. The urea includes urea from diet, saliva, and rumen epithelium. The protein degradation depends on dietary (structure, solubility, number of disulfide bonds and cross linkages between amino acid) and ruminal (type of bacteria, species, ammonia concentration, and pH)

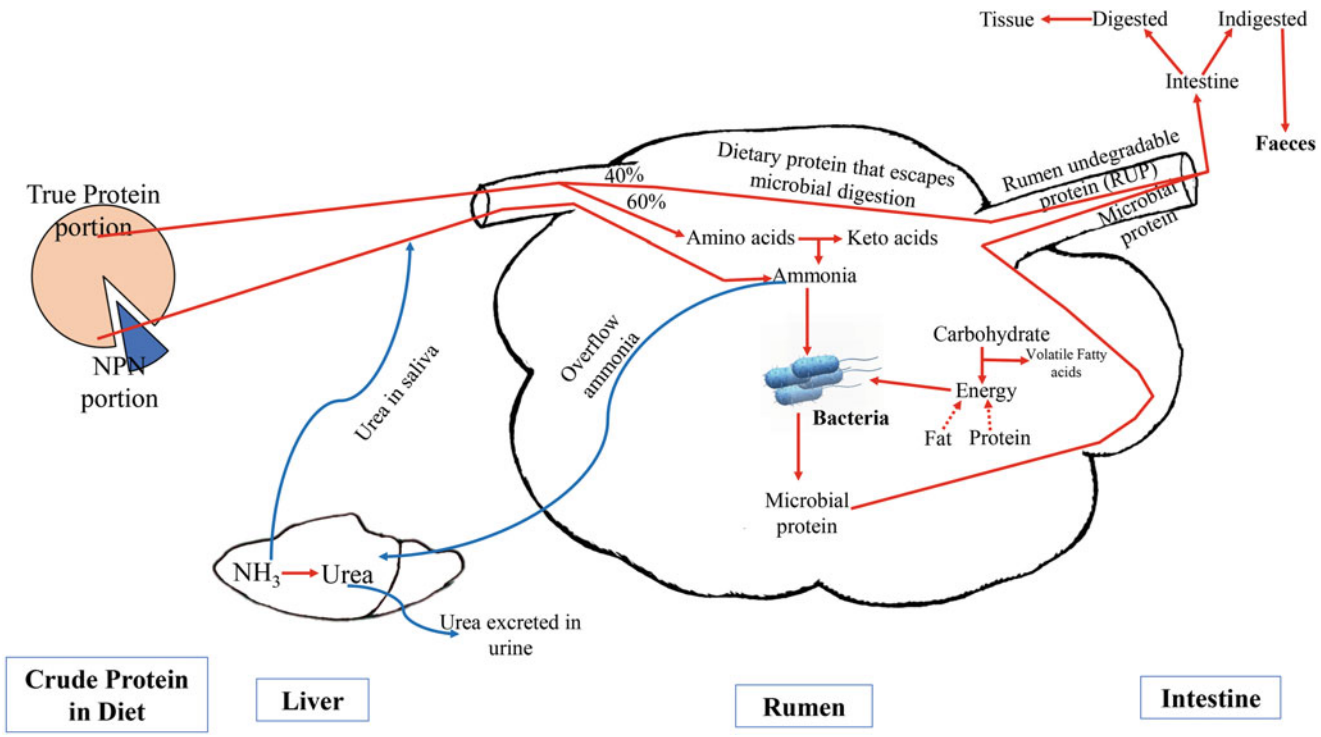


Fig. 14.8 The pathways of digestion and metabolism of nitrogenous compounds in ruminants. [The proteolytic bacteria break down the amino acids into ketoacids and ammonia, which in turn is used to prepare microbial protein]

conditions. The bacteria producing highest proteolytic enzyme concentration include *Butyrivibrio* sps., *Bacteroides* sps., *Selenomonas* sps., *Succinivibrio dextrisolvens*, and *Megasphaera elsdenii*.

More than 80% of the rumen bacteria utilizes ammonia as nitrogen source for growth. The concentration of ammonia nitrogen in rumen liquor varies with the diet from as low as 2 mg/dL in low-protein diets and as high as 100 mg/dL in high-protein diets. Urea in diets is converted by ureolytic bacteria to ammonia. Although the ruminants are able to utilize NPN compounds such as urea, the urea poisoning is not an uncommon phenomenon. Urea poisoning is mainly because of consuming higher quantities of urea, consequently increasing rumen pH and ammonia absorption rate into blood stream.

14.4.3.2 Metabolism of Amino Acids

Certain reactions occur for synthesis of non-essential amino acids, interconversion of amino acids, energy production, and ammonia excretion. These reactions include transamination, deamination, and decarboxylation.

14.4.3.2.1 Transamination

Transamination refers to a process whereby amino groups are removed from amino acids and transferred to acceptor ketoacid without the intermediate formation of ammonia. The most common transaminases are alanine transaminase and aspartate transaminase.

14.4.3.2.2 Deamination

Deamination refers to a process of removal of an amino group from an amino acid. The reaction is catalyzed by deaminases. They are of either oxidative or non-oxidative type.

Oxidative deamination: Oxidative deamination is a form of deamination involving oxidation in the conversion of amino acid to ketoacid and amino group to ammonia.

Non-oxidative deamination: Non-oxidative deamination refers to the deamination process involving non-oxidative steps and is catalyzed by amino acid dehydratase.

14.4.3.2.3 Decarboxylation of Amino Acids

Decarboxylation refers to reactions involving the removal of a carboxyl group from amino acids releasing biogenic amines and CO_2. The decarboxylases may be either specific or nonspecific.

14.4.4 Lipid Metabolism in Rumen

The uniqueness of lipid metabolism in ruminant calves is the presence of pregastric esterases in saliva, providing the

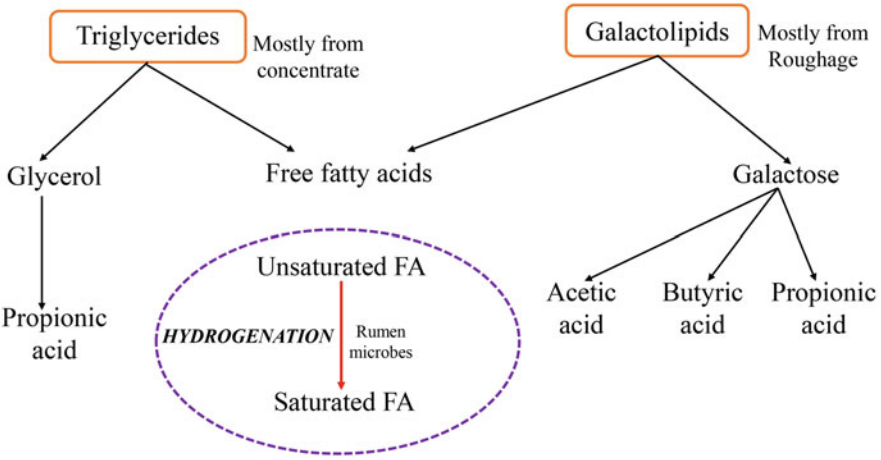


Fig. 14.9 Ruminal metabolism of lipids. [Lipolytic bacteria of rumen cause the breakdown of triglycerides into glycerol and free fatty acids and the galactolipids into galactose and free fatty acids]

ability to start digestion of milk fat from mouth. Dietary lipids include structural lipids of forages and storage lipids of oil seeds. Majority of the lipids in forages are phospholipids, whereas the oil seeds mainly comprise lipids as free fatty acids. A typical ruminant diet contains unsaturated fatty acids at higher proportion. They may be either from the galactolipids of forages or triglycerides of cereal grains and oil seed cakes. The rumen microbes hydrolyze the galactolipids and triglycerides to free fatty acids and glycerol. Glycerol is fermented to propionic acid. The ruminal metabolism of lipids is shown in Fig. 14.9.

The metabolism of lipids by rumen microbes involves a four-stepped process.

14.4.4.1 Hydrolysis of Esterified Fatty Acids

The triglycerides are hydrolyzed to fatty acids through hydrolysis. The lipids are subjected to hydrolysis by microbial lipases viz. cell bound esterases and lipases produced by rumen bacteria. Feeding concentrates at higher levels leads to production of higher concentration of unesterified fatty acids. Less than 10% of polyunsaturated fatty acids escapes the ruminal hydrogenation.

14.4.4.2 Biohydrogenation of Unsaturated Fatty Acids

The unsaturated fatty acids are biohydrogenated to saturated fatty acids. The linolenic acid of grasses is rapidly converted in rumen producing stearic acid, cis-transmonoenoic acid, and cis-trans dienoic acid as end products. Incomplete biohydrogenation generally produces conjugated linoleic acids (CLA), which are proven to benefit human health. Although the biohydrogenation ability is found in both bacteria and protozoa, the extent varies with higher ability in ruminal bacteria such as *Ruminococcus albus* and *Butyrivibrio fibrisolvens*. The biohydrogenation procedure is continuously monitored by the presence of metabolic hydrogen as end products of carbohydrate fermentation.

14.4.4.3 Lipid Biosynthesis in the Rumen

The ruminal fauna, especially bacteria synthesize odd chain fatty acids from propionate and branch chain fatty acids from valine, leucine, and isoleucine. The presence of odd chain and branch chain fatty acids in milk and higher stearic and oleic acids of ruminant fat depots are related to the biohydrogenation and rumen synthesis of fatty acids.

14.4.4.4 Metabolism of Phytal to Phytanic Acid

Phytal is an isoprenoid alcohol present in the chlorophyll of leaves. On consuming forages, the ruminant bacteria hydrogenate phytal to dihydrophytal, consequently producing phytanic acid on oxidization. The resultant phytanic acid is incorporated into rumen organisms and is reported to activate the transcription factors.

14.4.5 Lipid Digestion in Small Intestine

The short-chain fatty acids are mostly absorbed from rumen wall. The lipids leaving the rumen include 85–90% free fatty acids and 10–15% phospholipids. The neutral pH conditions render most of the free fatty acids assaults of calcium, sodium, and potassium. Reaching the acidic abomasal pH conditions dissociates the free fatty acids from the minerals. The free fatty acids adsorb on the degraded feed particles and pass to duodenum through pylorus.

In non-ruminants, monoacylglycerols play an important role in the formation of micelles. However, in ruminants, lysophosphatidyl choline acts as emulsifying agent. Micelle of saturated fatty acids forms under the influence of bile salts and lysolecithin. The pancreatic phospholipase hydrolyzes lecithin into a fatty acid and highly polar lysolecithin. The higher percent of lipid absorption occur in lower part of the jejunum. The bile salts are absorbed in ileum and reaches back to liver to contribute to bile. After entering into mucosal cells, resynthesis of triglycerides occurs via the glycerophosphate pathway. The triglycerides combine with

the proteins inside the Golgi body to form chylomicrons. The chylomicrons and very low-density lipoproteins (VLDL) are carried to adipose tissue by capillaries.

Learning Outcomes

Ruminants possess large compartmental gastrointestinal tract viz. rumen, reticulum, omasum, abomasum, and intestine, which favors handling large amounts of fibrous plant materials. In adult ruminants, the rumen harbors vast range of microbes enabling microbial fermentation of ingesta before exposing to gastric juices of abomasum. The fermentation of complex carbohydrates produces short-chain fatty acids (acetate, propionate, and butyrate), and gases such as CO_2, CH_4, and H_2. The protein metabolism in ruminants depends upon the ability of rumen microbes utilizing ammonia to produce microbial proteins. Ruminal bacteria split the fatty acids and sugars from glycerol backbone through lipolysis. The metabolism of lipids by rumen microbes involves a four-stepped process viz. hydrolysis of esterified fatty acids, biohydrogenation of unsaturated fatty acids, lipid biosynthesis in the rumen, and metabolism of phytal to phytanic acid.

Exercises

Objective Questions

1. The juice which plays an important role in the digestion of fats is _____.
2. The feedstuff that is regurgitated and remasticated in mouth of ruminants is _____.
3. Important parameter that stimulates chewing activity and saliva production is _____.
4. An example for lipolytic bacteria is _____.
5. Rumen Holotrichs use _____ and entodinomorphs utilize _____for survivability.
6. _____ are able to penetrate the cuticle and degrade plant cell wall.
7. The roughage fraction composed of beta-linked galacturonan structure is _____.
8. Feeding roughage and concentrate-rich diets leads to the production of _____ and_____ as fermentation end products, respectively.
9. The first step of bacterial degradation of carbohydrate is _____.
10. _____ reaction of pyruvate causes the formation of acetic acid and formic acid from two molecules of pyruvic acid.
11. _____ is an example for hydrogen sink in rumen.
12. The ratio of acetate, propionate, and butyrate ranges from _____ for high forage diets.
13. Feeding rapidly degradable starch substances at huge level may leads to _____.
14. _____ is the desired carbohydrate fermentation end product for milk fat synthesis.
15. _____ is the desired carbohydrate fermentation end product for weight gain and lactose production.
16. _____ acts as energy source for rumen epithelium.
17. On total nitrogen basis, rumen bacteria contain about _____ protein.
18. During lipid metabolism, glycerol is fermented to _____ volatile fatty acid.
19. _____ is an isoprenoid alcohol present in the chlorophyll of leaves.
20. The short-chain fatty acids are mostly absorbed from _____.

Subjective Questions

1. Explain in detail about the mechanical factors involved in ruminant digestion.
2. Write about the microbial habitat of rumen and classify the bacteria according to the substrate.
3. Elucidate the metabolic pathways of pyruvate degradation.
4. Explain clearly the pathways of digestion and metabolism of nitrogenous compounds in ruminants.
5. Describe the role of rumen biohydrogenation procedure in lipid metabolism.

Answers to Objective Questions

1. Bile juice, pancreatic juice
2. Lighter roughage pieces
3. Physically effective NDF
4. Micrococcus sps.
5. Soluble sugars and starch
6. Fungi
7. Pectin
8. Acetate and propionate
9. Adherence
10. Carbon dioxide fixation
11. Propionate, sulfate, and nitrate
12. 70:20:10
13. Subacute rumen acidosis
14. Acetate
15. Propionate
16. Butyrate
17. 65%
18. Propionic acid
19. Phytal
20. Rumen

Keywords for Answer to Subjective Questions

1. Mastication, deglutition, rumination, eructation, regurgitation, remastication, reinsalivation, redeglutition
2. Rumen fermentation, nitrogen metabolism, carbohydrate metabolism, cellulolytic bacteria, proteolytic bacteria, lipolytic bacteria
3. Phosphoclastic split, oxidative decarboxylation, acetyl Co-A, lactyl Co-A, acetate, propionate, butyrate
4. Non protein nitrogen, urea, microbial protein, rumen degradable protein, rumen undegradable protein
5. Triglycerides, galactolipids, glycerol, galactose, unsaturated fatty acid

Further Reading

Books

Duncan AJ, Poppi DP (2008) Nutritional ecology of grazing and browsing ruminants. In: Gordon IJ, Prins HHT (eds) The ecology of browsing and grazing. Ecological studies, vol 195. Springer, Berlin, Heidelberg

Hyder I, Reddy PRK, Raju J (2017) Alteration in rumen functions and diet digestibility during heat stress in sheep. In: Sejian V, Bhatta R, Gaughan J, Malik P, Naqvi S, Lal R (eds) Sheep production adapting to climate change. Springer, Singapore

McDonald P, Edwards RA, Greenhalgh JFD, Morgan CA, Sinclair LA, Wilkinson RG (2011) Animal nutrition, 7th edn. Pearson Education Limited, London, UK

Reddy DV (2021) Principles of animal nutrition and feed technology, 3rd edn. Oxford and IBH Publishing Co. Pvt. Ltd., New Delhi, India

Research Articles

Baldwin RL, Vi McLeod KR, Klotz JL, Heitmann RN (2004) Rumen development, intestinal growth and hepatic metabolism in the pre-and postweaning ruminant. J Dairy Sci 87:E55–E65

Fiorentini G, Carvalho IP, Messana JD, Canesin RC, Castagnino PS, Lage JF, Arcuri PB, Berchielli TT (2015) Effect of lipid sources with different fatty acid profiles on intake, nutrient digestion and ruminal fermentation of feedlot Nellore steers. Asian Australas J Anim Sci 28(11):1583–1591

Reddy PRK, Kumar DS, Rao ER, Seshiah CV, Sateesh K, Rao KA, Reddy YPK, Hyder I (2019a) Environmental sustainability assessment of tropical dairy buffalo farming vis-a-vis sustainable feed replacement strategy. Sci Rep 9:16745

Reddy PRK, Kumar DS, Rao ER, Seshiah CV, Sateesh K, Rao KA, Reddy YPK, Hyder I (2019b) Assessment of eco-sustainability vis-à-vis zoo-technical attributes of soybean meal (SBM) replacement with varying levels of coated urea in Nellore sheep (*Ovisaries*). PLoS One 14(8):e0220252

Shen H, Lu Z, Xu Z (2017) Associations among dietary non-fiber carbohydrate, ruminal microbiota and epithelium G-protein-coupled receptor, and histone deacetylase regulations in goats. Microbiome 5:123