Common Terms Used in Animal Nutrition

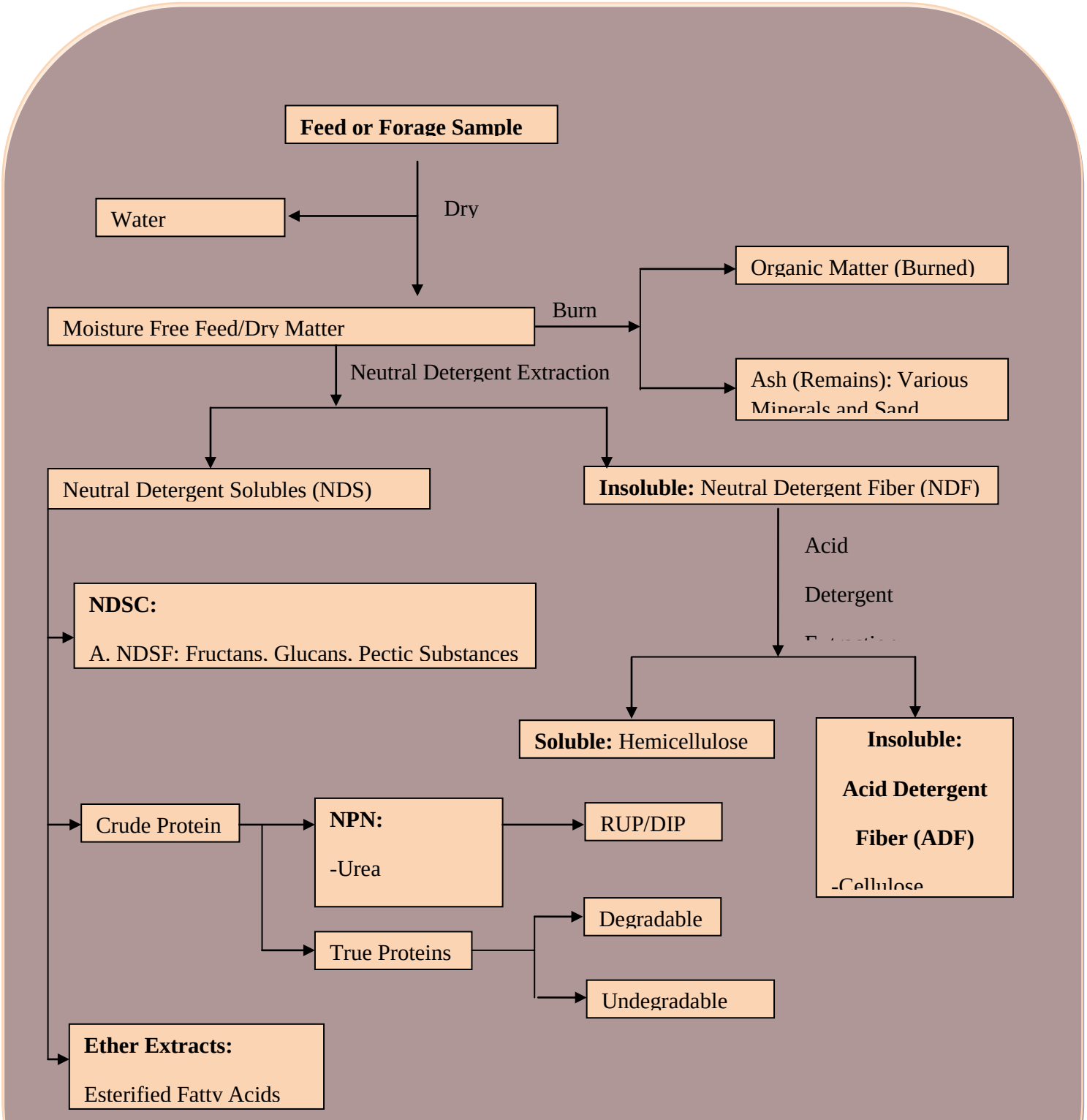
The feed bill is the largest operating cost in animal production. To decrease this cost, we must supply the balanced amount of feed to the animals according to type production. Overfeeding is wasteful. Underfeeding will decrease animal productivity and profitability. Therefore, proper animal nutrition are crucial to the profitability of the livestock enterprise.

Analyses of the composition of feed or forage are used to assess their nutritive value (Figure 1). A typical feed analysis includes measurements of some important quality items or parameters (e.g., crude protein, fiber, digestibility, etc.) used to define the nutritive value. Other items are analyzed under some special circumstances (i,e., acid detergent insoluble crude protein (ADICP) is usually only measured if heat damage to the feed is suspected).

Many of the parameters included in laboratory analysis are estimated from measured feed quality attributes. For example, digestible energy, total digestible nutrients and intake potential are all estimated from the concentration of the various fiber components and the relationship between them.

Our primary objective is to list the common terms used when discussing animal nutrition so that one might be better informed when consulting with nutritionists, feed salesmen, professional personnel, veterinarians, feed laboratory managers and other industry professionals.

These terms listing will be helpful when reading articles on animal feeding and nutrition, feed analysis reports or tags associated with feeds.



Abbreviations: *DIP = Degradable Intake Protein; NDSC = Neutral Detergent Soluble Carbohydrate; NDSF = Neutral Detergent Soluble Fiber; NPN = Non Protein Nitrogen; RUP = Rumen Undegradable Protein.*

Figure 1. A figure that describes the partitioning of organic and mineral components in a feed sample

Acid Detergent Fiber (ADF)

The fibrous component represents the least digestible fiber portion of forage or other roughage. This includes lignin, cellulose, silica and insoluble forms of nitrogen. The higher ADF, the lower digestible energy, which means that as the ADF level increases, digestible energy levels decrease. During analysis, ADF is the remaining after boiling a forage sample in acid detergent solution. ADF is often used to calculate digestibility, total digestible nutrients (TDN) and/or net energy for lactation (NE).

Acid Detergent Insoluble Crude Protein (ADICP);

Acid Detergent Fiber Crude Protein (ADFCP)

acid detergent insoluble protein (ADIP),

acid detergent insoluble nitrogen (ADIN),

acid detergent fiber protein (ADFP)

ADICP is the insoluble protein fraction remaining in the acid detergent fiber residue. ADICP escapes ruminal breakdown and represents the portion of the protein that is not degradable and is therefore unavailable to the animal. It contains any heat-damaged protein that may result from heating during processing. In this case, a portion of the protein reacts with carbohydrates to form an indigestible complex. It is expressed as a percent of crude protein. It is an adequate estimate of heat-damaged protein in forage feeds but not in non-forage feeds.

Additive

a substance/mixture of substances added to the ration to meet a specific purpose. An additive may enhance the nutritive value, sensory value or shelf life of the ration. Additive is involved in the production, processing, packaging and/or

storage of the feed without being a major ingredient.

Aflatoxins

Mold growth in or on feed can result in the production of many different types of toxic outputs. As a group, these toxic substances are commonly called mycotoxins. The term aflatoxins describe a group of toxins produced by some species of Aspergillus. There are four major aflatoxins named B1, B2, G1, G2 besides, two metabolic products known as M1 and M2 that are of significance as direct contaminants of foods, feeds and milk.

Fungal growth and aflatoxin contamination are the result of interactions among the fungi, the host (foods or feeds) and the environment. On a crop, aflatoxin contamination of peanuts and corn is favored by adverse condition (high temperatures, prolonged drought conditions and high insect infestation), while after harvest production of aflatoxins on them is favored by higher water content, warm temperatures and high humidity. Forages are generally not analyzed for aflatoxins but in some situations (e.g., corn or sorghum silage that is at risk) this analysis may be necessary.

The presence of aflatoxins in feeds, forages and foods is an important risk factor and is associated with many diseases in wild animals, domestic animals and humans that are generally termed aflatoxicosis. Aflatoxicosis is consider a hepatic disease. Liver damage, decreased reproduction, production, embryonic death, teratogenicity, cancer and suppressed immune function are caused by low levels of aflatoxin.

The FDA's action level for human food is 20 ppb total aflatoxins, with the exception of milk, 0.5 ppb for aflatoxin M1. The FDA action level for most feeds is 20 ppb (Table 1).

If the level of aflatoxins in forage is higher, it may be fed in combination with

other feeds containing no aflatoxins.

Table 1. FDA action levels for aflatoxins	
Commodity	Action level(µg/kg or ppb)
All products, except milk, designated for humans	20
Corn for immature animals and dairy cattle	20
Corn for breeding beef cattle and mature poultry	100
Corn for beef cattle	300
Cottonseed meal (as a feed ingredient)	300
All feedstuff except corn	20

Amino Acids

A molecules containing an amine group, a carboxylic acid group and a side chain that varies between different amino acids. Protein is made from amino acids. There are 20 known standard amino acids forming various proteins. When taken up into the body in the diet, the 20 standard amino acids are either used to synthesize proteins and other biomolecules or broken down into urea and carbon dioxide. There are ten essential amino acids and the other ten are called non-essential amino acids. Animals cannot synthesize the essential amino acids from other compounds at the level needed for normal growth, so they must be obtained from food.

Anorexia

loss of appetite in disease condition.

Anti-quality Factors (deleterious substances)

Forages may contain various harmful compounds/metabolites that can adversely affect animal performance even cause sickness or death. These compounds are called anti-quality factors and include tannins, nitrates, alkaloids, cyanoglycosides, estrogens and mycotoxins. The occurrence and severity of these

factors depend on the forage and weed species present, season, environmental conditions and sensitivity of the animal. High quality forages should be free from these deleterious materials.

Appetite

It is a desire or inclination for feed. It is a conditioned reflex. It is related to taste, smell and appearance of food. Well developed in man than in farm animals.

Ash

An inorganic matter of feed sample. It is determined in a laboratory by burning the sample at a high temperature and weighing the residue.

As-fed Basis "As-is Basis" or "As-received Basis."

Feed analyses reports state results based on the feed's natural condition (including water) and/or on a dry matter basis. It is affected by the sample's moisture level before drying. When comparing two or more analyses, it is better to utilize the data from the "Dry Matter Basis" rather than the "As- fed Basis" unless you are mixing a ration.

Balanced Ration

Complete feed formulated to provide a specific animal species and class with appropriate amounts of all nutrients required for maintenance and a given level of performance during 24hrs.

Balanced diet

It the feed that supplies all the essential nutrients in the proper amounts required for optimum performance of the animal.

Basal (Energy) Feeds

Nutritionally, basal feeds are mainly concentrated sources of energy being

especially rich in starches and sugars. They are grains and grain by-products that contain not more than 16% protein and 18% crude fiber.

Botulism

Botulism is a muscle-paralyzing disease caused by botulinum toxin, a potent neurotoxin produced mainly by an aerobic bacterium Clostridium botulinum and also by a few strains of C. baratii and C. butyricum.

Botulism can result from the ingestion of the toxin or the growth of C. botulinum on anaerobic food/feed tissues. Seven types of botulinum toxin, designated A through G, have been identified. Types A, B, E and F cause illness in humans. Type C is the most common cause of botulism in animals. Type D is sometimes seen in cattle and dogs, and type B can occur in horses. Types A and E are found occasionally in birds. Type G rarely causes disease, although a few cases have been seen in humans. All types of botulinum toxin result in the same disease; however, the toxin type is important for treatment.

The toxins come from a variety of sources. Decaying vegetable matter (e.g., grass, hay, grain, spoiled silage) and carcasses can cause botulism in animals. Ruminants may inadvertently be fed hay or silage contaminated by carcasses of birds or mammals that may contain the toxin. Horses usually ingest the toxin in contaminated forage.

Botulinum toxins are large proteins that can be easily denatured. Toxins exposed to sunlight are inactivated within one to three hours or 0.1% sodium hypochlorite, 0.1 M NaOH, heating to 80°C for 30 minutes or 100°C for 10 minutes. Chlorine and other disinfectants can destroy the toxins in water.

By-pass Protein (undegradable intake protein (UIP),

rumen undegradable protein (RUP) or escape protein.

The portion of protein, that has a slow rate of degradability in the rumen. It is

reach the lower gastrointestinal tract essentially intact and be digested directly in the small intestine as it would be in non-ruminants. This can provide a balance of amino acids unaltered by microbial digestion and synthesis.

Carbohydrates

Carbohydrates are composed only of the elements carbon, hydrogen and oxygen, and consider the main source of energy for animals. Animals have the majority of their energy from the carbohydrates in feeds. The plant carbohydrates are divided into non-structural and structural. The non-structural carbohydrates serve as storage and energy reserves and available for more rapid metabolism to supply energy (e.g., sugars, starch, and pectin). The structural carbohydrates are not used for energy storage and provide fiber and anatomical features for rigidity and even water transport (e.g., fibrous cellulose and hemiellulose).

Cellulose

Cellulose is a major structural carbohydrate that is present in plant cell walls. Cellulose is an unbranched chain of 7,000 to 15,000 glucose molecules that are linked together by β-1,4 bonds. Cellulose is a major constituent of the structural fiber in forages and can be utilized by microorganisms in the rumen. Cellulose is estimated as follows:

Cellulose = ADF – (ADL + Ash), where ADF is acid detergent fiber and ADL is acid detergent lignin.

Cob

It is the fibrous inner portion of the ear of maize from which the kernels have been removed.

Complete feed

It is a balanced ration for the animal. It provides all the nutritional

requirements (except water) needed to maintain normal health and to promote production.

Concentrates

Concentrates refer to animal feeds that are rich in energy and/or protein but low in fiber, such as corn, soybean meal, oats, wheat, molasses, etc. Also described as feed/feed mixture which supplies primary nutrients (protein, carbohydrates and fat). It is a commercially supplement which refers to a concentration of protein, minerals or of vitamins in excess of those found in basal feed.

Crude Fat

Crude fat is an estimate of the total fat content of feeds taken from older collection of methods known as proximate analysis. The crude fat is estimated using ether extraction. Crude fat contains true fat (triglycerides) as well as alcohols, waxes, terpenes, steroids, pigments, ester, aldehydes and other lipids.

Crude Fiber (CF)

Carbohydrates divide into digestible and indigestible fractions by proximate method. When CF content is higher, the energy content of the feed is lower because indigestibility of it. Measuring crude fiber was one part of the original system of analyzing the "digestible" fraction in feedstuffs. This method is often referred to as the Weende System of proximate analysis. CF is partially digestible by micro-organisms in the rumen. Crude fiber accounts for most of the cellulose but only a portion of the lignin and no ash, so it underestimates true fiber and is less than ADF. Thus, CF is not a good indicator of digestibility, and the use of it in feeds for ruminants is declining.

Even though CF is not a very useful parameter for quantifying forage fiber where lignin content is substantial, the CF is a reasonable estimate of the fiber in grains because of their low lignin content. So, it is commonly used for analysis of

feeds for monogastric animals. Crude fiber is used as the legal measurement of fiber content in grains and finished feeds.

Crude Protein (CP)

Proteins are composed of amino acids. They are a major component of vital organs, tissues, muscles, hair, skin, milk and enzymes. Protein is daily required for maintenance, lactation, growth and reproduction.

The crude protein content of a feed represents the total nitrogen (N) in the diet, which includes both true protein and non-protein nitrogen (urea and ammonia in a feed; nitrate is not included in non-protein nitrogen). Because N is an integral part of any amino acid, non-protein nitrogen has the potential to be used for protein synthesis by microflora. In Lab, total N is first determined and then the amount of protein is calculated by multiplying the total N by a factor. This factor is 6.25 for forages because proteins generally contain 16 percent nitrogen. For seeds, this factor is different (e.g., 5.70 for wheat and 5.90 for other cereal grains).

Crude protein in feeds for ruminants can be further fractionated according to their rate of breakdown in the rumen, as discussed below for neutral detergent fiber insoluble crude protein (NDFICP) and discussed previously for acid detergent fiber insoluble crude protein (ADFICP).

CP is an important indicator of the protein of a forage, and even estimates of non-protein nitrogen are important in evaluating nutritive value. However, it is a false perception that protein is always the most limiting nutrient in the animal's diet and CP is the ultimate measure of a forage quality. The energy value of forages is often the most limiting attribute for meeting an animal's requirements in most forage-based feeding.

Degradable Intake Protein (DIP)

The DIP, Rumen Degradable Protein (RDP), represents the portion of intake

crude protein that is degraded to ammonia and amino acids in the rumen by microbes. This fraction of CP consists of non-protein nitrogen and the true proteins that are soluble and those having intermediate ruminal degradability. The DIP is expressed as a percentage of CP, where DIP = NPN + Soluble True Protein + True Protein of Intermediate Degradability.

Detergent Fiber Analysis

Since crude fiber has been found to have an unsatisfactory relationship with livestock performance, it has limited value in ruminant nutrition. Most laboratories do not use the proximate analysis system and have replaced it with the Van Soest analysis system. The idea behind the detergent fiber analysis is that plant cell materials can be divided into less digestible cell walls and the highly digestible cell contents (starch and sugars).

Generally,

NDF = Hemicellulose + Cellulose + Lignin + Ash

ADF = Cellulose + Lignin + Ash

Hemicellulose, cellulose and lignin are indigestible in non-ruminants, while hemicellulose and cellulose are partially digestible in ruminants. NDF is a good indicator of the "bulk" fiber and has been used to predict feed intake. In contrast, ADF is a good indicator of digestibility (negatively correlated) and thus energy intake.

The detergent fiber analysis system is the most widely accepted method for forage analysis. However, many organisation still base part of their regulations on proximate terms.

Digestibility

It refers to the degree at which a feedstuff is absorbed (used) in the animal body as it passes through an animal's digestive tract. It varies greatly with the type

of feedstuff and animal spp.

Digestible Dry Matter (DDM) or Dry Matter Digestibility (DMD)

DDM (or DMD) is the part of the dry matter in a feed that is digested by animals. It is often estimated by measuring in vitro or in situ digestibility. The DDM can be calculated as follows:

%DDM = 88.9- [0.779 × %ADF (on a dry matter basis)].

Digestible Energy (DE)

Digestible energy provides the actual amount of energy from a feed that can be available for use by the animal. It is estimated by subtracting energy lost in the feces from the gross intake energy (GE), (i.e., DE = GE – FE). Digestible energy is commonly used to evaluate poultry and horse feed. For poultry feed, DE is considered as an measure of feed quality, because FE is almost the sole form of energy loss during digestion.

Digestible Neutral Detergent Fiber (dNDF)

The 48-hour in vitro digestible fraction of Neutral Detergent Fiber (NDF) is expressed as a percentage of the dry matter content of a feed sample.

Distillers Grains

Distillers grains are residual grains or byproducts remaining after the starch from grains has been fermented to alcohol. Traditionally, alcohol was produced mainly for beverages. This increased demand has led to the development of ethanol production plants in various places around the world. With increasing ethanol production, the opportunity currently exists for using a substantial quantity of distillers grains as feed in livestock industry.

Dry Matter (DM)

Dry matter represents everything contained in a feed except water; this

includes protein, fiber, etc. It is determined by drying the feed sample in an oven until the sample reaches a stable weight.

However, estimates of the DM of fermented materials such as silage are complicated by the presence of volatile fatty acids. These acids are removed in the drying process but they are part of the dry matter and are digestible. Analysis of the fodder without ensiling provides a more accurate estimate of fiber fractions and digestibility contained in the silage.

Dry Matter Basis "Dry Basis," "Dry Results" or "Moisture-free Basis."

Dry matter basis indicates the nutrient percentage in a sample based on its dry matter content. As the water has variable content in forages, excluding the water or expressing the nutrient content on a dry matter basis eliminates the dilution effect of the water, thereby providing the essential common basis for direct comparison of the nutrient contents in different forages.

Dry Matter Intake (DMI)

Dry matter intake is the amount of dry matter consumed by the animal. Intake increases as the digestibility of the forage increases. When, the percent of NDF increases in the feed, animals consume less. This relationship, along with estimates of NDF digestibility, is used to estimate DMI for grasses and legumes using the following equations:

$DMI_{Grass} = -2.318 + 0.442 \times CP - 0.0100 \times CP^2 - 0.0638 \times TDN + 0.000922 \times TDN^2 + 0.180 \times ADF - 0.00196 \times ADF^2 - 0.00529 \times CP \times ADF$

Where DMIGrass is expressed as % of BW, and CP (Crude Protein), ADF (Acid Detergent Fiber), and TDN (Total Digestible Nutrient) are expressed as % of DM .

$DMI_{Legume} = 120/ NDF + (((NDFD - 45) \times 0.374)/ 1350) \times 100$

Where DMI_{Legume} is expressed as % of BW, NDF (Neutral Detergent Fiber) as % of DM, and NDFD (48-hour in vitro NDF digestibility) as % of NDF.

Dry matter intake is affected by the animal condition (age, body weight, pregnancy status, level of milk production, etc.), feed factors (palatability, balance of the diet, anti-quality factors in the feed) and the environment (temperature, humidity, etc.).

Ensiled

Ensiled refers to the plant materials preserved by anaerobic fermentation and typically stored in a bag, wrapped bale, bunker or upright silo.

Ethanol Soluble Carbohydrates (ESC)

ESCs are the carbohydrates that can be solubilized and extracted in 80% ethanol. ESC includes primarily monosaccharides and disaccharides.

Ether Extract

Ether extract is a portion of dry matter extracted with ether. It is a laboratory test to approximate the total fat (or crude fat) content of a feed and includes some waxes, pigments and other lipids to a minor degree in addition to true fats.

Fat

Fats are “triglycerides of fatty acids” that are a rich source of energy for animals. It contains 2.25 to 2.8 times the energy found in carbohydrates and is highly digestible. Fat is supplemented to rations to enhance energy content when intake may be limited due to poor animal condition, less palatable feed or environmental stress. Some concentrates (such as oil bearing seeds) contain relatively high levels of fats and oils. Fats are composed of fatty acids. Fats in feed

samples are typically determined through ether extraction (EE). In addition to fat, EE may solubilize some other compounds like plant pigments, fat soluble vitamins, esters and aldehydes. This is why the measurement of fat through EE is called crude fat. True fat can be measured by determining the content of fatty acids or it can be estimated in forages as ether extract minus one.

Feed

It is a material, which after ingestion by the animal is capable of being digested, absorbed and utilized i.e. before transformed into body elements of the animal. A feed is merely the carrier of nutrients. No single feed has been found that is fully nutritionally for and balanced to the need of a certain animal.

Feedstuff/Feed ingredients

a feeding stuff is any product, either of natural origin or artificially that when properly used has nutritional value in the diet. It includes natural feeds of animal origin, plant origin, synthetic and other pure nutrients.

Feeding

It is a practical application of nutrition, i.e. consideration of management, formulation, palatability, economics, etc.

Feedmill

It is an firm/place where feeds/commercial feeds are provided using specialized equipment according to the feed formulation.

Feedmillers

Owner of a feedmill, for commercial/personal use.

Feed Microscopy

This is the science of identification, evaluation of feeds/feedstuffs by visual appraisal using a microscope, hand lenses. Essentially it involves physical and textural examinations.

Formulation

It is the process of constructing a feed or diet formulation.

Forage

Forage refers to any material (plants or plant parts) other than separated grains fed to or grazed by animals and contains more than 18% crude fibre. Forage may be fresh, dry or ensiled (e.g., pasture, green chop, hay, haylage).

Forage Quality

Forage quality refers to the ability of a forage to support desired levels of animal performance. It is a function of voluntary intake and nutritive value (nutrient content and digestibility).

GIT

Gastro intestinal tract, responsible for the digestion, absorption and assimilation of feed and nutrients.

Gross Energy (GE)

The total energy in a feed before accounting for losses due to normal digestive, metabolic and productive functions. It is estimated by measuring the amount of heat produced when a feed is completely oxidized in a bomb calorimeter. It is not a useful measure as the gross energy in most common feeds is about the same, while they do not result in similar animal performance. For

example, GE in oat grain = GE in oat straw.

Heat Damage

Heat damage is the result of exposing the feedstuff to excessive heat during processing or storage, which irreversibly binds protein to the carbohydrates portion of the feed through a chemical reaction called “Maillard Reaction” or “Browning Reaction,” thus making proteins partially or wholly unavailable for digestion.

Hemicellulose

Hemicellulose is a carbohydrate that exists in almost all plant cell walls along with cellulose. Whereas cellulose is composed only of glucose, hemicellulose is composed of many other sugars (e.g., glucose, xylose, mannose, galactose, arabinose, etc.) in chains of 500 to 3,000 sugar units. Hemicellulose is a branched polymer. In contrast, cellulose is not branched. As hemicellulose content increases in animal feed, the voluntary feed intake typically decreases.

Husks

It is leave enveloping an ear of maize or outer covering of kernels or seeds especially in the dry form.

Hulls

Outer covering of grain or kernel.

International Unit (IU)

A standard unit of potency of a biological agent (e.g., vitamin, hormone, antibiotic, antitoxin); also called a USP unit.

In Vitro

In vitro generally refers to the technique of performing a given biological procedure in a controlled environment outside of a living organism. In other words, it is a process that is carried out in a test tube. In feed testing, in vitro refers to a feed sample that is digested in test tubes or tested outside the animal.

In Vitro Digestibility or *In Vitro* Dry Matter Digestibility (IVDMD)

In vitro digestibility of a feed is determined by incubating a feed sample with rumen fluid in a beaker or test tube for 24 to 48 hours, followed either by addition of acid and pepsin and further incubation for 24 hours or by boiling in neutral detergent fiber solution.

In Situ Digestibility

In situ digestibility is determined by incubating a ground forage sample in a porous nylon bag placed within the rumen via a fistula or port in the animal's side (in situ) for a fixed time period.

Kernel

It refers to a whole grain.

Lignin

Lignin is a complex carbohydrate compound, a major structural component of mature plants, contained in the fibrous portion of plant stems, leaves, cobs and hulls. It is undigestible and so, has a negative impact on cellulose digestibility. As the lignin content in a feed increases, digestibility of its cellulose decreases, thereby lowering the amount of energy potentially available to the animal.

Lipids

Lipids are substances found in plant and animal tissues that are insoluble in water but soluble in benzene or ether; lipids include glycolipids, phosphoglycerides, fats, oils, waxes and steroids.

Lysine

Lysine is an essential amino acid for protein synthesis. It is the first limiting amino acid in corn/soybean-based diets. It can be added to diets in a synthetic form.

Macro-minerals

Macro-minerals, also called major minerals, are the elements present in the animal body in relatively larger amounts than micro minerals. Therefore macro-minerals are required in large amounts, generally in gram (g) per head per day, if the animal is to live and function properly. Macro-minerals perform specific roles in the body's tissues and functions (help to build body tissues (e.g. bone) or to regulate metabolic activities). Macro minerals interact with each other and must be supplied in proper amount and ratios to maintain appropriate animal function. The seven essential macro-minerals are: calcium (Ca), phosphorus (P), sodium (Na), magnesium (Mg), potassium (K), sulphur (S) and chlorine (Cl). The total mineral content of the body is approximately (3-4 %) of body weight. Calcium makes up approximately 1.5-1.75 % of total body weight, phosphorus makes up approximately (1-1.10 %) of total body weight, and magnesium makes up approximately 0.04 percent of total body weight.

Megacalorie (Mcal)

The amount of energy required for an animal to perform a specific function is

most often expressed as a unit of heat, the smallest unit of which is called a "calorie."

- 1,000 Calorie (cal) = 1 Kilocalorie (Kcal)
- 1,000 Kcal = 1 Megacalorie (Mcal)
- Therefore, 1 Mcal = 10^6 cal

Metabolizable Energy (ME)

Metabolizable energy equals the gross feed energy minus the energy lost in the feces, urine and gaseous product of digestion:

- ME = GE – FE (energy in feces) – (energy in urine)– (energy in gases)
- ME = DE (digestible energy) – (energy in urine) –(energy in gases)
- Because, DE = GE – FE

A commonly used formula to estimate ME in beef feedstuffs is: ME = 0.82 × DE.

Methionine

An essential sulfur containing amino acid involved in many vital enzymatic processes in the human and animal body.

Micro-minerals (trace minerals)

Micro-minerals, are present in the animal body tissues in extremely low concentrations. They are required in small amounts, generally in milligram (mg) or microgram (μg) per head per day, but play important roles in animal nutrition. There are 10 micro-minerals recognized in animal nutrition: iron (Fe), manganese (Mn), copper (Cu), zinc (Zn), selenium (Se), cobalt (Co), iodine (I), chromium (Cr), molybdenum (Mo) and nickel (Ni).

Copper is particularly toxic in sheep and selenium can also be toxic.

Deficiencies and toxic levels are quite regional.

Minerals

In feed analysis, minerals refer to inorganic essential for life.

Monogastric

Animals having a single compartment or simple stomach system (poultry, horse).

Mycotoxins

Mycotoxins are toxic to animals, and are produced on plants by fungi, particularly during weather stress during the growing or harvest seasons or during feed storage (e.g., ochratoxin, vomitoxin, zearalenone, aflatoxin and T-2, etc).

Net Energy (NE)

The amount of feed energy actually available for animal maintenance, growth and production. Conceptually, total NE is the portion of metabolizable energy (ME) remaining after the energy expended in body heat "heat increment" is deducted, (i.e., NE = ME - heat increment).

NE is further partitioned into the net energy for maintenance (no gain or loss of body weight), growth (or gain in body weight) and lactation (production of milk). The NE requirements for maintenance, growth and lactation are denoted by NE_m, NE_g, and NE_l, respectively.

Net Energy for gain (NEg): NEg is an estimate of the energy in a feed used for body weight gain. Net Energy for Lactation (NEl): NEl is an estimate of the energy in a feed used for maintenance plus milk production during lactation. Net Energy for Maintenance (NEm): NEm is an estimate of the energy in a feed used

to keep an animal in energy equilibrium, neither gaining nor losing weight.

Neutral Detergent Fiber (NDF)

NDF is the residue or insoluble fraction left after boiling a feed sample in neutral detergent solution. The NDF contains plant cell wall components except for some pectins. The NDF is considered a close estimate of the total fiber constituents of feedstuffs since it measures cellulose, hemicellulose, lignin, silica, tannins and cutins. The hemicellulose, cellulose and lignin represent the fibrous bulk of the forage. Because they give the plant rigidity, these three components are classified as structural carbohydrates. Though lignin is indigestible, hemicelluloses and cellulose can be digested by microorganisms in ruminants (e.g., cattle, goats or sheep) or hind-gut fermentation (e.g., horses, rabbits, guinea pigs) as part of their digestive tract. NDF concentration is negatively correlated with dry matter intake

Neutral Detergent Fiber Digestibility (NDFD)

NDFD is the 48-hour in vitro digestible fraction of NDF expressed as percentage of the Neutral Detergent Fiber (NDF) content.

Neutral Detergent Solubles (NDS)

The NDS represents ingredients in a feed sample that are soluble in neutral detergent solution. That means it represents everything that is not NDF. About 98 % of the NDS is assumed to be digestible.

Nitrogen-free Neutral Detergent Fiber (NDFn)

Nitrogen-free NDF (NDFn) is determined as:

- NDFn = NDF – NDFICP (Neutral Detergent Fiber Insoluble Crude Protein)
- Also estimated as NDFn = NDF × 0.93.

Non-fibrous Carbohydrate (NFC) or Neutral Detergent Soluble Carbohydrates (NDSC)

NFC or NDSC represents forms of digestible carbohydrates that are solubilized after boiling a feed sample in neutral detergent solution. These are all forms of non-cell-wall carbohydrates (starch, sugar, pectin and fermentation acids), which are digestible and serve as energy sources. Because NFC includes some other digestible compounds in addition to starch and sugars included in NSC, NFC generally shows a higher value than non-structural carbohydrate (NSC) on the feed analysis report. NFC is calculated as follow:

• NFC% = 100% – [CP% + (NDF% –NDFICP%) +EE% + Ash%]

where EE% is the ether extract% or Fat%.

Non-protein Nitrogen (NPN)

Nitrogen in a feed sample that is not in the form of protein but can be used by the microbial population in the rumen or gastro-intestinal tract to synthesize amino acids and proteins. Common forms of NPN are urea and ammonia.

Non-structural Carbohydrate (NSC)

NSCs are simple carbohydrates, such as starches and sugars, stored inside the cell that can be rapidly and easily digested by the animal. Hence, NSC is considered to serve as an available energy source.

Nutrient

Nutrients are any feed constituent, elements, compounds or groups of compounds that that aids in the support of animal life and nourishment. Also known as constituents of a feed that are capable of being transformed into body

elements are known as nutrients. Common nutrients are: carbohydrates (or energy), proteins, fats, minerals, vitamins and water.

Nutrient Requirements

It refers to the specific amounts of nutrients (energy, protein, fat, minerals and vitamins) necessary to meet an animal's real needs for maintenance, growth, reproduction, lactation or work.

Nutrition

Science involving various chemical and physiological activities, which transforms feed elements (nutrients) into body elements.

Nutrition evaluation

Aassessment of feedstuff for its nutritional adequacy, it can be physical, chemical, biological or microbiological in nature.

Nutritive Value (NV)

Nutritive value refers to a feed's protein, mineral and energy composition, availability of energy, and efficiency of energy utilization.

Nutritive value of feed

An indicator of a feed to its digestibility of dry matter(%). Feeds and fodders can be classified as shown below based on their digestibility.

Above 70% = good

60-70% = moderate

40-60% = low

Less than 40% = very low

Nutritive ratio (NR):

It is the ratio of the digestible protein to the sum of digestible non protein compounds (carbohydrates and fat), the latter being multiplied by 2.25. It is also termed *albuminoid ratio*. It is computed on the fact that protein serves some functions in the animal body, which cannot be done by the digestible non-protein nutrients present in the TDN. Feeds higher in protein have narrow nutritive ratios while feeds lower in protein content have wide nutritive ratio. It is usual to consider that rations with wide nutritive ratio (1:9) are suitable for idle horses and cattle; a medium ratio (1:6) for early fattening, lactation, working animals, etc. and a narrow ratio (1:0.7) for young.

Example 1. Calculate NR of groundnut cake using the following data :

DCP = 42 ; DEE = 6; DCF=l; DNFE = 14.5

NR = (DEE x 2.25 + DNFE + DCF)/ DCP

= ((6 x 2.25) + 14.5 + 1)/42 = 29/42= 0.7

A narrow ratio of 1: 0.7, since groundnut cake is a protein supplement

Example 2: Calculate NR of maize grain:

Maize has 82% TDN and 7% DCP.

NR = (TDN – DCP)/DCP = (82-7)/7 = 75/7 = 10.7

A wide nutritive of 1:10.7. It means that for each kg of DCP, maize contains 1.07 kg digestible non-protein nutrients

Palatability

Palatability refers to the acceptability of feedstuffs to an animal. Palatability is affected by the feed's odor, texture, moisture, physical form and temperature. Palatability is a plant trait that can be measured when the animal has the opportunity to selectively feed.

Parts Per Million (ppm)

Parts per million is a unit of measurement used to state the concentration of specific nutrients, compounds or elements present in small quantities in a feedstuff (e.g., 1 ppm = milligrams per kilogram (mg/kg), 1 pound per million pounds, 1 milligram per liter (mg/L), or 1 microliter per liter (μL/L).

pH

pH is a measure of acidity or alkalinity. Values ranged from 0 (most acidic) to 14 (most alkaline or basic). A pH value of 7.0 is neutral. The values give the negative log of the hydrogen ion concentration.

Pectin

Pectin is an intercellular polysaccharide that functions as cellular glue. Like the nonstructural carbohydrates, it is easily degraded in the rumen. Unlike the NSCs, though, it does not lower rumen.

Protein

Protein is an essential nutrient. Proteins are composed of long chains of various kinds of amino acids. Animals cover their protein needs by breaking down plant and microbial protein and reassembling them as animal proteins.

Proximate Analysis

Proximate analysis is a chemical method that separates, identifies and quantifies the major categories of compounds in a mixture. In feed and food analysis, it serves as a tool for assessing and expressing the broad nutritional value of a feed sample.

It is important to remember that proximate analysis is not a nutrient analysis. Rather, it is a partitioning of both nutrients and non-nutrients into categories based

on common chemical properties. The system consists of the consecutive steps of separations and determinations of six categories of components and expressing the percentage of each that is present in a feed sample (Figure 2):

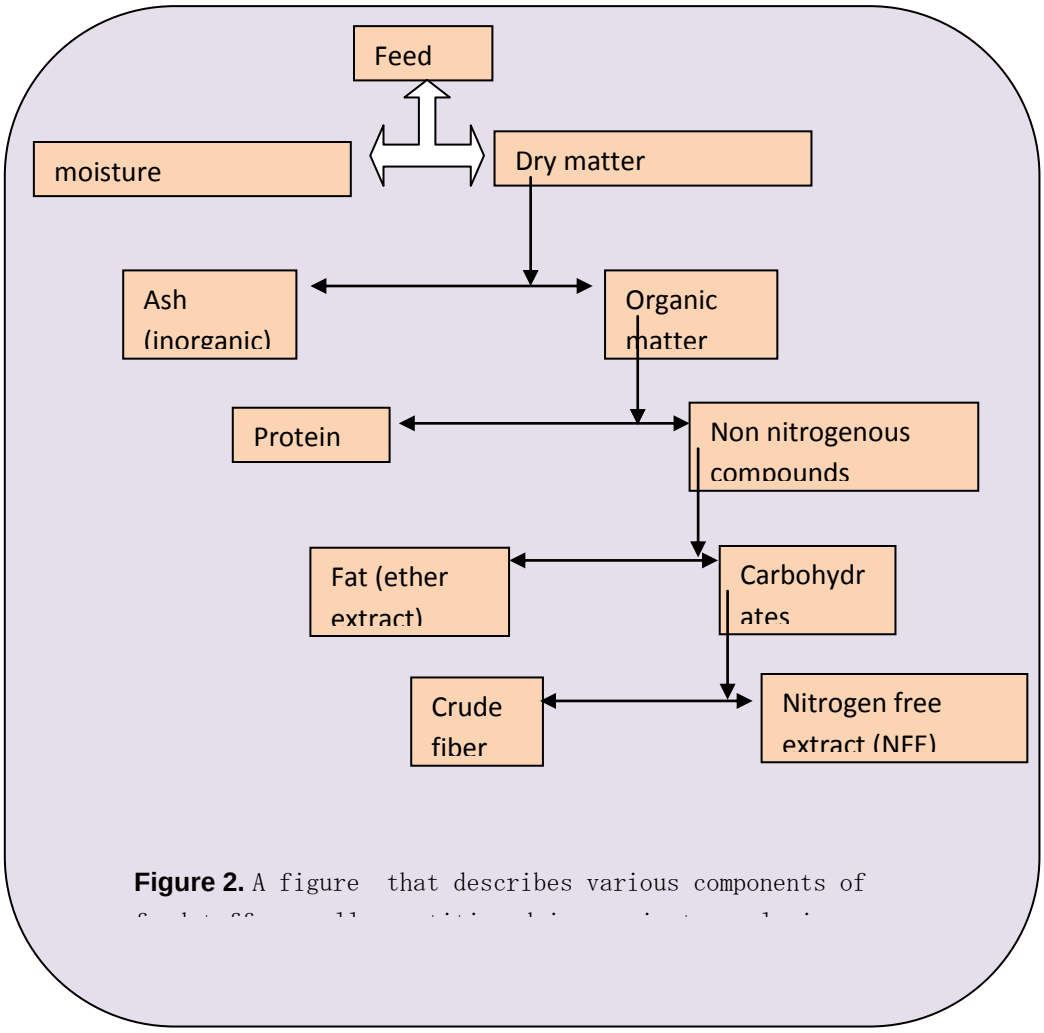


Figure 2. A figure that describes various components of

- Water/moisture (or dry matter)
- Ash (minerals)
- Total or crude protein (total nitrogen×6.25)
- Total or crude fat (or ether extract)
- Crude fiber (incompletely digested carbohydrates)
- Nitrogen-free extract (readily digestible carbohydrate)

Ration

Ration refers to the 24-hrs feed allowance for an individual animal.

Ration Formulation

The art of combination and mixing in specific ratios of feed ingredients/feedstuffs to obtain mixture for the nutrient requirement of farm animals.

Relative Feed Value (RFV)

Relative feed value is a forage quality term that is used to rank feeds, especially forages, according to their nutritive value. This ranking consider nutritive value of full bloom alfalfa hay as standard. Full bloom alfalfa hay, containing 41 % ADF and 53 % NDF on a dry matter basis, has an RFV of 100 and is considered to provide the average score. The RFV has no units, it compares the potential of two or more forages on the energy intake basis. Thus, it consider an index of forage quality for comparing forage lots. Forages with RFV greater than 100 are of higher quality than full bloom alfalfa hay, and forages with a value lower than 100 are of lower value than full bloom alfalfa. Such a single suitable parameter is useful for practical forage pricing and marketing. The RFV is calculated based on two parameters: NDF and ADF. NDF is used as an indicator of forage intake and ADF is used as an indicator of digestibility. Thus, together, ADF and NDF estimate intake potential and digestibility, and calculate as:

• RFV = DDM (% of DM) × DMI (% of BW) ÷ 1.29

• Where, DDM (digestible dry matter) and DMI (dry matter intake) can be calculated from ADF and NDF as:

• DDM (% of DM) = 88.9 – 0.78 × ADF (% of DM)

• DMI (% of BW) = 120 ÷ NDF (% of DM)

Due to the variability of measuring ADF and NDF, absolute RFV numbers

are not recommended for making direct comparisons. Rather, a range of RFV values (±5 points of the target) is a more reasonable way to classify a forage (e.g., if an RFV of 140 is desired, any forage with an RFV of 135 to 145 should be considered to have an equivalent value). One of the limitations of the RFV system is that it assumes constant relationships between NDF and intake, and between ADF and digestibility. However, two forages can have identical NDF levels but very different digestibilities and, therefore, intakes. This often results in the RFV of high-quality forages being underestimated because their intake is underestimated.

Relative Forage Quality (RFQ)

RFQ is a forage quality term, used to rank forages according to their relative nutritive value. RFQ shares many of the properties of RFV (e.g., its basis of comparison is 100, the typical nutritive value of full bloom alfalfa hay; it has no units; it compares the potential of two or more like forages on the basis of energy intake; it serves as a useful index of forage quality for comparing forage lots; and it is very useful for practical pricing and marketing of forage lots). Unlike RFV, however, RFQ takes into account digestible fiber. RFQ is based on intake and true TDN. This makes RFQ a better predictor of forage quality than RFV. This is because RFQ accounts for NDF digestibility (NDFD) and the contribution of other nutrient fractions when calculating TDN, rather than calculating DDM based on ADF. RFQ is calculated

as:

• RFQ = DMI (% of BW) × (TDN (% of DM) ÷ 1.23

The equation for RFQ includes the correction factor 1.23, which allows the RFQ to retain the value of 100 for full bloom alfalfa (similar to RFV), which serves as the base value. The equations used to calculate DMI and TDN for

legumes and legume/grass mixtures are specific to those forages and are different from those used to calculate DMI and TDN for warm and cool season grasses. Proper identification of forage type will therefore be essential before RFQ calculation. The two recommended equations for DMI and TDN calculations depend on whether or not the primary forage is legume or grass and are explained in the definition of "Dry Matter Intake" and "Total Digestible Nutrients"

Roughage

A bulky and/or coarse feed high in fiber (greater than 18 percent crude fiber) but lower in energy. For example, forage, hay, silage and haylage are sometimes called roughage.

Rumen

The rumen is a large, hollow muscular organ that is the site of most of the fiber digestion that occurs in ruminant animals, such as cattle, sheep and goats.

Ruminal Microbes

Ruminal microbes include the whole community of microorganisms present in the rumen. They accomplish the degradation or fermentation of feed. An estimated 150 billion microorganisms per teaspoon are present in the fluid of the rumen. This microbial community consists of bacteria, protozoa and fungi.

Ruminants

Ruminants are a group of animals that have multiple organs working together to accomplish digestion. The digestive tract consists; the reticulum (involved in rumination and in passage from the rumen to the

omasum); rumen (large compartment used for fermentation); omasum (once

called the manyplies, it removes excess liquid and nutrients moving out of the reticuloomasal orifice); and abomasum (acid-pepsin digestion similar to a monogastric), where monogastric animals (e.g., horse) have a simple or single-chambered stomach that utilizes an acid-pepsin digestion process to extract nutrient from the ingested food.

Rumen Degradable Protein (RDP)

RDP is also known as Degradable Intake Protein (DIP). See DIP.

Rumen Undegradable Protein (RUP)

RUP is another name of by-pass protein, escape protein or undegradable intake protein (UIP). See By-pass Protein.

Silage

The feed preserved by an anaerobic fermentation process (e.g., corn silage, haylage, high moisture corn) in which lactic acid and volatile fatty acids (produced by fermentation of lactic acid producing bacteria) lower the pH of the silage. The low pH preserves the silage, when it reach 4. See Ensiled.

Silage Additives

Silage additives refer to the mater aialsdded during the ensiling process to enhance production of lactic acid and/or a rapid decrease in pH of the feed.

Soluble Intake Protein (SIP)

SIP includes the non-protein nitrogen and that part of true proteins which are readily degraded to ammonia in the rumen. They are used to synthesize microbial protein in the rumen.

Starch

Starch is an intracellular carbohydrate found primarily in the seed and/or root portions of plants. Starch is the main energy source.

Structural Carbohydrates

Structural carbohydrates are the complex carbohydrates that form the plant cell wall and involve cellulose, hemicellulose, lignin and pectin. They are typically measured in the laboratory as neutral detergent fiber.

Supplement (feed additives)

A supplement feed or feed mixture is used to improve the nutritive balance of the ration. A supplement is rich in one or more of protein, energy, vitamins or minerals, and, in combination with the base feeds, produces a more complete balanced feed.

Total Digestible Nutrients (TDN)

TDN is a measure of the quality of a feedstuff. Nowadays, TDN values are calculated, not measured. Formulas for calculating TDN originally were based on ADF and varied by region and the nutritionist doing the calculation. Their procedure is based on the assumption that forage classes have more uniform and predictable digestion coefficients. So, it could be assumed that the TDN for alfalfa, clovers and legume/grass mixtures be calculated as follows:

- TDNlegume = (CP × 0.93) + (FA × 0.97× 2.25) +[NDFn × (NDFD ÷ 100)] + (NFC × 0.98) – 7

Where:

- NDFn = nitrogen free NDF = NDF – NDFICP, also estimated as NDFn = NDF × 0.93
- NDFICP = neutral detergent fiber insoluble crude protein
- NDFD = 48-hour in vitro NDF digestibility (% of NDF)

• NFC = non fibrous carbohydrate (% of DM) = 100– (NDFn + CP + EE + ash)

• EE = ether extract (% of DM)

The TDN for warm and cool season grasses is calculated as:

• TDNgrass = (NFC × 0.98) + (CP × 0.87) + (FA × 0.97 × 2.25) + [NDFn × (NDFDp ÷ 100)] – 10

Where:

• All other terms are as defined previously and NDFDp = 22.7 + 0.664 × NDFD

The most significant issue with TDN is that it does not account for additional energy losses, particularly heat increment and, to some extent, gaseous losses, especially regarding ruminant systems. Consequently, TDN is known to over-estimate the energy value of roughages compared to grains.

Total Mixed Ration (TMR)

A total mixed ration is a mixture of mechanically mixed ingredients that typically contains both roughages and concentrates such as grains to optimize animal performance. TMRs are commonly used in large dairy ranches or beef feedlot operations.

Undegradable Intake Protein (UIP)

By-pass protein, escape protein or

Rumen Undegradable Protein (RUP)

The use of UIP is consider a misnomer since it generally refers to material that is not degraded in the rumen but is degraded in the abomasum and so is not truly “undegradable.” See By-pass Protein.

Vitamins

Vitamins are compounds that typically act as parts of enzyme systems essential for many metabolic functions.

Water-soluble Carbohydrates (WSC)

The carbohydrates that can be solubilized and extracted in water. WSCs include mono-saccharides, disaccharides and some short chain polysaccharides, mainly fructans, which are consider a major storage carbohydrate in some cool season grasses.

"Wet Chemistry" Analysis

Wet chemistry is a term that refers to a number of techniques involving direct analyses with solvents, acidic or basic solutions, other chemicals and other traditional methods used to analyze feed samples. The procedures are based on chemical and biochemical principles, but require more time to complete than the newer methods. Wet chemistry is the basis for all modern, instrument-based, analytical methods and for calibration of new dry methods. It is the most accurate methods for determining nutrient values of feeds/forages and are frequently used for quality assurance purposes or in the development of new techniques/calculations.

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SAMPLING

Samples for Analysis

Scope: These procedure steps are used to obtain samples for analysis (different methods are used for hay and silage).

Principle: The accuracy of analysis depends on accuracy of the sent sample to the lab. It is important that the sample represents the composition of the "lot" of sampled; otherwise analysis will be without value.

A "lot" is a hay or silage taken from:

- Same feedstuff. (pure or mixed) & variety;
- Same pit/bunker;
- harvested (collected) within 24-48hrs.

a "lot" is influenced by other factors include:

- Environmental (rain damage, humidity, wind, etc.);
- Field (weed content, other crop, toxic plants, etc.);
- Soil type;
- Treatment after harvest; and
- Storage condition.

A "lot" of cubes or baled hay should be 150- 200 tonnes.

Equipment: Representative samples can be obtained with a probe or core sampling device, (Do not rely on a handfuls from one bale).

- A corer can be made from 2-3cm steel tubing, with at least 45cm long with a slightly scalloped and sharp cutting edge.
- Corers are driven using either a hand brace or by a continuously variable speed, electric drill (preferred and practicable). A portable generator is useful, when many samples are to be taken.

Sample Size Reduction:

The original sample will reduced in size before further processing. Two ways can be achieved, by use of a riffle box or by sample coning and quartering. The riffle box is preferred for fine chopped maize forage, because it is faster and yields a better sub-sample.

***Riffle Box**:* This tool also known as sample splitters, where it split sample into two equivalent portions via a series of openings. Half is usually discarded, and the other remains for analysis, or for further reduction.

Coning and Quartering: This procedure used to reduce the size of a sample to a convenient amount prior submission to the lab.

Involves the following steps:

a) Spread the sample over a clean surface and mix well to ensure good distribution of leaf and stem;

b) Fold the outer surrounding back into the middle (form a Cone or Mound);

c) Divide this "Cone" into equal quarters;

d) Select two diagonally opposed quarters and repeat this process until get the correct sample weight. Discard the unselected quarters. Transfer this sub-sample to a suitable container for sending to the lab.

Sampling Procedure: HAY

1. Small square bales:

a) Sample between 15- 20 small bales, selected at random from the "lot".

b) Take single core from each bale selected, probing at the center and at right angles to the surface.

c) Gather the cores into one sample in a bucket and mix well. Keep the whole sample intact (not subdivide now).

2. Large round or square bales:

a) Sample (8-10) large bales, selected randomly from the lot.

b) Take single core from each side of all selected bales, probing at right angles to the surface at different heights.

c) Gather the cores into a one sample in a bucket and mix well. Keep the whole sample intact (not subdivide now).

3. Cubes or pellets:

a) Select a handful of cubes or pellets from at least 5 locations from the whole lot.

b) Gather the sub-samples in a bucket and mix well.

c) Use "Riffle Box" or "Coning and Quartering" procedure to reduce sample size to at least 250g.

Sampling Procedure: SILAGE

Silage sample preferred to be taken at least 3 weeks after ensilaged process and practically at the time of feeding.

1. Bunker silage:

a) Closed bunker- Obtain samples for analysis using a coring device that goes deeply into the bunker. (At least 5 locations to be representative sample).

b) Opened bunker - Take random samples from minimum 10 locations across a freshly cut surface of the stack, (this not provide a good representative sample as multi-site coring).

c) Gather the multiple samples into one sample in a bucket and mix well. (The whole sample not subdivide at this stage).

2. Wrapped baled silage

a) Sample up to 10 large bales at random using a coring tool in the same manner as for large hay bales.

Note: Great care is taken to reseal the holes made in the plastic by the corer.

b) Gather into a single sample in a bucket and mix well. (Not subdivide now).

Sample Handling: As soon as possible, use the "Coning and Quartering" procedures to minimize the sample size. The sample must be placed in a plastic bag and tightly sealed to expelled air. This to ensure, that the lab sheet of dry matter will reflect the actual DM content of the lot when it was taken.

Sample Dispatch: Samples must be delivered to the lab as soon as possible after being collected. If silage samples be delayed to reach the lab, more than 12hrs, it must be frozen at once. (It is important during hot weather.)

Ensure to follow the laboratory's instructions for identifying samples and filling out all the required details on the submission tag.

Preparation of Fodder Samples for Analysis

Scope: This procedure is used to prepare samples for lab analysis.

Principle: homogeneous samples are essential to achieve representative analytical results. This generally involves grinding and/or drying. Most samples received at a lab fall into one of the following categories:

a) DM 90 to 95%, grind and analyze immediately;

b) DM too wet, coarsely ground (to pass a 4mm sieve) but to be finely ground; or

c) Those samples, (need to be partially dried), can be ground coarsely (DM less than 85%). This wet sample (less than 85% DM) is dried at 55 to 60oC in a forced hot air oven not more than 24hr to reduce water content before grinding.

Each sample type differs in procedure of handling. Most forage can be milled to pass a 4mm sieve pores using a cutting mill at 80 to 85% dry matter without sticking in the mill, loss of moisture. However, when using a cyclone mill to grind forage samples to pass a 1mm sieve pores, DM need to be 90 to 95% for proper grinding.

Samples with DM 85% or greater, that are too large to grind in their entirety are first ground with a large mill to pass a 4mm screen. The coarse ground sample is then decreased in size by Coning and Quartering procedure. When necessary, the reduced sample is partially dried then ground again to fit our desire for analysis.

-Care should be taken at sample preparation to avoid contamination with other samples.

-Sample should be kept in airtight containers and away from heat and light.

Equipment:

Forage chopper.

Cyclone pulverizing mill - 1mm screen (NIR).

Cutting mill - 4mm screen.

Sample container.

Freezer and refrigerator.

Personal precautions:

- The noise level is hazardous, so wear hearing protector.
- Dust, so wear a dust mask to avoid inhaling.
- Operating instructions and safety should be observed.
- Avoid inserting fingers or objects into the mill.

Procedure:

1) At samples >85% DM, grinding:
    a) Remove sample from shipping bags and discard roots and brush off dirt materials.
    b) Identify and report any material discarded from the sample and any other sample handling.
    c) Chop samples into 2cm long pieces using either hand shears or the lab forage chopper. Include any ears attached to maize plants. (Not necessary for silage samples or cored hay).
    d) Grind whole sample to pass 4mm screen in large mill.
    e) Reduce the grind sample by coning and quartering to the desired amount.
    f) Kept to airtight container, seal and label as the coarse sample.

g) Grind the coarse substances in a cyclone mill fitted with a 1mm screen to have a fine ground material suitable for analysis.

h) Mix the ground materials. Transfer to airtight container, seal and label as the final ground sample.

2) At sample < 85% DM, ('Wet' samples) grinding:

a) Remove sample from shipping bag and remove roots and brush off dirt materials.

b) Identify and report any material discarded from the sample and any other sample handling.

c) Chop samples of into approximately 2cm pieces using either hand shears or the lab forage chopper. Chop stalk and maize cob pieces to help drying, besides any ears attached to maize plants.

d) Place the chopped sample on a clean surface and mix well. If the whole sample cannot be dried, reduce the sample size.

e) Care about that representative ratios of stem and leaf occur when reducing size.

f) Transfer reduced sample to a tared container for determination of partial dry matter (Method 1.3R, Step 1).

g) Store the remainder in a refrigerator as a reserve (seal and label) until sample preparation is complete.

h) Dry the reduced sample using the forced hot air oven.

i) Grind the partially dried sample with a 4mm screen of large mill.

j) Reduce the sample by coning and quartering to the desired amount.

k) Transfer to airtight container, seal and label as the coarse sample.

l) Grind the coarse substances in a cyclone mill fitted with a 1mm screen to have a fine ground material suitable for analysis.

m) Mix the ground materials. Transfer to airtight container, seal and label as the final ground sample.

Comments:

- Using NIR, samples must be dried and ground by the same method.
- Equipment should be cleaned to avoid samples contamination.
- avoid sample heating during grinding process. Heating affect chemical analysis.

- Equipment should be maintained on a routine schedule. Condition of the sieves, rotors, blades or grinding surfaces should be monitored and recorded in a designated maintenance lab book.
- Do not leave portions from sample in mill.
- Store samples in airtight containers away from heat and light.

Method for Silage Samples

Scope: Silages contain a volatile DM portion that is lost with oven drying and freeze drying. This fraction contains VFA, alcohols and volatile nitrogen compounds. The loss of it varies with the extent and type of fermentation, and is commonly greater with low DM silages where there is usually a more extensive fermentation. With higher DM silages (> 50 % DM) there is a restricted fermentation so the volatile loss on oven drying is less.

As the volatile compounds are completely digestible, it is important to take this into account when estimating digestibility, otherwise it will be underestimated. Owing to volatile N losses, the total N content of silages will be underestimated if analyses are conducted on oven dried samples. All other analyses on silages will be overestimated unless a correction is made for the volatile DM content.

Principle: A laboratory method that specifically determines water content is needed to allow the calculation of the true DM content of silages and other fermented feeds. The Karl Fischer method has been found to be suitable. The method involves the extraction of silage moisture in methanol, and then determining water content of the extract using a Karl Fischer titrator.

The above procedure is less appropriate for commercial feed testing laboratories. So, an alternative procedure is to use a correction equation to predict actual DM from an oven DM. As the correction required will be influenced by the oven drying regime performed.

Procedure:

The correction equation calibrated actual DM against oven DM determined (dried) at 80ºC for 24 hours, **one-stage method**, and at 60ºC for 24 hours, **first stage of two-stage method**, (**Partial DM**).

(a) Correction for silages dried at 80ºC

Estimated Actual DM (%) = 3.846 + (0.96 × oven DM %)

(b) Correction for silages dried at 60ºC

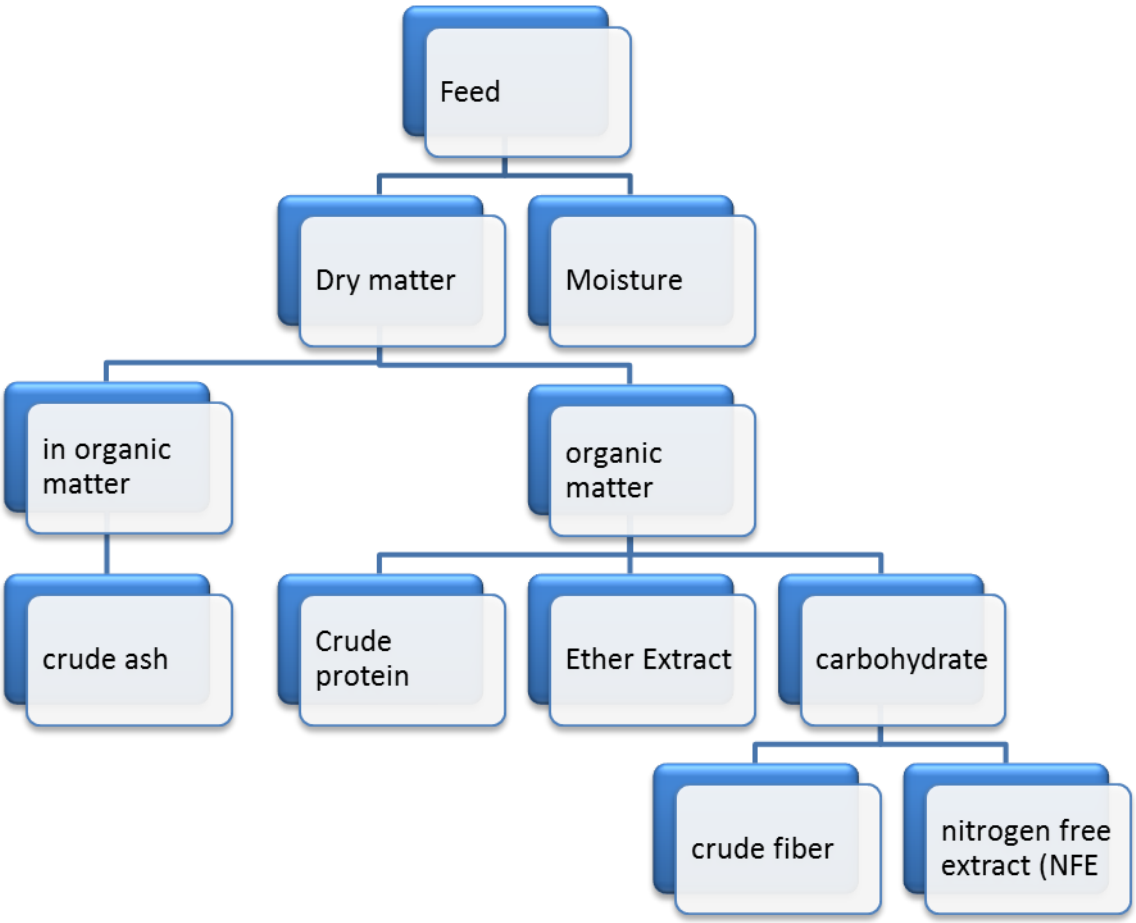
Estimated Actual DM (%) = 4.686 + (0.89 × oven DM %)

Proximate composition and detergent fiber

Summary

Proximate composition is the term usually used in the field of feed/food and means the 6 components of moisture, crude protein, ether extract, crude fiber, crude ash and nitrogen free extracts, which are expressed as the content (%) in the feed, respectively.

The composition as the feed is classified as shown below:



The measured values of these 6 components in feed are important factors to understand the nature and the properties of the subject feed.

The 6 components in the feed and substances contained in them are listed in the table shown below:

Proximate analysis composition			Substances
Moisture			Water and Volatile substances
Dry Matter	Organic matter	CP	Protein, AA, NPN
		EE	Fat, complex lipids, sterols, FA, fat soluble pigments and vit.
		CF	Cellulose, hemicellulose, lignin
		NFE	Soluble carbohydrate, pectin, hemicellulose, lignin, organic acids, tannin, water-soluble dyes
	Inorganic matter	Ash	Pure ash, organic residue, soil

1. Moisture

The ingredients from the view of nutritionist are present in the dry matter part; therefore the moisture content is an important factor in both economy and storage. At high temperature and humidity in Egypt, the risk of growth of molds, etc., or self-digestion by enzymes in the feed when water in the feed, is more than about 15 %. For that reason, the moisture content in formula (mixed feed) distributed in Egypt is usually less than 10- 13 %.

Volatile matter

The feed measures loss on drying by heating at normal pressure as moisture, the result includes most of volatile substances other than H_2O. Therefore, it may be more precisely to be referred to as volatile matter rather than moisture for accuracy.

Organic acids like lactic acid, butyric acid and acetic acid in silage as well as ammonia and flavor components in feed are also vaporized and thus overestimated the value of moisture. Because of the low content of these in the feed, it has hardly been a need to consider their influence on the measured value. However, in silage, etc., with high moisture content, contents in solid matter may be slightly affected according to the content of volatile acids.

When the feed is spread on flat sheet and left, moisture absorption or release happens, and according to a constant relative humidity comes to equilibrium at the moisture content (equilibrium moisture content), which indices comparatively stable moisture content of the feed in the air-dry basis. The equilibrium moisture content differs by the kind of the feed and affected by materials such as salt when mixed in the feed. When the annual mean RH in Egypt is presumed to be 70 %, the moisture content that is at equilibrium with it is in the range of 13-15 % for most of feeds.

Precautions for the procedure (moisture estimation):

(1) Check if the thermometer is acting well.

(2) Errors may be caused when overcooling for the weighing dish.

(3) Errors may be caused by the location of the weighing dish in the dryer.

(4) A rough standard for the analysis value can be obtained when the standard sample is analyzed.

Methods of determination

1- Determination of both water and Dry Matter content

Scope: This procedure is used to determine the DM% of all fodder except silage. Silage loses volatile compound during the drying.

Principle: Three steps are involved in the determination of %DM:

Step 1: Wet (< 85% dry matter, is dried at 55 to 60oC in a forced air oven for a maximum of 24 hours to reduce moisture content prior to grinding. The loss of moisture is recorded partial DM%. This has minimal effect on composition. Samples containing < 15% moisture are ground as received.

Step 2: Ground material is dried at 105oC in a forced air oven for 2-4 hours. The loss of moisture is recorded (water %).

Step 3: Calculation of Total Dry Matter %

Total dry matter % (%DM) = { (Partial DM%) X (Lab DM%) } /100.

Equipment:

Step 1 - Partial dry matter %

Forced hot air oven set at 55 to 60oC

Digital balance, accurate to 0.01g

Containers to hold 100 to 250g coarse sample

Recording sheets / Computer

Step 2 - Laboratory dry matter %

Digital balance, accurate to 0.1mg

Aluminium moisture dishes

Forced hot air oven, 105°C

Desiccator

Tongs

Recording sheets / Computer

Procedure:

Step1. Partial dry matter %

a) Weigh empty container on a balance and label the container and weight (W1) to nearest 0.01g.

b) Tare balance against empty labeled container.

c) Add (100 - 200g) coarse forage to the container with depth of 3.5cm. Record sample number and wet weight to nearest 0.01g (W2).

d) Dry in a forced hot air oven at 55 to 60oC for 16 to 24 hours.

e) Remove container from oven and cool for at least 45 minutes before weighing the sample and container. Record the weight to the nearest 0.01g (W3).

Calculation:

Partial dry matter% = { (W3 - W1) / W2} X 100%

Where: W1 = Empty weight of container in grams

W2 = Initial weight of sample in grams

W3 = Dry weight of sample and container in grams

Step 2-Laboratory dry matter %

Note: Cotton gloves should be needed when handling moisture. (to avoid contamination of the moisture dishes with sweat and grease).

a) Dry aluminium dishes, at 105oC for at least 2 hours before use.

b) Cover dish with lid and cool in a desiccator to room temperature.

c) Remove dishes one at a time from the desiccator and weigh dish (plus lid) to 0.1mg (W4).

Record the dish number and weight against sample number.

d) Immediately weigh (to 0.1mg) about 2.0g of well-mixed sample. Record the weight (W5).

e) Shake the dish gently to distribute the sample evenly and ensure the maximum surface area for drying.

f) Place lids on each dish during transfer to the oven.

g) At the oven, remove the lids and place the dishes in upturned lids in the oven.

h) Leave the dishes in oven for 3 hours at 105oC.

i) Remove the dishes from the oven, quickly replace lids and transfer to a desiccator.

j) Cool for at least 30 minutes but not more before weighing. Record the weight (W6) to 0.1mg.

Calculation:

Laboratory dry matter % (Lab DM %) = { (W6 - W4) / (W5 - W4) } X 100%

Where: W4 = Empty weight of container

W5 = Initial weight of sample

W6 = Dry weight of sample and container

Step 3: Calculation of Percent Total Dry Matter

Total Dry matter % (%DM) = { (Partial DM%) X (Laboratory DM%) } /100.

= {(W3 - W1) / W2} X { (W6 - W4) / (W5 - W4)} X 100%

N.B The temperature should be 105±2 °C, and the time should be 3hrs for fish soluble adsorption feed, molasses adsorption feed, gluten feed and corn distillers dried grains with soluble.

Precautions and Notes

[1] The definition of water content varies according to the analysis purpose; moisture mainly means water of adhesion. The temperature

distribution in the temperature-controlled dryer commonly used differs widely depending on the place, but the range is comparatively smaller in the forced air-flow oven. Care should be taken with a light sample, where may be blown off depending on the location in oven.

[2] Water content in grains changed by grinding. In order to grind decrease moisture change as much as possible, a rough grinding is preferred, such as a manual roller mill. Care should be taken for the storage of the sample after grinding.

[3] A glass weighing dish can be used, but an aluminum dish is more preferred, because it is less fragile and lighter, shows better thermal conductivity, better airtightness, and is easier to handle.

Put the sample in the dish, and put it in oven with the lid below or at the side of it. After 2 hours of drying, cover the container with the lid, and let it cool in a desiccator. We recommended the use of cotton gloves, because the dish is hot.)

It is convenient to mark the lid and the dish with the same number.

[4] Collect the sample, and weigh together with the lid.

[5] A desiccator ≥ 20-22 cm in the diameter of the platform is preferred. Silica gel, calcium chloride, or phosphorus pentoxide, can be used as a desiccant. Silica gel is considered to be a common desiccant because it is easy to handle and regenerate; as, it should be re-dried to be used when the blue color of cobalt salt, the indicator of moisture absorption, fades even slightly. Re-drying should be conducted at 130-140 °C for 2-3 hours.

Hygroscopicity is reduced by the adsorption of fat and oil, etc., to silica gel. To minimize the error due to cooling, it is recommended to keep the cooling duration in a desiccator to be constant (45 minutes), and to always put not more than 8 dishes in a desiccator. The number of dishes to be contained

in a desiccator is preferably not more than 8 because errors are likely to occur between the start and the end of weighing when a large number of dishes are contained in a desiccator.

[6] For fish soluble adsorption feed, molasses adsorption feed, gluten feed and DDGS, the method was modified as "drying at 105±2 °C, 3 hours" because of vaporization or heat decomposition of volatile substances other than moisture if the normal method is applied to them.

The assay of moisture in muciform feed such as fish soluble and molasses is usually conducted by the following method:

Weigh accurately 2 g of a sample, put it in an aluminum dish (put 10-20 g of sand and a stirrer bar in it, dry, and weigh in advance), mix the sample and sand on a boiling water bath, and then dry for about 15 minutes stirring occasionally.

Then put it in a temperature-controlled oven, dry at 105±2°C for 3 hours, let it stand to cool in a desiccator, and then weigh, to calculate the moisture content based on the loss.

* Use sand (silica sand) of 350-250 μm (60-80 mesh). Wash sand with water, heat in hydrochloric acid (1+1) for a few hours, wash with water until there is no acid, dry and store in a desiccator.

Toluene distillation or loss on heating at 100 °C for 20hrs may be useful for a sample with a high content of volatile components such as silage. For a viscous liquid, adsorb it on filter paper and dry to calculate the moisture content by the loss.

[Other analysis methods]

2. Moisture assay methods

Sample	Weigh accurately about 5 g, and put in a container. Container, made from non-

	corrosive metal or glass with a sealable lid, with surface area of about 0.3 g/cm^2.
Drying	Dry at 103 °C for 4 hours using an electric dryer.
Cooling	Desiccator
Weighing	Weigh accurately to the order of 1 mg.

3. Distillation

Heat a sample in an organic solvent (toluene) immiscible with water, water in the sample or the mixed vapor of water and the solvent, cool it and calculate water in the sample based on the volume of water separated from the solvent. This is applicable to a thermo-stable sample with volatile components other than water as well as fat.

4. Kahl Fischer method

The method determine moisture in a sample utilizing the specific reaction of Kahl Fischer reagent, which contains iodine, sulfur dioxide and pyridine, with water under the presence of methanol, and is consider as the volumetric method and electrometric titration method.

The method is advantageous in that water alone can be quantitated when the sample contains volatile components other than moisture.

5. Heating furnace control method

A new type of thermogravitic analytical instrument that employs the heating furnace control method can measure 19 samples simultaneously and can quantitate ash content in addition to moisture.

6. Other methods

When the sample thermo-stable, dry under constant temperature and reduced pressure such as “60-70 °C, 26.7-33.3 kPa,” and the loss is obtained as the amount of moisture. Also, **a moisture meter** that employs infrared radiation can be used as field tool.

Methods of analysis of DM			
Procedure	*Applicability*	*Limitations*	*Capital costs*
Physical removal of water			
Air oven at 100 105ï¿½C	Most foods, except those rich in sugars and fats	Caramelization of sugars, degradation of unsaturated fats, loss of other volatiles	Low
Vacuum oven a 60ï¿½C	Most foods	Loss of volatiles	Low
Freeze-drying	Most foods	Slow, residual water in samples	Medium
Microwave ove	Medium or high moisture	Charring	Low
Dean & Stark distillation	Foods high in volatiles	Safety of solvents used	Low
Chemical reactivity			
Karl Fischer	Low moisture, hygroscopic foods		Low
Physical methods			
NMR	Most foods	Need for calibration with specific food	High
NIR	Established for cereals an some other foods	Need for extensive calibration with specific food. Particle size dependence	High
Chromatography			
GLC	Meat and meat products		High
GSC	Some meat products		High

2. Crude Protein (CP)

Summary of crude protein

Crude protein is the value obtained by estimating nitrogen in a sample by the Kjeldahl method and multiplying the result by the factor 6.25. Therefore, crude protein includes ammonia, etc., that are not of protein origin. Generally the nitrogen content of protein is 16 % on average; thus the factor is 100/16 =6.25. However, the factor is different between samples (5.83 for flour; 5.95 for rice), the crude protein of some feeds is different from the pure protein content; crude protein is measured to be excessively small in materials of milk product origin such as casein, and excessively large in flour and soybean.

Summary of the Kjeldahl method:

Add concentrated sulfuric acid to a sample and heat, and then degradation and oxidation/reduction occur simultaneously to turn nitrogen into ammonia, which is present in the form of ammonium sulfate in concentrated sulfuric acid. Dilute it with water, add excess of concentrated sodium hydroxide and heat, and then ammonium sulfate is degraded into ammonia that is distilled. Transfer the ammonia into the sulfuric acid standard solution of a constant concentration, and titrate excess acid with the sodium hydroxide standard solution to obtain nitrogen content. The chemical reaction in the degradation by concentrated sulfuric acid is considered to be as follows:

(1) Organic compound is dehydrated and carbonized by sulfuric acid to produce carbon compounds.

(2) Carbon compounds are degraded into carbon and hydrogen by hydrolysis.

(3) Carbon and hydrogen are oxidized into carbon dioxide and water to be driven out.

(4) Sulfuric acid is reduced by a part of carbon into sulfurous acid, and carbon turns into carbon dioxide.

(5) Sulfurous acid reduces nitrogen into ammonia, and itself turns into sulfuric anhydride.

(6) Hydrogen produced by the degradation of carbon compounds facilitates the formation of ammonia.

(7) Ammonia formed immediately combines with sulfuric acid to become ammonium sulfate.

(8) When potassium sulfate is added as an oxidizing agent, the boiling point is elevated and the effect on organic compound becomes more active.

The combustion method is added to the Analytical Standard of Feed in 2006. The combustion method is also called the modified Dumas method and is employed as the AOACI method and the ISO (International Organization for Standardization) method, and is frequently used in foreign countries. The measurement principle is to combust a sample in oxygen at high temperature and to quantitate freed nitrogen by a thermal conductivity detector. The combustion method is characterized in the following points:

(1) Rapid quantitation is possible with measurement time of 5-10 minutes per sample.

(2) No draft is needed because there is no need to degrade a sample. Also, environmental load is smaller because deleterious or toxic substances such as concentrated sulfuric acid and copper sulfate are not used.

(3) The analysis instruments can be easily operated.

As the combustion method is used worldwide instead of the Kjeldahl method, it is supposed that the combustion method will also become the main stream of crude protein analysis in Japan.

Determination Methods

1. Kjeldahl method

Scope: This enables the determination of all protein nitrogen (N) and most non-protein N in all types of feeds. Silage requires special procedures.

Principle: This method involves:

Step 1: Digestion of the sample in concentrated sulphuric acid with a catalyst. This results in the conversion of susceptible nitrogenous compounds to ammonium sulphate.

Step 2: The resultant solution is then neutralised with excess caustic soda. Ammonia is liberated by steam distillation and captured in boric acid. The borate formed is titrated with dilute sulphuric acid. Depending on equipment chosen, titration can be fully automated or carried out manually.

Application: The following Equipment, Reagent and Procedure Sections provide an accurate description of the Kjeldahl process based on block digestion, automated steam distillation and manual titration.

A. Reagent preparation

1) 0.1 mol/L sodium hydroxide standard solution

Prepare a saturated solution of sodium hydroxide, close the cap, leave at rest for not less than 10 days, and to 50 mL of the supernatant, add boiled and cooled water to be 10 L to prepare the 0.1 mol/L sodium hydroxide standard solution. Moreover, standardize its concentration by the following procedure:

Weigh accurately 2-2.5 g of amidosulfuric acid (standard reagent) (dried in a desiccator (vacuum) for 48 hours), put it in a 250-mL volumetric flask, add water to dissolve, and further add water up to the marked line to prepare the amidosulfuric acid standard solution. Transfer accurately 25 mL of the amidosulfuric acid standard solution into a 200-mL Erlenmeyer flask, add a few drops of bromothymol blue test solution, titrate with the 0.1 mol/L sodium hydroxide standard solution, and calculate the factor (f_1) of the 0.1 mol/L sodium hydroxide standard solution by the following formula:

$F1= (w*10^4)/(v*97.1)$

W: Weight (g) of amidosulfuric acid in the amidosulfuric acid standard solution (25 mL) used for standardization

V: Volume (mL) of the 0.1 mol/L sodium hydroxide standard solution required for titration 2) 0.05 mol/L sulfuric acid standard solution

Add 28 mL of sulfuric acid to 1 L of water gradually with stirring, let it stand to cool, and then add water to be 10 L to prepare the 0.05 mol/L sulfuric acid standard solution. Moreover, standardize its concentration by the following procedure:

Transfer accurately 25 mL of the 0.05 mol/L sulfuric acid standard solution into a 200-mL Erlenmeyer flask, add a few drops of methyl red test solution, titrate with the 0.1 mol/L sodium hydroxide standard solution, calculate the factor (f_2) of the 0.05 mol/L sulfuric acid standard solution by thefollowing formula:

$F2= (v*f1)/(25)$

f_1 : Factor of 0.1 mol/L sodium hydroxide standard solution

V : Volume (mL) of 0.1 mol/L sodium hydroxide standard solution required for titration

B. Sample solution preparation

Weigh accurately 1-5 g of an analysis sample, put it in a Kjeldahl flask, add 9 g of potassium sulfate and 1 g of copper sulfate (II) pentahydrate, further add 30-40 mL of sulfuric acid, and mix by shaking. Heat it gradually, and then strongly after foaming subsides, and heat for not less than 2 hours after the solution becomes clear, and then let it stand to cool. Transfer the solution with water into a 250-mL volumetric flask, and add water up to the marked line to be the sample solution.

C. Quantification

1) Absorption by the sulfuric acid standard solution

Transfer accurately a certain amount of the sample solution into a Kjeldahl flask, and add sodium hydroxide solution (50 w/v%) of a volume sufficient to turn the solution strongly alkaline. Connect the flask to the steam distillation apparatus to which a receiver containing a certain amount of 0.05 mol/L sulfuric acid standard solution in advance is attached, and distill until the distillate volume reaches about 120 mL.

Add a few drops of methyl red test solution to the distillate, titrate with the 0.1 mol/L sodium hydroxide standard solution, and calculate the nitrogen [N] content by the following formula. Multiply it by 6.25 (6.38 for samples of milk products or milk replacer for calves which contain milk products not less than 50 %) to calculate the crude protein content in the sample.

Nitrogen content (%)= $(1.4*10^{-3})*f1*(v1-v2)*(250/v)*(100/w)$

f1 : Factor of 0.1 mol/L sodium hydroxide standard solution

V1: Volume (mL) of 0.1 mol/L sodium hydroxide standard solution equivalent to the volume of 0.05 mol/L sulfuric acid standard solution in the receiver

V2: Volume (mL) of 0.1 mol/L sodium hydroxide standard solution required for titration

V : Volume (mL) of the sample solution used for distillation

W : Weight (g) of the sample used for analysis

2) *Absorption by boric acid solution*

Put a certain amount of boric acid solution (4 w/v%) into a receiver instead of the 0.05 mol/L sulfuric acid standard solution, and distill in the same way as 1).

Add a few drops of bromocresol green-methyl red test solution to the distillate, titrate with the 0.05 mol/L sulfuric acid standard solution, and calculate the nitrogen [N] content by the following formula. Multiply it by 6.25 (6.38 for samples of milk products or milk replacer for calves which contain milk products not less than 50 %) to calculate the crude protein content in the sample.

Nitrogen content (%)= $(1.4*10^{-3})*f2*(v1)*(250/v)*(100/w)$

f2 : Factor of 0.05 mol/L sulfuric acid standard solution

V1: Volume (mL) of 0.05 mol/L sulfuric acid standard solution required for titration

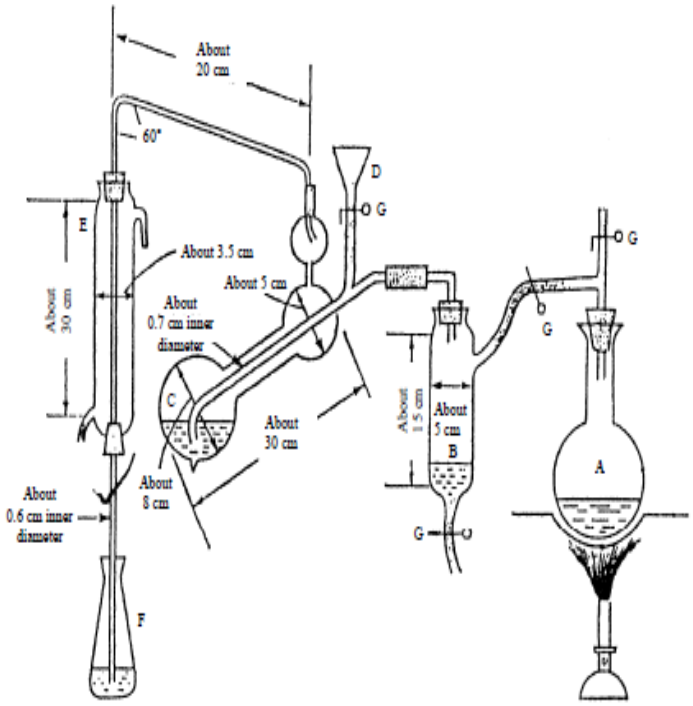
V : Volume (mL) of the sample solution used for distillation

W : Weight (g) of the sample used for analysis

«Summary of analysis method»

The method quantitates total nitrogen in a sample by the Kjeldahl method. However, nitrate nitrogen or nitrite nitrogen is not quantitated.

Generally, a macro-Kjeldahl nitrogen distillation apparatus by the indirect distillation method as shown in the following Figure is used.



A : Round-bottom flask for steam generation

B : Trap for fluid waste collection

C : Distillation flask

D : Funnel for the addition of the sample solution and sodium hydroxide

E : Condenser (if condensation occurs on the outer surface, it is recommended to wrap the condenser with cloth to prevent dripping off.)

F : Receiver (200-mL Erlenmeyer flask)

G : Pinch cock

Precautions and Notes

[1] Connect the container containing the sodium hydroxide standard solution prepared with a soda lime tube or a bottle containing sodium hydroxide solution to avoid the entrance of carbon dioxide in the air; however, it is desirable to standardize it once every 2-3 months.

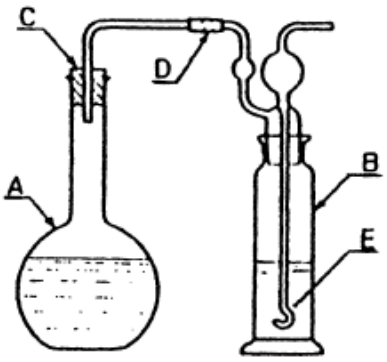
Commercially available 0.1 mol/L sodium hydroxide solution may as well be used.

[2] Because sodium hydroxide is highly hygroscopic and is likely affected by carbonic acid, theoretically it is difficult to obtain a solution of accurate concentration. Use saturated solution to avoid the effect of carbonic acid (concentrated sodium hydroxide solution contains little carbon dioxide).

About 80 g of sodium hydroxide is soluble in 74 mL of water at 20 °C, thus it is recommended to prepare saturated solution by adding slightly excessive sodium hydroxide and leave at rest to collect clear supernatant to be used (the concentration is about 20 mol/L at 20 °C).

[3] Use purified water that is boiled and then cooled to remove carbon dioxide. See JIS K 8001 “General rule for test methods of reagents.”

Boil water in a flask for 15 minutes, and then shut out carbon dioxide in the air by attaching a gas washing bottle as shown in the following Figure containing potassium hydroxide solution (25 w/v%) or a soda lime tube, and cool.



A : 1-L flat bottom flask
B : Gas washing bottle
C : Ground-joint stopper or silicon rubber stopper
D : Ground joint or silicon rubber tube
E : Potassium hydroxide solution (25 w/v%)

[4] Vacuum by suction with a vacuum pump, etc. (not more than 2.0 kPa). [5] See the appendix table for the preparation method.

[6] The endpoint is where the yellow color disappears and becomes greenish blue and the tone is maintained for not less than 30 seconds.

$$HO{\cdot}SO2{\cdot}NH2 + NaOH \rightarrow NaO{\cdot}SO2{\cdot}NH2 + H2O$$

During titration, use the fixed range (such as the graduation range of 10-20 mL) of a burette. [7] The number 97.10 means the molecular weight of amidosulfuric acid.

[8] Commercially available 0.05 mol/L sulfuric acid may as well be used.

[9] Connect the container containing the sulfuric acid standard solution prepared with a bottle containing dilute sulfuric acid to avoid the entrance of ammonia gas.

[10] Dissolve 0.1 g of methyl red in ethanol to be 100 mL. Filter the solution if needed.

Methyl red-methylene blue mixture test solution (dissolve 0.2 g of methyl red and 0.1 g of methylene blue respectively in ethanol (90 v/v%) to be 100 mL, and mix them.) may as well be used. When the mixture test solution is used for titration, the endpoint is where the red-purple color turns to blue and then to green.

[11] The number 25 in the calculation formula means the volume of the 0.05 mol/L sulfuric acid standard solution (25 mL) contained in the Erlenmeyer flask.

[12] Potassium sulfate and copper sulfate are used as degradation accelerators.

Degradation accelerators for Kjeldahl degradation includes (a) copper sulfate - potassium sulfate, (b) copper sulfate - selenium - potassium sulfate, (c) titanium dioxide - copper sulfate - potassium sulfate, and (d) mercuric - potassium sulfate, etc. As a measure to prevent environmental pollution and in order to avoid troubles caused such as by the influence of degradation accelerators when the same sample solution is used for colorimetric determination of phosphorus, only copper sulfate - potassium sulfate were employed as degradation accelerators.

Copper sulfate is a catalyst to facilitate degradation, while potassium sulfate elevates the concentration of sulfuric acid and the boiling point as well as facilitates degradation via the following reactions:

$K_2SO_4 + H_2SO_4 \rightarrow 2KHSO_4$

$2KHSO_4 \rightarrow K_2SO_4 + H_2O + SO_3$

$2KHSO_4 \rightarrow K_2S_2O_7 + H_2O$

$K_2S_2O_7 \rightarrow K_2SO_4 + SO_3$

[13] Degradation time may be further reduced by letting it stand to cool after adding sulfuric acid and heating while gradually adding about 1 mL of

hydrogen peroxide solution (not less than 30 v/v%). Make sure to conduct degradation in a draft chamber.

[14] Care should be taken for samples with high oil content such as plant oil cake because it may foam violently and spill out of the Kjeldahl flask when heating is strong. When it foams strongly, stop heating and leave at rest for a while, and then heat again with lower heat. Additionally, it is recommended to add a small amount of paraffin to a sample that foams strongly. [15] Because it is not appropriate in some cases to consider that degradation is completed when the solution becomes clear, it is needed to heat further.

Care should be taken so that the solution is not less than 10 mL because it is said that there is the loss of ammonia gas when the heating temperature is too high or the solution volume after degradation is too small.

Crude protein is degraded by sulfuric acid and turns into the form of (NH4)2SO4.

[16] The two methods, absorption by the sulfuric acid standard solution and absorption by boric acid solution, are listed in the Analytical Standard of Feed as the quantification methods for crude protein. [17] This method is frequently used in fertilizer analysis, food analysis and plant analysis, etc.

[18] It is judged by the emergence of the blue color of copper oxide.

[19] When 10 mL of the 0.05 mol/L sulfuric acid standard solution is used, distillation can be conducted according to the rough standards shown below based on the crude protein content in the sample:

Not more than

15 % 2 g-250 mL-50 mL

35 % 2 g-250 mL-25 mL

45 % 2 g-250 mL-20 mL

60 % 2 g-250 mL-10 mL

[20] At first ammonia is generated in a large amount; care should be taken to avoid the loss of ammonia by controlling the amount of steam introduced.

[21] One milliliter (1 mL) of the 0.1 mol/L sodium hydroxide standard solution corresponds to 1.40 mg of nitrogen.

[22] The value corresponds to the case when the degradation solution of the analysis sample is accurately 250 mL.

[23] This method is widely used in the Japanese Pharmacopoeia, JIS, and food analysis, etc. The method titrates ammonia with a strong acid when the ammonia (NH3) generated from an alkalinized sample solution is passed through dilute boric acid solution and completely is dissociated as shown in the following formula:

$$NH_3 + H_3BO_3 + H_2O \rightarrow H_3BO_3(OH)^- + NH4^+$$

The method is advantageous in that it is not needed to strictly define the concentration of the boric acid solution or the amount to take the boric acid solution, because the amount of boric acid is not directly involved in titration.

In addition, it is convenient to add methyl red - bromocresol green test solution in the boric acid solution in advance.

Prepare by dissolving 400 g of boric acid, 100 mL of 0.1 % bromocresol green solution in ethanol, and 70 mL of 0.1 % methyl red solution in ethanol in 10 L of water.

Ammonia is absorbed by boric acid, which is a weak acid. Ammonia can be sufficiently captured at the boric acid concentration of not less than 3 %; however, the concentration is stipulated as 4 % including the margin of safety.

Additionally, care should be taken for the boric acid solution in the receiver not to exceed 40 °C. Ammonia absorption decreases at high temperature, leading to loss.

[24] Dissolve 0.15 g of bromocresol green and 0.1 g of methyl red in 180 mL of ethanol. Add water to be 200 mL.

[25] The endpoint is when the green color disappears and changes into slightly grayish blue and then to slightly grayish red purple.

[26] One milliliter (1 mL) of the 0.05 mol/L sulfuric acid standard solution corresponds to 1.40 mg of nitrogen.

[27] The value corresponds to the case when the degradation solution of the analysis sample is accurately 250 mL.

2- Combustion method

Quantification

Weigh 100-500 mg[2] of an analysis sample*2[1][3], put it in a nitrogen (protein) analyzer,*3[4] and run the analyzer to obtain the response peak of nitrogen gas with a detector.

Similarly, weigh the reagent for calibration curve preparation*4 accurately, and put it in the analyzer to obtain the response peak of nitrogen gas with a detector. Calculate the area from the response peak obtained to prepare the calibration curve, calculate the nitrogen [N] amount in the sample, and multiply the nitrogen [N] amount by 6.25 (6.38 for samples of milk products or milk replacer for calves which contain milk products not less than 50 %) to be the crude protein content in the sample.

Requirements for the analysis instrument

i) Capable of thermolysis of a sample in oxygen gas (purity not less than 99.9 %), maintaining the temperature in the reactor at 870 °C at the minimum

ii) Capable of separation of free nitrogen gas from the other combustion products

iii) Equipped with the system to convert nitrogen oxide (NOx) into nitrogen gas (N2), or capable of measuring nitrogen as NO2

iv) Capable of measuring nitrogen gas with a thermal conductivity detector

1. For samples with high nitrate nitrogen content such as Sudan grass, it is quantitated as higher crude protein, and thus measure nitrate nitrogen [N] content separately and subtract the value.

2. All the amount of the analysis sample is screened through a net sieve of 0.5-mm mesh.

3. Use the instrument according to the combustion method, and measure under the conditions appropriate for the instrument.

4. Use reagents specified for the nitrogen (protein) analyzer used, such as disodium dihydrogen ethylenediaminetetraacetate dihydrate, DL-aspartic acid, etc.

Summary of analysis method

The method is a quantification method using an automatic analyzer applying the Dumas method, in which a sample is degraded by combustion at high temperature and released nitrogen gas is quantitated by a thermal conductivity detector (TCD) for crude protein in feeds.

The method is advantageous in that the analysis time is reduced and that facilities such as a draft are not needed.

The schematic diagram of the analyzer is shown in Figure 3.2-3.

«Notes and precautions»

[1] Particle size is set as 0.5 mm because the measured value may vary in a sample that passed a net sieve of 1-mm mesh; however, 1 mm will do in some samples.

[2] The sampling amount should be adjusted according to the nitrogen content in the sample and to the specifications of the analyzer used.

[3] Sampling boats to load a sample are made of quartz, ceramic, etc.

Other methods

3. Crude protein quantification by international standards

ISO 5983 (1997) Animal feeding stuffs - Determination of nitrogen content and calculation of crude protein content - Kjeldahl method

Flow sheet of the analysis method

Sample	• 0.5-2.0 g (800-mL Kjeldahl flask) • 15 g of potassium sulfate • 0.3 g of copper (II) oxide or 0.9-1.2 g of copper (II) sulfate tetrahydrate • Add 25-40 mL of sulfuric acid and, depending on the sample, a defoaming agent (paraffin)
Heating	• After the sample solution becomes clear, heat for 2 hours.
Cooling	
	• Add 250-350 mL of water and dissolve. • Put 25 mL of 0.1 mol/L or 0.05 mol/L sulfuric acid in a trapping flask, add 100-150 mL of water, and add 2-3 drops of mixed indicators (MR and MB).
Distillation	
	• Put 100 mL of 33 % sodium hydroxide solution in a flask, and connect it to the distiller. • Titrate with 0.1 mol/L or 0.05 mol/L sodium hydroxide standard solution. (When boric acid is used as the trapping solution, titrate with 0.05 mol/L or 0.125 mol/L sulfuric acid standard solution.)
Titration	

4. Analysis with automated instruments

Recently, automated instruments have been developed to accelerate quantification procedures applying the Kjeldahl method by degradation with sulfuric acid.

(1) SuperKjel automatic nitrogen/protein analyzer

(2) Kjeltec auto system

(3) Kjeldahl method nitrogen/protein analyzer VAP series

In the field of crude feed, near-infrared spectrophotometers have been becoming popular.

Methods of nitrogen analysis			
Procedure	***Applicability***	***Limitations***	***Capital costs***
Total nitrogen			
Kjeldahl	Manual, all foods	Minor interference from inorganic nitrogen	Low
	Automated, at several levels of complexity	Minor interference from inorganic nitrogen	Medium
Dumas	Automated, all foods	Includes inorganic nitrogen. Analytical portion size	High
Radiochemical methods	Most foods	Instrumentation required	Very high
Protein			
Total N x facto	All foods	Variations in NPN	Low
Protein N x factor	Preferable for vegetables, some fish, yeast foods, insect foods, breastmilk	Choice of procedure for measurement of NPN. Better to use amino acid N	Low
Methods applicable to specific foods			
Formol titration	Dairy products	Specificity	Low
Biuret	As above	Specificity	Low
Folin s reagent	As above	Specificity	Low
Alkaline distillation	Cereals	Specificity	Low
Dye-binding	Specific foods, some cereals, some legumes	Specificity	Low
NIR	Established for some foods	Number of calibration samples	High

Factors for the conversion of nitrogen to protein	
Foodstuff	***Factor***
Animal products	
Meat and fish	6.25
Gelatin	5.55
Milk and milk products	6.38
Casein	6.40
Human milk	6.37
Eggs	
whole	6.25
albumin	6.32
vitellin	6.12
Plant products	
Wheat	
whole	5.83
bran	6.31
embryo	5.80
endosperm	5.70
Rice and rice flour	5.95
Rye and rye flour	5.83
Barley and barley flour	5.83
Oats	5.83
Millet	6.31
Maize	6.25
Beans	6.25
Soya	5.71
Nuts	
almond	5.18
Brazil	5.46
groundnut	5.46
others	5.30
a specific factor is not listed, 6.25 should be used until a more appropriate factor has been determined.	

3. Ether Extract (EE) or crude fat

Summary of ether extract

A sample is extracted with ether using the Soxhlet extractor, to obtain the extract as ether extract or crude fat.

Ether extract contains, in addition to fat, oil-soluble dyes (such as chlorophyll and carotenoids), wax, free fatty acids, lecithin, cholesterin, and phospholipids, etc.

Generally, ether extract of oil meal is around 1 % and the major component is oil and fat, therefore the energy value is high. Ether extract in cereals and bran is 4-5 % and the major component is oil and fat but contains a large amount of contaminants, therefore the energy value is less than that of oil meal; however, it is characterized in that it contains a lot of oil-soluble vitamins such as carotene and tocopherol. Also, when feeds are stored for a long period, a phenomenon is observed in that moisture does not change while ether extract decreases gradually. This is because unsaturated fatty acids contained in feeds are oxidatively polymerized absorbing oxygen in the air and becomes insoluble in ether.

Determination Methods

1. Diethyl ether extraction

Quantification

Weigh accurately 2-5 g of an analysis sample, put it in a filter paper thimble; (22 mm in diameter, 90 mm in height), put absorbent cotton on it pushing lightly, and dry at 95-100 °C for 2 hours.

Transfer the thimble into a Soxhlet extractor, connect to a fat weighing bottle (dried at 95-100 °C, cooled in a desiccator, and accurately weighed in advance), add diethyl ether, and extract for 16 hours.

Then, remove the filter paper thimble, and collect diethyl ether. Remove the fat weighing bottle to evaporate diethyl ether, dry at 95-100 °C for 3 hours, let it stand to cool in a desiccator, and weigh accurately to calculate the ether extract content in the sample.

N.B: When it is difficult to grind the sample due to high fat content, prepare the analysis sample to obtain the ether extract content in the original sample as follows.

To the volumetric flask containing diethyl ether used in the preliminary extraction, add diethyl ether to the marked line, transfer accurately a certain amount of it to a fat weighing bottle (dried at 95-100 °C, cooled in a desiccator, and accurately weighed in advance), and obtain the ether extract content in the original sample after preliminary extraction according to the quantification method shown above.

Then, obtain the ether extract content in the sample after preliminary extraction by the quantification method shown above, and calculate the ether extract content in the original sample by the following formula:

Ether extract content (%) in the original sample= A+((100-B)*C)/100

A : Ether extract content (%) obtained from the original sample after preliminary extraction

B : Loss (%) by preliminary extraction

C : Ether extract content (%) in the sample after preliminary extraction

Summary of analysis method

Put a sample in a filter paper thimble, load it on a Soxhlet extractor, add diethyl ether, heat and circulate to collect diethyl ether-soluble components in a

fat weighing bottle below, evaporate diethyl ether, dry, and weigh the weight of the extract.

Notes and precautions

[1] Select according to the size of the extractor tube. Use the thimble of 22×90 mm when the apparatus shown in [5] below is used.

[2] Place absorbent cotton on the bottom of the filter paper thimble and put an analysis sample on it, so that the removal of the analysis sample after extraction will be easier. It is convenient to use a glass funnel of about 10 mm in the diameter of the stem to transfer the analysis sample into the filter paper cylinder.

[3] Absorbent cotton is stuffed on the analysis sample in order to infiltrate diethyl ether into the whole sample and to prevent release of the sample from the top of the filter paper thimble.

[4] Drying for a long time may cause evaporation of lower free fatty acids or oxidation of unsaturated fatty acids in some samples, while insufficient drying may cause extraction of water-soluble substances such as sugars into the diethyl ether extract, resulting in a higher measured value.

[5] The Soxhlet extractor (Figure 3.3-1) circulates diethyl ether by the siphon principle.

Adjust the water bath or heater so that 2-3 drops per second of diethyl ether dribbles from the condenser (at the level of 16-20 circulations per hour).

[6] Dust or dirt on the hand may be adhered on the outer surface of the weighing bottle; wipe the bottle with gauze, etc., before weighing.

[7] Remaining diethyl ether causes danger in a dryer; therefore evaporate diethyl ether by putting the fat weighing bottle in a water bath.

[8] The cooling time in a desiccator should be the same as in the pre-drying.

[9] Recently, automated Soxhlet apparatus is also used in which extraction time is shortened and the solvent amount is reduced.

2. Acid degradation/diethyl ether extraction

Scope of application: Expanded feeds, formula feeds using fat powder material (only formula feeds for milk replacer for calves in the milk-feeding period and formula feeds for piglets in the in the milk-feeding period), formula feeds for dairy cows using fatty acid calcium salt material, soy oil foots and rapeseed oil foots

Quantification

Weigh accurately 2 g of an analysis sample, put it in a 100-mL beaker, add 2 mL of ethanol, mix with a glass bar to moisten the sample, then add 20 mL of hydrochloric acid (4+1), cover with a watch glass, heat in a water bath at 70-80 °C with occasional stirring for 1 hour, and then let it stand to cool.

Transfer the content of the said beaker into a 200-mL separatory funnel A, wash the beaker sequentially with 10 mL of ethanol and 25 mL of diethyl ether, and add the washing to the separatory funnel A. Further add 75 mL of diethyl ether to the separatory funnel A, mix by shaking and leave at rest. Collect the diethyl ether layer (upper layer) with a pipette, etc., and transfer it to a 300-mL separatory funnel B containing 20 mL of water in advance.

Add 50 mL of diethyl ether to the separatory funnel A, repeat the same procedure twice, and collect each diethyl ether layer with a pipette, etc., and add it to the separatory funnel B.

Shake the separatory funnel B to mix, and leave at rest, and discard the water layer (lower layer). Further add 20 mL of water to the separatory funnel B, and repeat the same procedure twice. Filter the diethyl ether layer[5] with a funnel that is stuffed with absorbent cotton and a proper amount, not less than 10

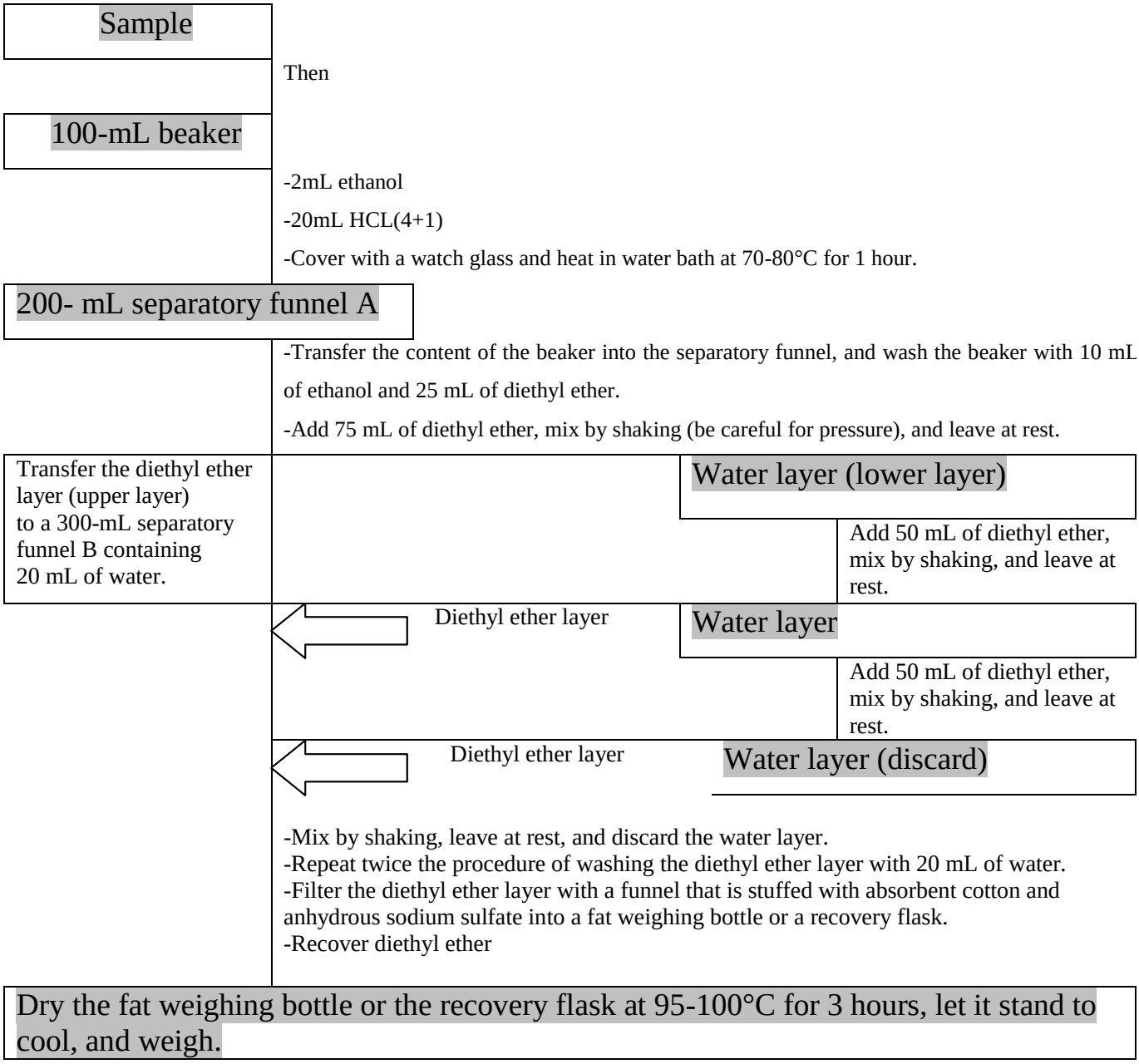
g, of sodium sulfate (anhydrous) in advance into a fat weighing bottle or a 300-mL recovery flask (dried at 95-100 °C, cooled in a desiccator, and accurately weighed in advance).

Then, recover diethyl ether in the said fat weighing bottle using a Soxhlet extractor, or in the said recovery flask using a rotary evaporator. Remove the fat weighing bottle or the recovery flask to evaporate diethyl ether, dry at 95-100 °C for 3 hours, let it stand to cool in a desiccator, and weigh accurately to calculate the ether extract content in the sample.

Summary of analysis method

In this method, a sample from which fat is not directly extracted completely with diethyl ether is hydrolyzed using hydrochloric acid solution to disperse free fat in the sample, and then is subjected to liquid-liquid extraction with diethyl ether. A process to wash the diethyl ether layer with water is added in order to prevent higher measured value caused by water-soluble substances contained in the diethyl ether layer other than ether extract.

The scope of application of the method is specified to avoid confusion in analytical testing. The flow sheet of the analysis method is shown in the following Figure.



«Notes and precautions»

[1] Ethanol is added to prevent the solidification of the sample when hydrochloric acid is added.

[2] The solution turns from brown to blackish brown along with degradation.

[3] Pressure in the separatory funnel increases immediately; release pressure sufficiently before shaking.

[4] Care should be taken because a plus error is caused by the transfer of the blackish brown water layer to the separatory funnel B.

[5] White solid content may be left on the funnel in samples with a high content of hydrogenated fat; filter it in with heating to avoid the precipitation of the white solid content.

[6] An error may be caused by ethanol remaining in the fat weighing bottle or the recovery flask; evaporate ethanol by heating with a water bath or purging with nitrogen gas.

[7] A Mojonnier flask has the shape shown in Figure 3.3-3, and is made so that it can be centrifuged and the upper layer can be collected only by tilting.

Other methods

3. Ether extract quantification by international standards

ISO 6492 (1999) Animal feeding stuffs - Determination of fat content Flow sheet of the analysis method

Sample	5.0 g (put in a filter paper cylinder and cover with absorbent cotton)
Soxhlet extractor	Extract for 6 hours by light crude oil.
Fat weighing bottle	Add 2 mL of acetone, and evaporate acetone on a heating device.
Drying	Dry the fat weighing bottle at 103±2 °C for 10±0.1 minutes.
Cooling	Desiccator
Weighing	

4. Analysis with automated instruments

The following instruments are commercially available, which are considered to be equivalent to the Soxhlet extractor in 1:

(1) Soxtest SER (Actac)

(2) Soxtec (FOSS Tecator, distributed by Foss Japan)

(3) Soxtherm (Gerhardt, distributed by Gerhardt Japan)

These automated instruments are characterized in that analysis time can be completed in about 1/5 of the

time for the method by the conventional Soxhlet extractor, that less amount of solvent is used, and that solvent can also be collected automatically.

Moreover, Hydrotherm (Gerhardt, distributed by Gerhardt Japan) was developed as an instrument to automate acid degradation in 2 before an ordinary Soxhlet extractor or an automated extractor.

However, data should be compared with the data from the conventional Soxhlet extractor before using a newly released instrument without history of use

Table 7.5 Methods of analysis for lipids			
Procedure	***Application***	***Limitations***	***Capital cost***
Total fat			
Continuous extraction	Low moisture foods (single solvent)	Incomplete extraction from many foods. (dry analytical samples) Time consuming. Extracts cannot be used for fatty acid studies	Low
Acid hydrolysis	All foods except dairy and high sugar products	Some hydrolysis of lipids. Extracts cannot be used for fatty acid studies	Low
Acid hydrolysis and capillary GLC	Cereal foods (NLEA compliant)		High
Mixed solvent extraction	Rapid, efficient for many foods. Extract can be used for fatty acid measurements	Complete extraction from most foods. Extractsoften need clean-up	Low
Alkaline hydrolysis	Dairy foods	Validated for dairy foods only	Low
NIR	Established for cereals	Requires extensive calibration against other methods	High
Triacylglycerols			
Range of chromatographic methods	All foods	Free fatty acids can interfere. TLC checks usefu	Medium
Fatty acids			
G L C	All foods after transmethylation	Validated for most foods	High
HPLC	Under development	Not found to have advantages over GLC at present	High
***Trans*fatty acids**			
GLC with infrared analyses	All foods	Availability of authentic standards for some isomers	Medium to High
Infrared absorption	All foods	Some interference	High
G L C	All foods	Capillary techniques are required	High/medium
Notes: GLC = gas-liquid chromatography; NLEA = United States Nutrition Labeling and Education Act NIR = near infrared reflectance; TLC = thin-layer chromatography; HPLC =			

high-performance liquid chromarography.

4. Crude Fiber (CF)

Summary

A sample is boiled sequentially with dilute acid and then with dilute alkali, and then sequentially washed with ethanol and diethyl ether, and the residue is subtracted by its ash, and the result is defined as crude fiber. Crude fiber is primarily measured to comprehend indigestible parts in feeds, and is consisted mainly of a part of lignin, pentosan, chitin, etc., in addition to cellulose.

These compounds are collectively called as fiber; however, the sum of their individually measured contents is significantly different from the crude fiber content obtained by the method shown above, and the former is always larger. This is because a part of lignin and hemicellulose is dissolved during the boiling procedure, and the percentage of dissolution varies depending on the feed type and thus is not constant.

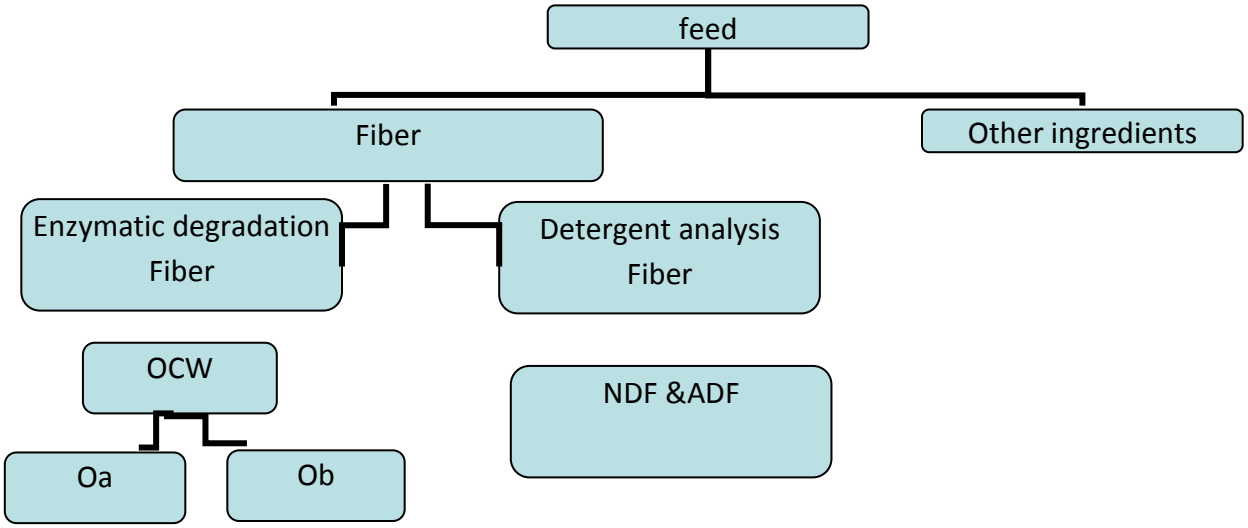
Crude fiber clearly corresponds only to feeds of plant origin considering the component compounds; however, a small amount of it is contained in feeds of animal origin. This is because organic residue that is not dissolved by acid/alkali boiling is observed in feeds of animal origin, and the residue is chitin and some of scleroprotein (albuminoid), which are completely different from so-called crude fiber in content. There is a close relationship between the crude fiber content and the nutrition value of the feed, and generally the higher the crude fiber content is, the lower the nutrition value is.

On the other hand, fiber is an important nutrient for ruminant livestock. Fiber that was previously in the scope of feed analysis was only crude fiber of general nutrients, which was insufficient for the evaluation of fiber. This is because plant fiber is mainly consisted of cell wall which is comprised of substances such as cellulose, hemicellulose, lignin and pectin, but crude fiber quantitated in general analysis does not include hemicellulose, etc. To measure

fiber as accurate as possible, detergent analysis using detergents was developed in Europe and the US, while enzymatic analysis using enzymes was developed in Japan led by the National Institute of Animal Industry.

In detergent analysis, a feed is heated using a neutral detergent and is separated into soluble and insoluble parts. Organic matter in the insoluble part corresponds to cell wall, and is defined as neutral detergent fiber (NDF). The "low digestivity" fraction in cell wall is measured by heat treatment with an acidic detergent, and is defined as acid detergent fiber (ADF).

In enzymatic analysis, starch and protein are degraded by enzymes and is separated into soluble and insoluble parts. Organic matter in the insoluble part corresponds to cell wall, and is defined as organic cell wall (OCW). The "indigestible" fraction after the degradation of starch and protein is measured by degrading cellulose with enzymes, and is defined as fiber with low digestivity or organic b fraction (Ob).



NDF: mainly consisted of cellulose, hemicellulose and lignin (total fiber)

ADF: mainly consisted of a part of cellulose and lignin (fiber with low digestibility)

OCW: mainly consisted of cellulose, hemicellulose and lignin (total fiber)

Oa: =OCW-Ob (fiber with high digestibility)

Ob: mainly consisted of a part of cellulose and lignin in OCW (fiber with low digestibility)

Determination Methods

1. Standing method

Quantification

Weigh accurately 2-5 g of an analysis sample, put it in a 500-mL tall beaker, add 50 mL of sulfuric acid (1+34), and further add water to be 200mL. Then, cover the tall beaker with a watch glass or a condenser, boil for 30 minutes while supplying moisture that is evaporated, add 300 mL of water and leave at rest overnight, aspirate the supernatant, add water again to be 200 mL, and subject to the same procedure.

To the residue (acid-insoluble fraction), add 50 mL of sodium hydroxide solution (5 w/v%), add water to be 200 mL, cover with a watch glass or a condenser, and subject to the same procedure as acid treatment. Filter the residue (acid/alkali-insoluble fraction) with filter paper (No.5A) (placed in an aluminum weighing dish, dried at 135±2 °C for 2 hours, cooled in a desiccator, and accurately weighed in advance). Wash the residue on the filter paper with hot water until the alkaline reaction of filtrate disappears, further wash 2-3 times each sequentially with a small amount of ethanol and diethyl ether, and then air-dry for 3-4 hours. Then put the acid/alkali-insoluble fraction with the filter paper in the said weighing dish, dry at 135±2 °C for 2 hours, let it stand to cool in a desiccator, and then weigh accurately, and calculate the amount of the acid/alkali-insoluble fraction in the sample. Transfer the residue in the weighing dish to a crucible (heated at 550-600 °C for 2 hours, cooled in a desiccator, and accurately weighed in advance). Heat this gently to be charred, then heat at 550-600 °C for 2 hours to incinerate, let it stand to cool in a desiccator, and accurately weigh to obtain the ash content in the sample.

Subtract the ash content from the amount of the acid/alkali-insoluble fraction to calculate the crude fiber content in the sample.

Summary of analysis method

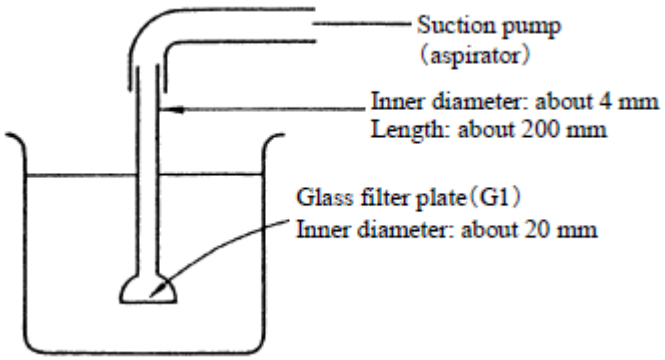
Treat a sample sequentially with sulfuric acid (1+34), sodium hydroxide solution (5 w/v%), ethanol and ether, and weigh the residue (designated as A).

Then, incinerate the residue and weigh (designated as B), and calculate A-B s crude fiber. Crude fiber contains most of cellulose and a part of hemicellulose, lignin and pectin.

«Notes and precautions»

[1] If the content adhered to the inner wall of the tall beaker, drop it off so that the content is continuously treated with sulfuric acid. Make sure to keep the acid concentration constant by adding hot water when moisture is evaporated.

[2] When an aspiration tube with a filter plate (G1) is used, as shown in this figure , wash off adhered matter on the glass filter plate after the aspiration procedure with water into the tall beaker.



[3] Conduct the procedure of boiling, leaving at rest, and then aspirating the supernatant.

[4] Bumping often occurs when boiling with sodium hydroxide solution; this can be prevented by adding boiling stones weighed in advance to the solution.

In addition, make sure to keep the alkali concentration constant by adding hot water when moisture is evaporated.

[5] See 1. Moisture, Precautions and Notes.

Write the number of aluminum weighing dish on filter paper with a pencil, etc.

[6] For fish meal and feather meal, etc., filter immediately because filtration becomes difficult at lower temperature when the solution is left after treatment.

[7] Check that there is no alkaline reaction with litmus paper or pH test paper.

[8] The major objectives are the reduction of air-dry time and defatting.

[9] Obtain according to the quantification method for crude ash.

2. Filtration method [Analytical Standard of Feed, Chapter 3, 4-2)] Quantification

Weigh accurately 2-5 g of an analysis sample, put it in a 500-mL tall beaker, add 50 mL of sulfuric acid(1+34), further add water to be 200 mL, cover with a watch glass or a condenser, boil for 30 minutes while supplying moisture that is evaporated, and then filter the residue with 0.045-mm stainless wire mesh[1], and wash with hot water.

Transfer the residue (acid-insoluble fraction) to the said tall beaker using 130-140 mL of water, add 50 mL of sodium hydroxide solution (5 w/v%) 50 mL, and further add water to be 200 mL.

Then, cover the tall beaker with a watch glass or a condenser, and boil for 30 minutes while supplying moisture that is evaporated.

Filter the residue (acid/alkali-insoluble fraction) with filter paper (No.5A) (placed in an aluminum weighing dish, dried at 135±2 °C for 2 hours, cooled in a desiccator, and accurately weighed in advance). Wash the residue on the filter paper with hot water until the alkaline reaction of filtrate disappears, further wash 2-3times each sequentially with a small amount of ethanol and diethyl ether, and then air-dry for 3-4 hours.

Then put the acid/alkali-insoluble fraction with the filter paper in the said weighing dish, dry at 135±2 °C for 2 hours, let it stand to cool in a desiccator, and then weigh accurately, and calculate the amount of the acid/alkali-insoluble fraction in the sample.

Transfer the residue in the weighing dish to a crucible (heated at 550-600 °C for 2 hours, cooled in a desiccator, and accurately weighed in advance). Heat this gently to be charred, then heat at 550-600 °C for 2 hours to incinerate, let it stand to cool in a desiccator, and accurately weigh to obtain the ash content in the sample.

Subtract the ash content from the amount of the acid/alkali-insoluble fraction to calculate the crude fiber content in the sample.

Summary of analysis method

The principle of this analysis method is the same as the standing method in 1. in this section. In the standing method, there is a step of standing overnight after boiling with sulfuric acid, while filtration with stainless wire mesh or nylon paper is conducted in this method to speed up the analysis.

Precautions and Notes

[1] Formerly nylon filter paper, but the product is discontinued; so, the method is modified to use stainless wire mesh of 0.045 mm (325 mesh) which has a filtering effect similar to the nylon paper employed in the AOACI method.

Another product equivalent to the previously used nylon paper has been developed.

[Other analysis methods]

3. Crude fiber quantification by international standards

ISO 6865 (2000) Animal feeding stuffs - Determination of crude fiber content Flow sheet of the analysis method

Sample	1.0 g, 500 mL beaker
	150 mL of 0.13 mol/L sulfuric acid
Boiling	30 minutes; 2-3 drops of a defoaming agent when it foams
Filtration	Add filter aid to a filter crucible (a Gooch crucible with glass fiber filter paper) and filter by suction.
Washing	Wash with 10 mL of hot water×5, add acetone, leave at rest for 2-3 minutes, and transfer the filtered residue to a beaker.
	150 mL of 1.29 % potassium hydroxide
Boiling	30 minutes
Filtration	Add filter aid to a filter crucible and filter by suction.
	Wash 3 times with 30 mL of acetone.
Drying	130 °C, 2 hours
Cooling	Desiccator
Weighing	
Charring	Put the filter crucible in a muffle furnace, and char at 500 °C.
Cooling	
Weighing	

4. Analysis with automated instruments

Recently, automated instruments for crude fiber have been becoming fairly popular. Types commercially available in Japan are shown below:

(1) Fibertest FIWE (Actac)

(2) Fibertec, FiberCap (FOSS Tecator, distributed by Foss Japan)

(3) Fiber analyzer SA-120 (ANKOM, distributed by Sanshin Industrial)

(4) Fibretherm FT12 (Gerhardt, distributed by Gerhardt Japan)

(1) and (2) automate the manual procedure of the analysis method in 2. in this section, and can treat 3 or 6 samples simultaneously. High-speed filtration and washing can be conducted by a built-in suction pump, significantly reducing analysis time.

(3) and (4) automate reagent injection and washing, with samples loaded in filter bags.

All the instruments can also analyze ADF, NDF, cellulose, hemicellulose and lignin, etc. Comparison with manual analysis should be conducted before using these instruments.

5. Heat-stable α-amylase-treated neutral detergent fiber (aNDF and aNDFom)

[Summary of heat-stable α-amylase-treated neutral detergent fiber]

Neutral detergent fiber (NDF) was developed as an analysis method for total fiber in grass. Various modification have been done later to be applicable to feed crop other than grass, and several modified methods have been introduced. Therefore, there are many kinds of analysis methods, and results by different treatment methods have all been reported as the NDF value. For that reason, NDF may differ for the same sample between different laboratories, thus the handling of the analysis value is a problem. With that, heat-stable α-amylase-treated neutral detergent fiber (hereinafter referred to as aNDF) was developed as an analysis method that is commonly applicable to all feed crop. Today the Nutrient Requirements of Dairy Cattle (NRC) used in the United States employs aNDF instead of previously used NDF, while AOAC International and ISO standardize

the aNDF analysis method. These aNDF methods use heat-stable α-amylase and sodium sulfite commonly for all feeds.

Substances soluble in neutral detergent are digested by bacteria in the digestive organs of livestock. Therefore, NDF is the part that is not digestible or is slowly digested by livestock, and corresponds to nonsoluble fiber. Its major components are cellulose, hemicellulose and lignin.

[Methods listed in the Analytical Standard of Feed]

1. Heat-stable α-amylase-treated neutral detergent fiber

A. Reagent preparation

1) Neutral detergent solution

Weigh 18.6 g of disodium ethylenediaminetetraacetate dihydrogen dihydrate, 6.8 g of sodium tetraborate decahydrate, and 4.6 g of disodium hydrogen phosphate, put in a 1-L volumetric flask, and dissolve by the addition of 500 mL of water. To this solution, add 30.0 g of sodium n-dodecyl sulfate, 10 mL of triethylene glycol and 250 mL of water, mix, and further add water up to the marked line of the volumetric flask to prepare neutral detergent solution.

Check that the pH is in the range of 6.95-7.05 before use.

2) Amylase stock solution

Put 1 mL of heat-stable α-amylase in a 10-mL amber volumetric flask, dissolve by the addition of water, and further add water up to the marked line to prepare the amylase stock solution.

3) Amylase solution

Check in advance the sufficient volume a μL of the amylase stock solution to add by a confirmatory test. Transfer the volume of the amylase stock solution to

take b µL calculated by the following formula to a 100-mL amber volumetric flask, and add water up to the marked line to prepare the amylase solution.

Volume of amylase stock solution to take b (µL) =

sufficient volume of amylase stock solution to add a (µL) ×50

4) Iodine solution

Dissolve 2.0 g of potassium iodide and 1.0 g of iodine in water to be 100 mL.

B. **Quantification**

Weigh accurately 0.5 g of an analysis sample, put it in a 500-mL tall beaker, add 0.5 g of sodium sulfite and 50 mL of neutral detergent solution, and then cover the tall beaker with a watch glass or a condenser, and heat until it boils by a fiber boiler heated in advance. Immediately after the start of boiling, add 2 mL of the amylase solution to the tall beaker, and boil for 1 hour supplying evaporating moisture. Remove the tall beaker from the fiber boiler, further add 2 mL of the amylase solution to the tall beaker, mix by shaking mildly, and then leave at rest for 60 seconds. Filter by suction the content of the tall beaker using a glass filter*6 (heated at 520-550 °C for 2-5 hours, incubate at 150 °C for 2 hours, cooled in a desiccator, and weighed accurately in advance). Wash the residue in the glass filter (neutral detergent-insoluble fraction) with 40 mL each of hot water for 3 times, wash further with 10-20 mL of acetone for 3-4 times, and then air-dry until the acetone odor disappears.

Then, dry the said glass filter at 135±2 °C for 2 hours, let it stand to cool in a desiccator, and then weigh accurately to calculate the content (%) of neutral detergent-insoluble fraction (aNDF) in the sample.

Moreover, heat the said glass filter at 520-550 °C for 2-5 hours to incinerate neutral detergent-insoluble fraction, incubate at 150 °C for 2 hours, let it stand to

cool in a desiccator, and then weigh accurately to obtain the ash content (%) in the sample.

Subtract the ash content obtained in advance from the neutral detergent-insoluble fraction content to calculate the heat-stable α-amylase-treated neutral detergent fiber (aNDFom) content (%) in the sample.

* 1. Store neutral detergent fiber at room temperature.

2. α-Amylase A3306, α-Amylase A3403 (both Sigma) or equivalents

3. Store the amylase stock solution at 2-8 °C.

4. Confirmation test

Weigh accurately 0.5 g of coarsely ground hominy (corn kernel endosperm)[10] (that passes a sieve of 1-mm mesh), put it in a 500-mL tall beaker; prepare 6 sets of this. Add 50 mL each of the neutral detergent solution to tall beakers, cover the tall beakers with a watch glass or a condenser, and heat by a fiber boiler heated in advance. Immediately after the start of boiling, add amylase stock solution 25, 50, 100, 200 and 400 μL (which contain 125, 250, 500, 1000 and 2000 units, respectively, when the enzyme activity of the heat-stable α-amylase is 50000 unit/mL) to the tall beakers, respectively. Do not add the amylase stock solution to a set of the tall beaker to be the blank test solution. Boil them accurately for 10 minutes. Remove the tall beakers from the fiber boiler, further add the amylase solution of the same amount as the previous step to the tall beakers, mix by shaking mildly, and then leave at rest for 60 seconds. Filter the content of the tall beakers using glass fiber filter paper (GA-100 (Toyo Roshi) or equivalents) into 100-mL beakers[11], cool in an ice bath for 5 minutes to be 1 °C or less, and return to normal temperature in an incubator at 20 °C.

Place the beakers on white paper, add 0.5 mL of the iodine solution quickly, leave at rest for 90 seconds, and check the color of the solution within 30 seconds. It is judged that the volume of the amylase stock solution added is sufficient when the solution is colorless to yellow, and insufficient when the solution is purple to pinkish light brown[12]. If the volume added was judged as insufficient at 400 µL of the amylase stock solution, conduct the same test using increased volume to add, and check the sufficient volume of the amylase stock solution to add.

5. Weigh immediately before use.

6. P2 (pore size 40-100 µm, Foss Tecator) or equivalents

«Summary of analysis method»

In this method, feeds are boiled and treated with heat-stable α-amylase simultaneously in a solution in which a neutral detergent is dissolved to remove starch and quantitate neutral detergent fiber. α-amylase-treated NDF is described as aNDF, while NDF without α-amylase treatment is described as NDF. NDF that does not contain ash is described as NDFom.

When the method is applied to feed rice, starch degradation may not be sufficient with the optimal amylase amount obtained by the confirmatory test depending on the rice cultivar. If amylase is changed from the usual method to the amount that can degrade starch sufficiently, it is required to describe as such in the analysis result.

Flow sheet of the analysis method.

0.5 g sample

500-mL tall beaker

0.5 g sodium sulfite

50 mL neutral detergent solution

Fiber boiler

Add 2 mL of α-amylase after boiling

Add 2 mL of α-amylase after boiling for 1 hour

Filter by suction

Wash 3 times with 40 mL of hot water

Wash 3-4 times with 10-20 mL of acetone

After air-drying, dry (135±2 °C, 2 hours)

Weighing

Incineration (520-550 °C, 2-5 hours)

Weighing

Precautions and Notes

[1] If it is stored in a cold place, warm it to 25 °C before use, and the solution should be uniform to be used.

[2] If it cannot be dissolved, dissolve by heating.

[3] When the pH is not within the range, adjust using hydrochloric acid and sodium hydroxide. If the pH is different by not less than 0.5, prepare newly the neutral detergent solution.

[4] Defat with acetone when the fat content in the sample is not less than 5 %. Acetone defatting should be conducted as shown below:

Weigh accurately 0.5 g of an analysis sample, put it in a glass filter, add 40 mL of acetone to immerse, and filter while mixing by stirring for 2-3 times. Repeat the procedure 3 times. On the third filtration, air-dry at room temperature until the residual acetone odor disappears, transfer to a tall beaker using 50 mL of the neutral detergent solution, and the add 0.5 g of sodium sulfite. The glass filter used here should be used in filtration after boiling.

[5] When a fiber extractor is used, weigh an analysis sample in a glass filter, further add 0.5 g of sodium sulfite, load on the fiber extractor, and then add 50 mL of the neutral detergent solution.

[6] If the content adhered to the inner wall of the tall beaker, drop it off so that the content is continuously treated with the neutral detergent solution. Make sure to keep the neutral detergent solution concentration constant by adding hot

water when moisture is evaporated. When a fiber extractor is used, use back pressure so that the content adhered to the inner wall of the glass filter is treated by the neutral detergent solution.

[7] This is because the rapid temperature decreasing in the glass filter from high temperature (520-550 °C) to room temperature causes distortion between the glass part and the filter part of the glass filter.

[8] Check every 6 months; and conduct when the lot is different.

[9] When the method is applied to high-starch feeds such as feed rice, it may not be sufficient with the optimal amylase amount obtained by the confirmatory test depending on the cereal cultivar.

[10] Coarsely ground hominy (corn kernel endosperm) is distributed for a charge by the Committee of Feed Quality Improvement.

There may be changes in the distribution method, etc.; see the website of FAMIC for the purchase procedure.

[11] If the blank test solution cannot be filtered, the supernatant can be used. [12] Examples of solution colors are shown below:

6. Acid detergent fiber (ADF and ADFom)

[Summary of acid detergent fiber]

The analysis method for acid detergent fiber (ADF) comprehends the fiber fraction in cattle feed, measuring fractions with no or low digestivity in fiber. The major components of ADF are cellulose and lignin.

Determination Methods

1. Acid detergent fiber

A. Reagent preparation

Acid detergent solution*1

To 1 L of sulfuric acid (1+37), add 20 g of cetyltrimethylammonium bromide and dissolve.

B. Quantification

Weigh accurately 1 g of an analysis sample, put it in a 500-mL tall beaker[1], add 100 mL of the acid detergent solution, and then cover the tall beaker with a watch glass or a condenser, and boil. After boiling for 1 hour supplying evaporating moisture[2], filter by suction the content of the tall beaker using a glass filter*2 (heated at 520-550 °C for 2 hours, incubated at 150 °C for 2 hours[3], cooled in a desiccator, and weighed accurately in advance). Wash the residue in the glass filter (acid detergent-insoluble fraction) with hot water sufficiently, wash further with 10-20 mL of acetone for 3-4 times, and then air-dry until the acetone odor disappears.

Then, dry the said glass filter at 135 °C for 2 hours, let it stand to cool in a desiccator, and then weigh accurately to calculate the content (%) of acid detergent-insoluble fraction (ADF) in the sample. Moreover, heat the said glass filter at 520-550 °C for 2 hours to incinerate acid detergent-insoluble fraction,

incubate at 150 °C for 2 hours, let it stand to cool in a desiccator, and then weigh accurately to obtain the ash content (%) in the sample.

Subtract the ash content from the acid detergent-insoluble fraction content obtained above to calculate the acid detergent fiber (ADFom) content (%) in the sample.

Summary of analysis method

In this method, feeds are boiled within an acid treatment solution in which a detergent is dissolved to quantitate acid detergent fiber (ADF). ADF that does not contain ash is described as ADFom.

Flow sheet of the analysis method is shown in Figure 3.6-1.

1 g sample

500-mL tall beaker

100 mL acid detergent solution

Fiber boiler

Boil for 1 hour
Filter by suction
Wash 3 times with 40 mL of hot water
Wash 3-4 times with 10-20 mL of acetone
After air-drying, dry (135±2 °C), 2 hours

Weighing

Incineration (520-550 °C, 2-5 hours)

Weighing

Flow sheet of the analysis method for acid detergent fiber

Precautions and Notes

[1] When a fiber extractor is used, weigh an analysis sample in a glass filter, load on the fiber extractor, and then add 100 mL of the acid detergent solution.

[2] If the content adhered to the inner wall of the tall beaker, drop it off so that the content is continuously treated with the acid detergent solution. Make sure to keep the acid detergent solution concentration constant by adding hot water when moisture is evaporated. When a fiber extractor is used, use back pressure so that the content adhered to the inner wall of the glass filter is treated by the acid detergent solution.

[3] This is because the rapid temperature decreasing in the glass filter from high temperature (520-550 °C) to room temperature causes distortion between the glass part and the filter part of the glass filter.

7. Crude Ash (CA)

[Summary of crude ash]

A sample is incinerated by heating to be crude ash.

When a sample is incinerated without special treatment, there always is contamination with charred organic matter, resulting in a blackish color. Therefore, it cannot be considered as pure ash (inorganic salts), and is referred to as crude ash.

Ash in feeds is useful for judging nutritional characteristics of the feed because ash has generally constant element composition by feed material type as long as the feed does not contain earth and sand, etc. Ash content in feeds of plant origin is not very good as an nutritional indicator because: it varies widely; silicate accounts for a large percentage of ash; and the other element composition is apt to vary depending on soil and fertilizers.

[Methods listed in the Analytical Standard of Feed]

1. Ignition [Analytical Standard of Feed, Chapter 3, 7]

Quantification

Weigh accurately 2-5 g of an analysis sample and put it in a crucible[1] (heated at 550-600 °C for 2 hours, cooled in a desiccator, and then weighed accurately in advance). Heat this gently to be charred, then ash at 550-600 °C for 2 hours, let it stand to cool in a desiccator, and accurately weigh to calculate the crude ash content in the sample.

«Summary of analysis method»

Crude ash is defined as the weight of a sample measured after heating at 550-600 °C for 2 hours to incinerate in a crucible.

«Notes and precautions»

[1] The container for incineration can be any of ceramic, platinum, and Pyrex; however, when crude ash is to be used for other elemental analysis after quantification, a platinum container is preferred depending on the purpose. A ceramic crucible can be about 40 mm in inner diameter, and about 37 mm in height.

A crucible stand shown in Figure 3.7-1 is convenient that it can be placed as is in a desiccator (with a stopcock on the top). This product is commercially available from Sanshin Industrial.

[2] For feeds with high sugar content or feeds of animal origin, the sample may be expanded and overflow out of the crucible; heat carefully.

[3] The cooling time should be the same as in cooling in the pre-heating.

[Other analysis methods]

2. Crude ash quantification by international standards

Determination of crude ash

Flow sheet of the analysis method

Sample	5.0 g (Put in a charring dish (crucible) weighed in advance)
Hot plate	Heat gradually until charred.
Muffle furnace	550 °C, 3 hours - Heat for another 1 hour if OM particles are observed.
Cooling	Desiccator
Weighing	

8. Nitrogen Free Extracts (NFE)

Subtract the contents (%) of the following 5 ingredients, moisture, crude protein, ether extract, crude fiber and crude ash, from the whole feed, and the result is shown as nitrogen free extracts (NFE) (%).
The content of NFE is those that are not contained in the 5 ingredients shown above, and the major ingredients are soluble carbohydrates such as starch and sugars, as well as organic acids and lignin. NFE is an important nutrient as an energy source for animals.

[Methods]

1. Calculation of Nitrogen free extracts

Quantification

Calculate the nitrogen free extracts content by the following formula:

Nitrogen free extracts content (%) = 100 − { moisture content (%) + crude protein content (%) + ether extract content (%) + crude fiber content (%) +crude ash content(%)}

Some crucial nutritional analysis

1-Determination of Silage pH

Scope: This method can be applied to the determination of pH in silages and forages.

Principle: Distilled water is added to a silage sample and the pH recorded using a pH meter.

Equipment:

pH Meter

pH Electrode

250mL Beaker

Scissors

Reagents:

1. Commercially available 4.0 and 7.0 buffer solutions
2. Potassium Chloride (KCl)

Procedure:

1. Calibrated pH Meter

Turn the pH meter on and stabilize.

Calibrate pH meter using 4.0 and 7.0 buffers

Once calibrated, rinse electrode thoroughly using distilled water.

Place electrode into a beaker of distilled water until required. The pH meter is now ready to read samples.

2. Chop the silage

Consistent results are obtained if the silage sample is finely chopped (size less than 0.6cm). This easily be achieved by chopping the material in a rotating bowl chopper. Alternatively the sample can be cut finely using scissors.

3. Preparation for pH measurement

Weigh out 20 g (+/-0.2g) fresh silage into a small beaker and cover with

100 ml distilled water.

4. Record pH

After allowing the sample to stand for 30 minutes at room temperature, place the electrode into the silage solution and read the pH to two decimal places after allowing the pH meter to equilibrate for 30 seconds (It is important at this stage to ensure the electrode is immersed in the solution and is not obstructed by the forage matter being tested.

Care about that rinsing electrode thoroughly with distilled water between measurements.

Store the electrode in 0.1M KCl solution following use.

Comments:

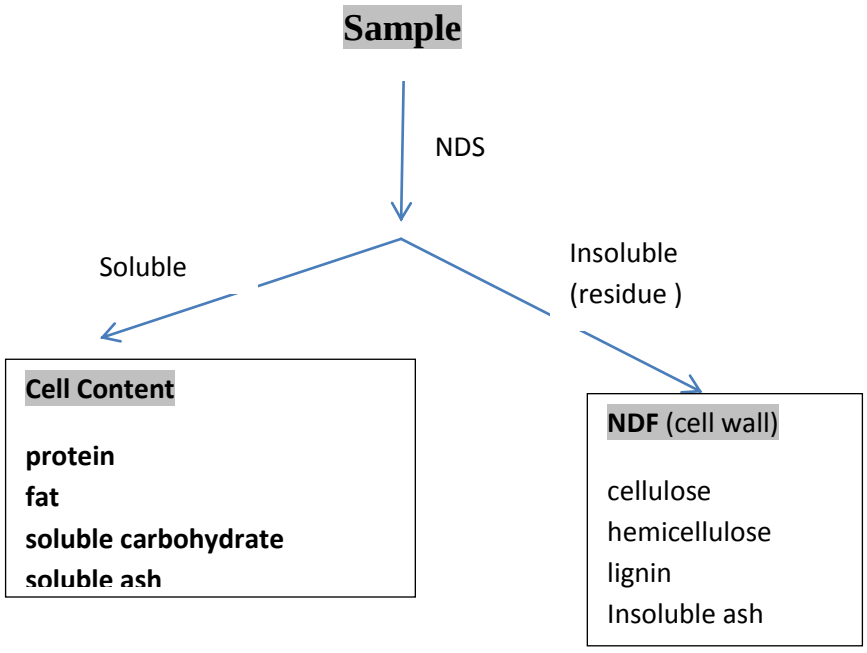
• Silage is a fermented feed and the silage pH can be used as an indicator of silage fermentation quality. The pH of well preserved silages is usually in the range 3.6-4.0. Higher pH values may indicate poor silage fermentation. In drier silages bacterial growth, and hence the fermentation, is restricted so the resulting silage pH is higher. For an interpretation, pH use as an indicator of silage fermentation quality.

• Silage is a perishable product and should be stored in an anaerobic condition at low temperature. If the sample is not to be analyzed immediately it should be frozen.

• If the sample is exposed to air it will decompose.

Proximate analysis limitations				
Component	Content supposed	contains	Missing	Excess
Crude fiber	Fibrous matter	Cellulose Lignin	Hemicellulose Part of lignin Insoluble ash	None
NFE	Soluble carbohydrate	Soluble carbohydrate Hemicellulose Part of lignin Insoluble ash	None	Hemicellulose Part of lignin Insoluble ash

VAN SOEST PROCEDURE



Exercise

1. **What are the limitations of proximate analysis?**
2. **How does Van-Soest analysis procedure avoid proximate analysis limitation?**
3. **Identify the main principle of kjeldahl method?**
4. **A feed has the following analysis :**

NUTRIENTS	%
CP	12
CF	4
ASH	1.2
NFE	20
EE	4
Ca	0.6

This composition on fresh basis

a) **What is the DM % on DM basis?**
b) **What is the DM % on oven DM basis?**
c) **What is the CP % on DM basis?**
d) **What is the water content?**

Task 1……

Task 2…….

Task 3……

Task 4……

Digestibility

Digestible part of foods or nutrients means the portion which is absorbed by the animal, which is not egested.

Digestible compounds:

protein	amino acids
starch	glucose
fats	fatty acids, monoglycerides

Calculation:

Digestible nutrient content = nutrient ingested – nutrient egested

Can be expressed as a coefficient or a percentage.

Apparent or True digestibility

The excreta contains not only the unabsorbed portion of food, but also endogenous compounds (erosion of epithelium, enzymes etc.).

True digestibility: correction with the endogenous compounds

Apparent digestibility: without correction with the endogenous materials

Apparent digestibility %

=((nutrient intake–nutrient excreted)/nutrient consumed)*100

True digestibility %

=((nutrient intake–nutrient excreted–endogenous loss)/nutrient consumed)*100

N.B: Endogenous losses is important mostly in the case of proteins and amino acids.

Endogenous losses in the faeces:

Sloughed off intestinal cells

Digestive juices (enzymes)

Microbial matter

Determination of digestibility

A***Animal experiments in vivo,

a. *Simple digestibility trial* (if the feedstuff could be given to the animals as the sole item of diet, corn with poultry species, hay for ruminants)

1- Preliminary period
2- Experimental period

Digestibility trial issues

Changeover designs

Necessary if period effects are an issue e.g.
Animal physiological changes
Forage physiological changes

Adaptation period

Necessary to adapt the animals to

New feed (microbial population changes)
Strange equipment
Strange housing

Period (6 –14) day is the normal

In the trial period the feed and the faeces must be measured exactly.

$$\text{Digestibility coefficient (\%)} = \frac{C-E}{C} \times 100$$

Where:

C = ingested nutrient content

E = egested nutrient

Digestibility (%) = (Nutrient in feed -Nutrient in feces/ Nutrient in feed) x 100

DM digestibility (DMD, g/kg) =(DM in feed -DM in feces /DM in feed) x 100

Organic matter digestibility(OMD, g/kg) = (OM in feed -OM in feces/ OM in feed) x 100

Can be expressed as a proportion, % or g/kg

b. Difference method (if the feedstuff cannot be given to the animal alone, otherwise it can cause digestive disturbances; for example cereal grains with ruminants)

Digestibility coefficient (%)= $\frac{A-(B \times b)}{d}$

where:

A= the common digestibility coefficient of the two feedstuffs

B= the digestibility of the accompanying feedstuff

b= ratio of the accompanying feedstuff

d= ratio of the test feedstuff

The animal number needed for digestibility trials

The ratio of the test feedstuff in the ratio (%)	number of animals
ruminants	
100	3
50	4
25	6
<25	8
poultry	
100	3
<50	6-8

The length of digestibility trials

	preliminary period	test period
ruminants		
substantial change in the feed	20	6-10
small change in the feed	10	6-10
poultry	5	5

c. Indicator method

Procedure

–Add indigestible marker to feed e.g chromic oxide.

–Measure concentration in feed & feces

–Estimate disappearance of marker from gut.

E.g. if a feed contains 1% Cr2O3 & feces contains 2% Cr2O3, diet digestibility = 50%
–Since Cr3O2 conc. has doubled, 50% of DM must have been digested

Particularly useful for grazing animals

- Do not have special balance cages,
- Total collection is not possible, (like ileal sampling),
- Taking representative excreta samples enough.

Indicators, markers:

--External (Chromic oxide, Dysporium, Polyamide etc.). Can contaminate forage
--Internal Indigestible natural compounds of the feed (Lignin, AIA, ADF, n-alkanes). Easier, less labor.

Indicators characteristics:

- Should not be absorbed
- Should not disturb digestion of nutrients
- Its transit time should be steady
- Its analysis should be accurate
- Could be incorporated into the test diet homogenously

$$\text{Digestibility coefficient (\%)} = \frac{A-(B \times It/Ib)}{A}$$

Where:

A= nutrient content of the feed

B= nutrient content of the faeces

It= indicator content of the feed

Ib= indicator content of the faeces

For the digestibility of a specific nutrient, must also know the % nutrient in feed & feces:

$$\%\text{Nutrient Digestibility} = 100 - (100 \times ((\%\text{ indicator feed}/\%\text{ indicator feces}) * (\%\text{ nutrient feces}/\%\text{ nutrientfeed}))$$

d. Continuous digestibility trial

- For measuring the digestibility of herbage by grazing animals.

- Lignin or acid insoluble ash can be used as an indicator.
- When fibre and lignin content of grasses increase, it reduce the digestibility of starch, fat and protein. So, the optimal grazing or harvest time can be determined.
- Representative faecal samples are collected after several days and the nutrient and lignin content of both grass and faeces are determined.

Problems with *in vivo*experiments

Animal trials are:
–Expensive

–Protracted

–Laborious

–Public concerns

–Animal stress.
Must estimate nutritive value with less animal dependent techniques

B Laboratory methods of estimating digestibility* in vivo,**

Ideal *in vitro* methods should be:
–Rapid (one step) & routinely practicable
–Accurate
–not expensive & not laborious
–Repeatable & robust
–Biologically meaningful
–Broad-based (apply to all forage types)
–Handle large no. of samples
–Laboratory-based

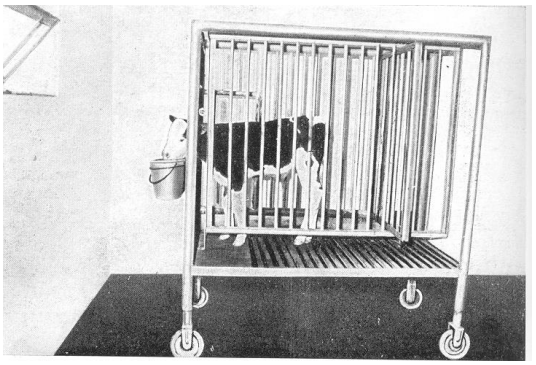
- **Protein digestion in non-ruminants** can be determined after incubation (39°C) the feed with pepsin and HCL acid, or by digestive tract secretion collected via cannula.
- **In the case of ruminants** the finely ground sample is incubated for 48hrs with rumen fluid under anaerobic conditions. After that the bacteria are killed by acidifying with HCL acid and then digested with pepsin for a further 48hrs. The insoluble remedies are filtered off,

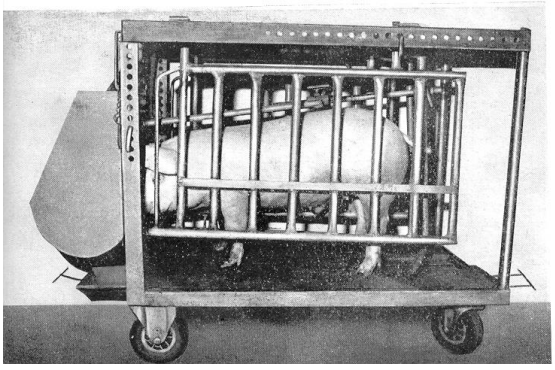
and its nutrient content is subtracted from the nutrient content of the feed. The difference is the digestible part.

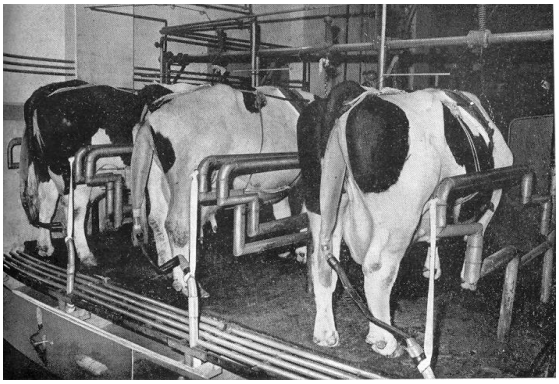
- **Rumen fluid problems**

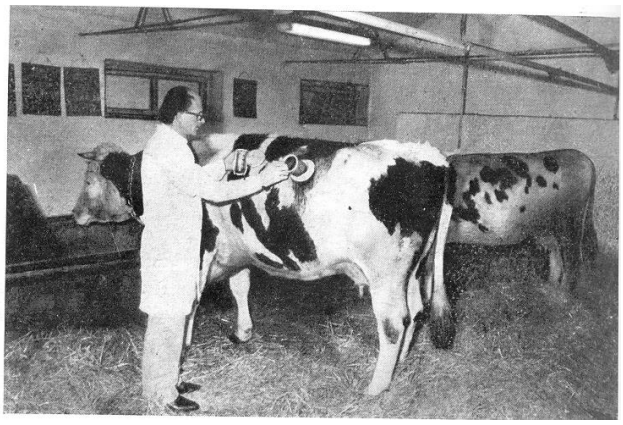
  Variation in Inoculum composition & activity due to

  - Host animal diet
  - Animal species
  - Collection time
  - Processing (blending vs. filtration)
  - Analytical issues
    - –Maintenance of anaerobic media; optimal pH, temp
    - –High viscosity hinders filtration
    - –Offensive odors
    - –Hygiene –(Prevent pathogen infection)









Factors affecting digestibility :

a- Animal factors :

Species (different digestive characteristics)

Breed (not significant differences found between various breeds of the same species, individual differences are sometimes higher)

Age *of the animal* (young animals with less enzyme secretion, so cannot digest the feeds as efficient as the adults)

Accustomed to a feed (changing feed composition suddenly decreases digestibility mainly in the case of ruminants)

b. Food factors:

Fibre content (if the fibre is too high, it negatively effect on the digestibility of the other nutrients)

Carbohydrates (sugar content of the food can cause depression in the rumen fermentation)

Enzyme supplementation of the food (it can be advantageous in young animals or against anti-nutritive factors)

The amount of food eaten. (If the consumed amount be increased, the transit time of digesta will be decreased, so the digestibility of nutrients will be less.

Preparation of foods (grinding, pelleting, heating and steaming of soybean, boiling of potato etc. helps in digestion)

Anti-nutritive compounds *of feed* (trypsin inhibitors, solanin, glucans and xylans etc. decrease digestion)

Digestibility indices that estimate energy value

Digestible organic matter content (DOMD) (% DM) =

(OM in feed -OM in feces/ DM in feed) x 100

TDN = DCP + DCF + DNFE + DEE(2.25)

- –DCP= Digestible Crude Protein
- –DCF= Digestible Crude Fiber
- –DNFE= Digestible Nitrogen-Free Extract
- –DEE= Digestible Ether Extract (2.25)

1. Calculate the digestibility coefficients of alfalfa hay, which was determined with sheep. Animals were fed 1,5 kg hay/day, their excreted 1,8 kg faeces/day.

	alfalfa hay (g/kg)	faeces (g/kg)	digestibility coefficient (%)	digestible nutrient content (g/kg)
dry matter	875	240		
crude protein	200	70		
crude fat	25	4		
crude fibre	272	88		
N-free extract	404	84		

2. Calculate the protein digestibility coefficient of barley protein. A difference trial was carried out with sheep.

- As an accompanying feed 1kg hay per day was fed in the first experiment. The daily faeces excretion was 1.2 kg. The crude protein content of the hay and faeces were 115 and 42 g/kg respectively.
- In a second experiment beside 1 kg hay 0.5 kg barley was fed. The crude protein content of the barley was 116 g/kg. In this experiment the daily amount of faeces was 1.5 kg, its protein content was 4%.

3. Calculate the fat digestibility coefficient and digestible fat content of a broiler diet.

According to the results of an indicator trial the fat and Cr2O3 content of the diet were 6% and 0.3% respectively. The same parameters of the faeces were 1.2% and 0.95%.

4. If lambs are fed a grass diet containing 7% protein & 1% chromic oxide, and their feces contains 5% CP and 2% chromic oxide. Calculate CP digestibility???

Task 1……

Task 2…….

Task 3……

Task 4……

Classifications of Feeds

All feedstuffs can be classified into categories based on their primary function in the animal diet, whether the feed is a protein or energy feed, for example. A good understanding of the classification of feeds and the nutrient content of popular feeds will make it faster and easier for producers to make feeding decisions.

Feeds classified into two main groups:

1- Roughage/forage or

2- Concentrates.

Feeds can be further sub-classified as shown in Figure 1. A variety of forages, grains, oilseeds, by-products, and crop residues can be used as feed to cattle. Some of the common feeds are described briefly next. Feed values are listed on a dry matter (DM) basis because dry matter contains all of the important nutrients, and we have to make one base for comparing. This is the nutrient values of the feed if all water was removed. This makes it easy to compare feeds and evaluate their nutrient composition.

A feed with 90% DM (i.e. corn) is a dry feed in its everyday form, whereas a 30% DM feed is a wet feed (i.e. silage). We could not be able to compare these two feedstuffs unless we look at the nutrient values on a DM basis.

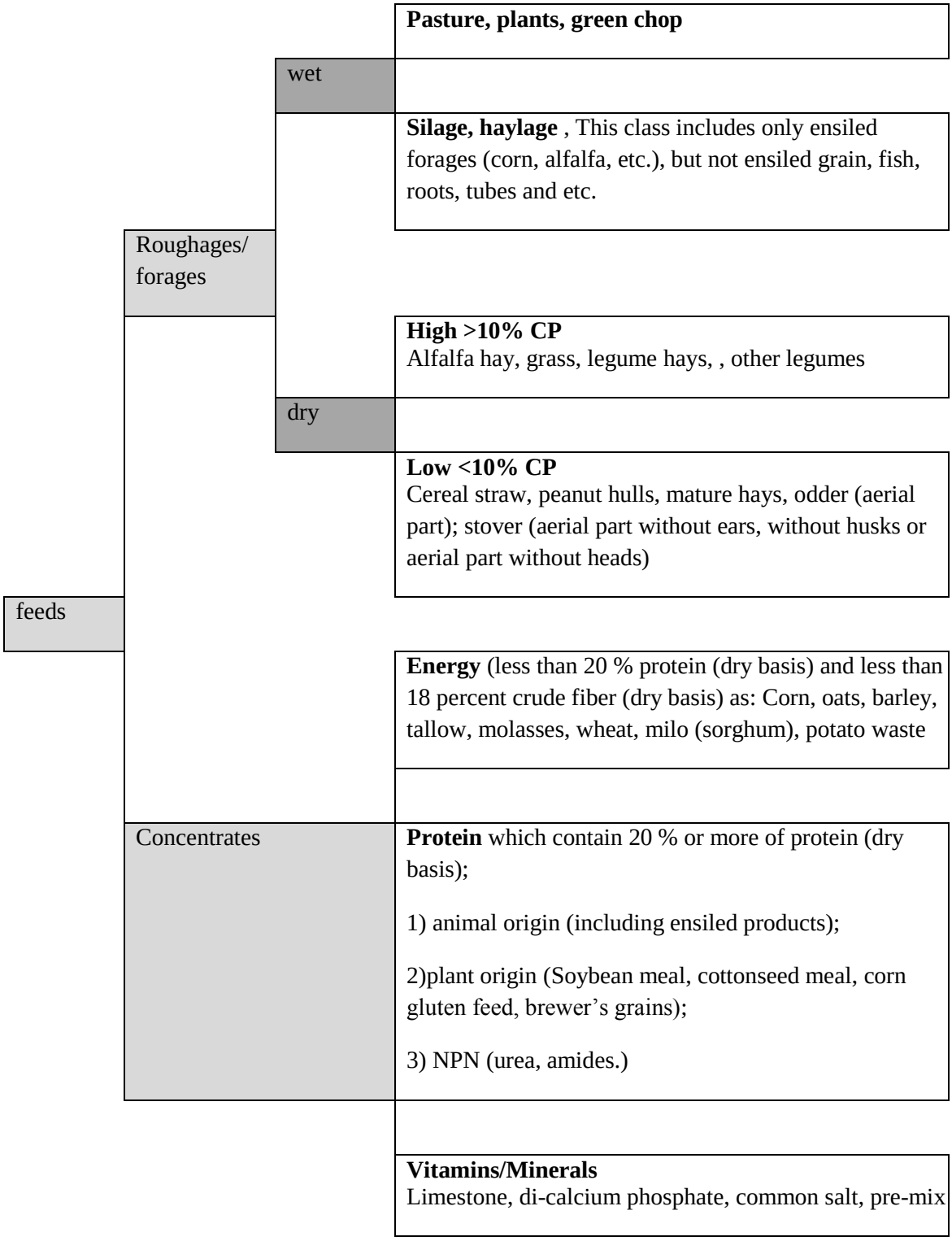


Fig (1) Classification of feedstuffs

Another Classification of Feeds

1. Carbonaceous concentrates (low protein)

2. Carbonaceous roughages (low protein)

3. Proteinaceous concentrates

(1) Supplements of plant origin

(2) Supplements of animal origin

a. Animal tissue residues

b. Fish products residues

c. Milk products(dry milk, milk replacers)

(3) Supplements non protein nitrogen

4. Proteinaceous roughages

5. Additive materials (Nutrients and non-nutrient additives)

I. Common Characteristics of Concentrate

A. Carbonaceous concentrates (high-energy feeds; mostly grains and their by-products. According to NRC, these are products containing less than 20% protein and less than 18% fiber.)

1. **Common characteristics**

a. High in energy (TDN)

b. Low in CF

c. Low in protein (in relation to oil seeds and mill feeds)

d. Protein quality (generally quite low)

e. Mineral content

f. Vitamin content

2. **Important carbonaceous concentrates**

a. Corn (widely used, 87% TDN;8% CP).

b. Sorghum grains (milo, hybrids, etc., 82% TDN;11% CP)

c. Oats-(76% TDN; 13% CP)

d. Barley-(84% TDN; 12% CP)

e. Rye-(75% TDN; 12% CP)

f. Wheat-(80% TDN; 12-14% CP)

g. Corn-and-cob meal-(73% TDN; 7-8% CP)

h. Dried beet.pulp-(65-70% TDN; 8-10% CP)

i. Molasses (over 48% sugar, 55-75% TDN), table 1.

B. Proteinaceous concentrates

1. Protein quality: it Refers to:-

a. Kinds, amounts and/or ratio of amino acids in a feed

b. Variability in composition (in non-ruminants). Supplementation of deficient amino acids will be important or a mixture of feeds provided to insure intake of all essential amino acids.

c. Non- protein nitrogen (urea) may be fed to ruminants but not poultry

d. All amino acids are essential to life. Some must be supplied in diet of non-ruminants.

2. The essential amino acids and sources:

a. Lysine

b. Methionine

c. Tryptophan

d. Threonine, Histidine, Arginine, Isoleucine, Valine and Phenylalanine - Grains fair source

e. Leucine

f. Glycine (with glutamic acid required by the chick). Corn is a good source.

g. When feeding poultry with high-grain rations, three specific amino acid contents must be carefully checked. They are:

(1) Lysine; (2) Methionine; (3) Tryptophan

3. Some protein supplements.

a. Urea (a source of nitrogen for protein synthesis). Ruminal bacteria convert it into microbial protein (1 kg. of urea contains as much nitrogen as 2.9 kg. of protein)

b. Supplements of plant origin (oilseed meals)

Manufacturing process for oil meals; (a) Expeller or hydraulic process, where seed is crushed, heated (steam or cooked), pressed and ground; (b) Solvent process, where seed is cracked, heated, rolled, extracted with hexane; flakes are then toasted and ground

Effect of processing; (a) Hydraulic or expeller processed meals contain more fat and less protein than solvent extracted. (Solvent process less than 1% fat; Expeller 4-5% fat); (b) Cooking improves palatability, color and increases protein quality. However, over heat can destroy some amino acids; (c) In soybeans, cooking destroys deleterious substances (antitrypsin factor) which suppresses growth and prevents trypsin action; also destroys urease enzyme.

Characteristics of the oil meals;

(a) Soybean meal

(1) Nutritive characteristics

(a) Solvent: 44 to 48% of crude protein

(b)Dehulled: 50 % of crude protein

(2) Must be heated or toasted

(3) Considered a high quality plant protein and gives good results in poultry rations with B-vitamins supplementation.

(4) On the farm processed soybeans

(a) Nutrients contents: (37-38% CP, 17% EE, 87-88% TDN)

(b) Special equipment will be needed to heat beans to a certain temperature which will destroy the Antitrypsin factor and the urease enzyme (undesirable in diet, containing urea)

(c) Using whole cooked soybeans will likely improve feed efficiency (4-8%) due to higher fat content.

(b) Cottonseed meal (commonly used in southern states)

(1) Nutritive characteristics

(a) 41-43% crude protein

(b) Low protein quality

(2) Danger in poultry:

(a) Contains gossypol (.03-.2%), toxic substance.

(b) signs of gossypol poisoning are similar to pneumonia except fluid develops in abdominal cavity which is typical of poisoning.

(c) cause green yolks and pink egg whites in eggs

(c) Supplements of animal origin

(1) Tankage (meat meal tankage & digester tankage)

(a) 55-60% crude protein (60%)

(b) If over 4.4% phosphorus, “meat and bone tankage”

(c) Contains blood meal, tendon, gut, and connective tissue which reduces biological value of the protein

(d) Excluded of hair, hoof, horn, manure, stomach contents and hide trimming

(2) Meat meal (Meat scraps)

(a) Contains 45-55% crude protein (55%)

(b) If over 4.4% P, “meat and bone scrap or meal”

(c) Does not contain gut, tendon, and connective tissue

(4) Fish meal

(3) Fish meal dried, ground whole fish

(1’) Crude protein (35-70%) according to product (whole fish or cutting)

(2’) High protein quality and good source of B-vitamins

(4) Milk products

(a) Whole milk (too expensive except to suckling animals)

(b) Dried skim milk (33% CP; EE removed)

(c) Dried buttermilk (32-33% CP; as skim milk, except slightly higher in EE)

(d) Dried whey (13% CP; casein and EE removed)

(5) Poultry by-products

(a) Poultry by-products meal.

(b) Hydrolyzed poultry feathers (Feather Meal).

Animal protein sources are generally used in poultry rations, while ruminants mainly use vegetable proteins and urea.

C. Feed grain by-products (also called mill feeds)

1. By-products of corn (from milling to manufacture corn starch, corn sugar, etc.)

a. Corn bran

b. Corn gluten meal (remains after removal of starch, germ and bran)

c. Corn gluten feed (remains after removal of starch, gluten and germ)

d. Corn germ meal (ground corn germ from which the soluble has been removed)

e. Hominy feed (corn bran, corn germ and a part of the starchy part on of the grain)

f. Distillers by-product feeds (made largely from corn and some rye); DDGS (distiller's dried grain with soluble)

2. Wheat by-products

a. wheat bran (outer coating of the kernel)

b. wheat middling (shorts); (consists of fine particles of wheat bran, wheat germ, wheat flour, and some offal from the "tail of mill")

c. wheat germ meal (consists chiefly of the germ, together with some of the bran and middling)

3. Oat by-products

a. Oat hulls (consists primarily of the outer covering of the grain)

b. Oat groats (hulled oats); (kernels produced from cleaned and dried grain which has had the hull removed)

c. Feeding oat meal (consists of broken rolled oat groats, oat groat chips and a small quantity of ground oat hull)

4. Barley by-products

Brewers dried grains, the dried extracted residue of barley alone or in mixture with other cereal grain or grain products resulting from the manufacturing of beverage.

5. Many of the by-product feeds are high in crude protein content. We should remember that the protein quality is similar as in the original cereal grains. These products are generally used to supply a part but not all of the protein for animal rations.

II. Roughage Characteristics

A. General characteristics

1. Low in energy and containing more than 18% CF.

2. Includes pasture, hay, silage; the crop may be the same but preservation procedures can influence the nutrient value.

3. Lower in energy and higher in fiber than concentrates. Protein content is variable.

4. Important for bulk in ruminant.

5. Higher in Ca and trace minerals than concentrates.

6. Legumes are higher in protein and B-vitamins.

7. Good sources of fat soluble vitamins

8. Palatable to ruminants

9. Limited or excluded in monogastric rations (not suited for finishing rations)

10. Limited in beef finishing rations and in high energy lactating rations

11. Required in lactating cattle rations to help maintain normal fat level in milk

12. More variable in nutritive values and palatability than concentrates because of variation in maturity degree and harvesting and storing procedures.

B. Proteinaceous roughages

1. Legume forages

a. Advantage

(1) Higher in protein than other forages

(2) Good quality protein

(3) High in Ca

(4) High in Vitamin A precursor activity at harvest

(5) Sun cured hay is rich in Vitamin D

(6) Good soil fertilizer

(7) Excellent in combination with grasses

b. Variety

(1) Alfalfa hay

(2) Alfalfa haylage (low moisture silage)

(3) Dehydrated alfalfa meal

(4) Other legume or legume grass mixture

(a) Red clover

(b) White clover

(c) Sweet clover

(d) Pasture mixes (alfalfa-brome; clover-grass)

2. Grass hays such as Brome, Timothy and various wild grasses may contain 10-14% crude protein (immature stage)

C. Carbonaceous roughages

Include:

1. Corn and sorghum silages

2. Sorghum pasture

3. Corn cobs

4. Stover

5. Straw

6. Ruminants make maximum use of low grade roughages, and must be supplemented with:

a. Protein

b. Readily available carbohydrate (starch from grain and/or sugar from molasses)

c. Additional minerals

d. Vitamin A

D. Soilage (green chop) refers to green forage taken directly from the field to the animals.

1. Advantages:

a. maximum yield per area; larger yield of nutrients in "hay stage" than if grazed.

b. Less waste of nutrients than with other harvesting procedures

c. Less fencing than if pastures; crop not trampled by livestock (tall crops, e. g., Sudan grass)

d. Reduce bloat problems with certain legume crops

e. Most commonly used in large dairy farms although not usually better than good pasture for dairy or beef cattle.

2. Disadvantages:

a. No uniform quality from day to day.

b. Weather can be a problem.

c. Expensive due to labor and machinery.

d. Could not have it around the year.

E. Hay

1-Good hay is; a) Cut at certain stage of maturity; b) Properly dried; c) Free from mold; d) Palatable; e). Less foreign material

2. Hay making

a. Purpose of curing is to dry down to 25 % moisture of less (22 % if to be chopped and stored)

F. Silages

1. Steps occur

a. **Plant cells respiration** continue till Oxygen is consumed and Carbon dioxide is produced (Mold will not grow)

b. **Temperature** of silage increases

(1) Desirable temperature of silage after respiration stops is 27-37˚C

(2) Lower temperature (lactic acid producing bacteria can not compete with butyric acid forming bacteria)

(3) Temperatures above 37 ˚C, preferable but much nutrient value has been lost

c. **Fermentation**

(1) Anaerobic condition necessary

(2) Bacteria (naturally present on field) begin to multiply and attack plant sugars, forming:

(a) Acetic acid; (b) Lactic acid; (c) Butyric acid; (d) Propionic acid

d. **The acidic pH** stop all bacterial growth and silage remains in a stable state in the silo.

2. Crops for silage

a. Corn

b. Sorghums

c. Small grains-oat crop

d. Legume or grass silage

3. Consideration for use of silage

a. Advantage

(1) Feed more livestock per area

(2) Harvesting, till feeding can be mechanized

(3) High-quality,

(4) Wide range of moisture (Earlier harvest)

b. Disadvantage

(1) Committed to livestock feeding

(2) Rate of gain is reduced when high levels are fed, which increases the amount of feed for maintenance

Ways to Measure Quality

Lignin- Non digestible part of a plant that is found within the structure of the plant itself

Lignin increases as fiber increase.

Young plants contain 20% crude fiber, but mature plant has 40% or more.

Digestibility decreases as fiber increases.

III. Characteristics of Common Nutrient "Additive" Feedstuffs

A. Sources of Minerals and their Potency (table 2)

1. Major Minerals (sources of minerals may be obtained individually or in some form of a mixture)

2. Trace minerals (trace minerals may be obtained individually or as a trace mineral mixture)

a. Iodine

b. Fe, Cu and Co

c. Potassium and Magnesium

d. Manganese

e. Zinc

B. Sources of Vitamins (table 3)

1. Vitamin A

2. Vitamin D (a. D2 (plant form); b. D3 (animal form))

3. Vitamin E(a. Vitamin E concentrates; b. Wheat germ meal or oil)

4. Vitamin K

5. B-vitamin concentrate

Methods of Feedstuff Preparation

A. Benefits of processing

1. Increase efficiency of handling

2. Increase efficiency of utilization

a. Palatability

b. Digestibility

(1) Increase surface area for bacterial and/or enzymatic activity

(2) Alter molecular structure to enhance digestion

B. Specific processes

1. Grinding

a. Feed particles may separate (all the course material in the middle and the powder around the edges)

b. Degree of fineness:

(1) More subject to wind loss

(2) Tend to "ball up" in animal's digestive system

(3) Result in cattle going off feed (reduced palatability)

(4) Does not "feed down" properly in a self-feeder

(5) Tends to have a faster rate of passage through digestive tract

(6) May cause digestive disturbances; (a) Stomach ulcers in swine; (b) Ruminal parakeratosis in feedlot cattle;

c. Grinding grain for sheep not necessary except for sorghum grains which could be coarsely ground.

d. Fine grinding of grain for dairy cattle will result in lowered milk fat production; grains should be coarsely ground; dry rolling and coarse grinding of corn are quite equal for beef cattle

f. Grinding hay for cattle and sheep

(1) As hay is fine ground, there is a noticed decrease in digestibility of the nutrients (due to a fast passage through GIT)

(2) Chopping hay results in increased intake, gain and efficiency compared to long hay.

(3) Chopping a poor quality hay increased intake(more advantageous than chopping a high quality hay)

2. Pelleting

(1) Advantages

(a) Reduced dustiness

(b) Less waste of fine particle in transport

(c) Prevent selective eating and feed wastage

(d) Reduced space of storage

(e) Increase utilization of fibrous part of feedstuffs

(f) Adaptable to bulk

(g) Partial gelatinization of starch making more susceptible to enzymatic action and better digestion

(2) Disadvantages

(a) Pelleting cost. (Quality sometimes not ideal)

(b) Transportation cost

(c) Improper procedures may cause feed spoilage

(d) Cereal grains require finer grinding

(e) High fat Rations are difficult to pellet

(3) In general, pelleting diets will result in increased feed consumption and improved feed efficiency

3. Steam Flaking of Grain

4. Roasted Grain: Roasting is a new process of treating dry corn before feeding. The dry corn is passed through a roaster of soybeans roaster type. The corn is heated to approximately 150˚C . DM content is reach to about 95 % or more, and the bulkiness of the grain is increased about 15percent.

Exercise

1. What are the categories of feedstuffs?

3. What is the difference between roughages and forage?

4. Define the energy feed? What is the definition of a protein feed?

5. Differentiate between protein supplements from the three major sources of protein supplements?

6. Compare between vitamin supplements and minerals?

7. What is the purpose of nonnutritive additive?

8. Differentiate between an energy feed and protein feed?

Table 1: Contents of crude protein and TDN % on dry matter basis in common feedstuffs		
Feedstuff	**Total CP, % DM**	**TDN, % DM**
Alfalfa cubes	18	57
Alfalfa, dehydrated,17% CP	19	61
Alfalfa, fresh	18	61
Alfalfa hay, early bloom	19	59
Alfalfa hay, midbloom	17	58
Alfalfa hay, full bloom	16	54
Alfalfa hay, mature	13	50
Alfalfa silage	18	55
Alfalfa silage, wilted	22	58
Alfalfa leaf meal	28	69
Alfalfa stems	11	47
Ammonium chloride	163	0
Ammonium sulfate	132	0
Bahiagrass hay	8	51
Bakery product, dried	12	90
Barley silage	12	59
Barley silage, mature	12	58
Barley straw	4	43
Barley grain	12	84
Barley grain, steam rolled	12	84
Beet pulp, wet	9	76
Beet pulp, dried	7	75
Beet pulp, wet, with molasses	10	77
Beet pulp, dried, with molasses	10	76
Bermudagrass, Coastal, dehydrated	16	62
Bermudagrass hay, Coastal	10	56
Bermudagrass hay	10	53
Bermudagrass silage	10	50
Birdsfoot trefoil, fresh	21	66
Birdsfoot trefoil hay	16	57
Blood meal	92	66
Bluegrass, Kentucky, fresh, early bloom	15	69
Brewers grains, wet	28	85

Brewers grains, dried	28	84
Bromegrass, fresh, immature	15	64
Bromegrass hay	10	55
Bromegrass haylage	11	57
Canarygrass hay	9	53
Canola meal, solvent	40	71
Citrus pulp, dried	7	79
Clover, ladino, fresh	25	69
Clover hay, ladino	21	61
Clover, red, fresh	18	64
Clover hay, red	15	55
Clover hay, sweet	16	53
Corn, whole plant, pelleted	9	63
Corn fodder	9	67
Corn stover, mature (stalks)	5	59
Corn silage, milk stage	8	65
Corn silage, mature, well eared	8	72
Corn grain, whole	9	87
Corn grain, rolled	9	87
Corn grain, flaked	9	93
Corn grain, high moisture	10	93
Corn and cob meal	9	82
Corn cobs	3	48
Corn screenings	10	91
Corn gluten feed	23	81
Corn gluten meal, 41% CP	46	85
Corn gluten meal, 60% CP	67	89
Cottonoseed, whole	22	95
Cottonseed, whole, delinted	23	95
Cottonseed hulls	4	45
Cottonseed meal, mechanical, 41% CP	45	80
Cottonseed meal, solvent, 41% CP	48	77
Diammonium phosphate	115	0
Distillers grain, wet	28	90
Distillers grain, barley	30	77
Distillers grain, corn, dry	28	90
Distillers grain, corn, wet	29	90

Distillers grain, corn with solubles	29	90
Distillers corn stillage	22	92
Distillers grain, sorghum, dry	32	85
Distillers grain, sorghum, wet	32	85
Distillers grain, sorghum with solubles	31	85
Distillers dried solubles	29	88
Fat, animal, poultry, vegetable	0	205
Feather meal, hydrolyzed	86	69
Fescue, Kentucky 31, fresh	15	64
Fescue hay, Kentucky 31, early bloom	18	65
Fescue hay, Kentucky 31, mature	11	52
Fish meal	66	74
Grass hay	10	58
Grass silage	11	61
Hominy feed	11	89
Lespedeza, fresh, early bloom	16	60
Lespedeza hay	14	54
Linseed meal, solvent	39	76
Meadow hay	7	50
Meat and bone meal, porcine/poultry	56	72
Molasses, beet	9	75
Molasses, cane	5	75
Molasses, cane, dried	10	74
Molasses, citrus	10	77
Molasses, wood, hemicellulose	1	76
Monoammonium phosphate	70	0
Oat hay	10	54
Oat silage	12	60
Oat straw	4	48
Oat grain	13	76
Oat groats	18	91
Oat middlings	17	90
Oat hulls	4	40
Orchardgrass, fresh, early bloom	14	65
Orchardgrass hay	10	59
Peas, cull	25	86
Peanut meal, solvent	50	77

Potatoes, cull	10	80
Potato waste, wet	7	82
Potato waste, dry	8	85
Potato waste, wet with lime	5	80
Potato waste, filter cake	5	77
Poultry byproduct meal	62	79
Poultry litter, dried	25	64
Poultry manure, dried	28	38
Prairie hay	7	50
Rice grain	8	79
Rice bran	14	68
Rice hulls	3	13
Rye grass hay	10	58
Rye grass silage	14	59
Rye grain	12	82
Sanfoin hay	14	61
Sorghum silage	9	59
Sorghum grain (milo), ground	11	82
Sorghum grain (milo), flaked	11	91
Soybeans, whole	40	93
Soybeans, whole, extruded	40	93
Soybeans, whole, roasted	40	93
Soybean hulls	12	77
Soybean meal, solvent, 44% CP	49	84
Soybean meal, solvent, 49% CP	54	87
Spelt grain	13	75
Sudangrass hay	9	57
Sudangrass silage	10	58
Sunflower seed, meal, solvent	38	65
Sunflower seed, meal with hulls	31	57
Sunflower seed hulls	4	40
Timothy, fresh, pre-bloom	11	64
Timothy hay, early bloom	11	59
Timothy hay, full bloom	8	57
Timothy silage	10	59
Triticale grain	14	85
Turnip roots	12	86

Urea, 46% N	288	0
Vetch hay	18	58
Wheat, fresh, pasture	20	71
Wheat hay	9	57
Wheat silage	12	59
Wheat straw	3	42
Wheat straw, ammoniated	9	50
Wheat grain	14	88
Wheat grain, hard	14	88
Wheat grain, soft	12	88
Wheat grain, flaked	14	89
Wheat grain, sprouted	12	88
Wheat bran	17	70
Wheat middlings	19	82
Wheat mill run	17	75
Wheat shorts	20	80
Wheatgrass, crested, fresh, early bloom	11	60
Wheatgrass, crested, fresh, full bloom	10	55
Wheat grass, crested, hay	10	54
Whey, dried	14	82

Table 2 Sources of Minerals and Their Potency			
Element	**Common source**	**Composition or potency**	**Remarks**
Calcium [a/]	Feeding bone meal	25% Ca	Also contains 18% CP and 11% p.
	Feeding bone meal	27% Ca	Also contains 12% CP and 14% p.
	Bone char	27% Ca	Also contains 13% p
	Tricalcium phosphate	13.5% Ca	9% P
	Dicalcium phosphate	21% Ca	19% P
	Monocalcium phosphate	18% Ca	21% P
	Ground limestone	38% Ca	Balance likely to be carbonate and mg
	Calcium carbonate	38% Ca	
	Oyster and other marine shells	38% Ca	Shells contain on the average 96% $CaCO_3$
Phosphoru	Bone meals and Ca phosphates	(above)	
	Rock phosphate	14% P	Rock phosphate is 75-80% tricalcium phosphate Not advised unless guaranteed to contain less than 1% fluorine
	Defluorinated rock	18% P	Should not contain more than 1 part fluorine to 100 part p
Iodine	Potassium iodide	76% I	K and Na salts may be used interchangeably
	Sodium iodide	84% I	
	Potassium iodate	59% I	
	Iodize salt		Stabilized iodine should be used,
			Amounts of iodine differ but 0.02%
			and 0.05% are commonly sold
Iron	Ferric oxide	35% Fe	
	Ferrous sulphate	20% Fe	commercial grade
	Reduced iron	80-100% Fe	May be 20% ferric oxide
Cobalt	Cobalt sulphate	34% Co	drench cobaltized salt, ingredient in the ration

Table 3: Some data on Vitamins Frequently Added to Rations					
Name	**Functions; units of potency, etc.**	**Deficiency symptom**	**natural sources**	**Other sources (synthetic)**	**amounts normally added/ ton meal**
Vitamin A (animal form) Carotene (plan form)	formulation and development of body cells; a growth; Involved in dim-light vision. 0.6 micro-gran carotene = 1 IU of vitamin A.	Retarded growth. Interference with reproduction. Impaire night vision.	Leafy forage, fresh or preserved. Milk or fish fat. Yellow corn.	vitamin A acetate, 000 000 IU/g. Mixed carotene 1000 000 IU/g.	1000000 IU
Vitamin D (D_2 is plant form; D_3 is animal form)	absorption of calcium bone formation. t sun effectively supplies animals needed vitamin D. Poultry do not utilize D_2	Rickets in young; osteomalacia in adults Swollen and sore joints. Poor reproduction.	Sun-cured, leafy hay to r animals (but not poultry).	Fortified fish oils, irradiated sterols fo both poultry and animals. Crystallin D_2 or D_3 4000000 IU/g.	200000 IU
Riboflavin	enzyme necessary for oxidation. Synthesize by micro-organisms ir herbivora.	Slow growth, nerve degeneration, diarrhea (calves). Curled toe paralysis, low egg	Liquid, condensed or dry milks. Dried leafy forage. DDG. Dried brewers' yeast.	Pure synthetic riboflavin, 400 000 BS units/g 1 g has potency of 100 lb skim milk powder.	300 mg
Pantothenic acid	Co-enzyme A, necessary for utilization of all energy-yielding nutrients.	None in herbivora. Stilted gait in pigs, especially with hind legs (often called goose-stepping). Dermatitis around bill and eyes in poultry.	Brewers' yeast. Dry milk or whey. Can molasses. Alfalfa.	Synthetic calcium pantothenate (92% pantothenic acid).	2 g
Vitamin B_{12}	A cobalt-containing substance active in treating pernicious anaemia. Probably active part of "the animal protein factor"	Poor growth, poor feathering, and poor hatchability. Its use usually increases gains of young by 10-20.	Feeds of animal or marine origin. Live meal.	Fermentation products and byproducts	5-10 mg for a young. Adul herbivora synthesize it.

EVALUATING FORAG QUALITY

ACTIVITY 1 - VISUAL EVLUATION OF FEEDS

The first step in forage evaluation and determine quality is to visualize for certain criteria.

Exercise, samples of haylage, silage, high moisture grain, and hay are available for physical examination. The samples came from various storage structures (bunker, upright, bag).

Answer the following questions?

Haylages

1. Determine stage of maturity of these hay crop forages?
   Consider leafs or stems of the samples.
   Is the sample mainly grass or alfalfa?
   Do you think the DM content is appropriate for the type of storage structure?
   Is there any evidence of mold?

Haylage	Haylage
Haylage	Haylage

2. Do you consider the length of cut (too fine, just right, or too coarse) for ensuring proper fermentation? For good rumen ecosystem?

Haylage 1	Haylage 2
Haylage 3	Haylage 4

3. Describe the odor of these forages?
    - Strong acid?
    - High ammonia?
    - Vinegar?
    - Alcohol?

Haylage	Haylage
Haylage	Haylage

Corn silages

1. Examine the grain amount present in the silage?
2. Is it a lot, not much, or typical amounts in the silage?
3. Is the grain present hard or soft?
4. Do you think the DM content is appropriate for the type of storage structure?
5. Is there evidence of any mold?
6. Is the length of cut too fine, just right, or too coarse for ensuring proper fermentation? For good rumen ecosystem?

Corn silage 1	Corn silage 2
Corn silage 3	Corn silage 4

7. Is the odor of forages could be described? Strong acid? High ammonia? Vinegar? Alcohol?

Corn silage 1	Corn silage
Corn silage 3	Corn silage

High moisture shelled corn

1. Evaluate the dry matter, odor, and particle size of the two samples.

High moisture corn 1

High moisture corn 2

Dry hay

1. Evaluate the following hay samples for stage of maturity, leafiness, color, and moldiness.

Hay 1

Hay 2

Suggested DM content for ensiling high moisture feedstuffs.

		Range (%)
Grains		
Shelled corn:		
Small		65-
Forages		
Haycrop		
	Oxygen-limiting	45-60
	tower silos Horizontal silos	35-
Corn		
	Oxygen-limiting	40-45
	tower silos Horizontal	

Recommended forage particle sizes using .

Screen	Particle Size (inches)	Corn Silage	Haylage	TMR
Upper Sieve	> 0.75	3 to 8%	10 to 20%	2 to 8%
Middle Sieve	0.31 to 0.75	45 to 65%	45 to 75%	30 to 50%
Lower Sieve	0.07 to 0.31	30 to 40%	20 to 30%	30 to 50%
Bottom Pan	< 0.07	< 5%	< 5%	≤ 20%

Common silage problems.

Visual	Possible
Caramelized dark brown	Generally a dark brown color caused by entrapment of oxygen during filing or air leaks in the silo. This indicates high heat fermentation.
Dark haylage with a cooked smell.	Excessive heat damage favored by high dry matter content, oxygen not eliminated, too long a chop, or poor compaction.
Moldy	Molds grow in presence of oxygen and crops. Slow filling, slow feed out, too long a chop, poor distribution and packing, and air infiltration
Rancid	Caused by clostridial fermentation with production of butyric acid. Favored by low dry matter content, and low plant sugar content.
Vinegar	Fermentation dominated by bacteria, which ferments sugars to acetic acid. Favored by low dry matter content, and low soluble plant sugar content.
Alcohol	Fermentation dominated by yeast, which ferments sugars to alcohol. Favored by slow feed out, air penetration, and limited heat generation.

Adapted from the Pioneer Forage Manual: A Nutritional Guide.

ACTIVITY 2 - pH OF FORAGES

pH can reveal some aspects related to the type of fermentation that has occured and the forage quality. It can also give clues to the wrong issues during the ensiling process if the pH is too high.

The ideal ranges for pH of various crops at different dry matter contents.

Expected range in pH for ensiled crops

Crop	pH range
Corn silage	
25% DM	3.5 - 3.
30% DM	3.8 - 4.
35% DM	3.9 - 4.
Legume silage	
<30% DM	4.6 - 5.
30-35% DM	4.6 - 5.
>35% DM	4.6 - 5.
Grass silage	
<30% DM	4.3 - 4.
30-35% DM	4.3 - 4.
>35% DM	4.4 - 5.

Determine the pH on the following forages:

Haylage 1 - pH_____ Haylage 3 - pH _____

Haylage 2 - pH_____ Haylage 4 - pH _____

Corn silage 1 - pH_____ Corn silage 3 - pH _____

Corn silage 2 - pH_____ Corn silage 4 - pH _____

Have any of the above forages undergone an undesirable fermentation?

ACTIVITY 3 - DETERMINING FORAGE DM

Ensiling forages at the suitable DM content is crucial for a proper fermentation. The perfect forage DM content is mainly dependent on the type of storage structure being used. Observing DM is important prior to ensiling and also when the forage is being fed out.

Listed below are some suggested guidelines for drying times. Times will vary depending on sample size.

Corn silages	<40% DM
Initial drying time	1:30 minutes
Second drying time	0:45 seconds
Third drying time	0:35 seconds
Fourth drying time	0:30 seconds*

Haycrop forages	<40% DM	>40% DM
Initial drying time	1:00 minutes	0:50 seconds
Second drying tim	0:35 seconds	0:40 seconds
Third drying time	0:25 seconds	0:25 seconds
Fourth drying time	0:15 seconds*	0:15 seconds*

*After each drying, observe sample for moisture; sample should get more brittle with each drying time. After the fourth drying time, weigh sample on scale and record weight. Place sample in microwave for another 10 to 20 seconds, weigh sample again. Continue process until the sample weight become stable.

Recording forage dry matter values:

Forage:________________________ Initial weight: ________

Final weight: _________

(Dry matter % = Final weight ÷ Initial weight x 100) Dry matter % ____

— —

Forage:________________________ Initial weight: ________

Final weight: _________

(Dry matter % = Final weight ÷ Initial weight x 100) Dry matter % ____

— —

Forage:________________________ Initial weight: ________

Final weight: _________

(Dry matter % = Final weight ÷ Initial weight x 100) Dry matter % ____

— —

Forage:________________________ Initial weight: ________

Final weight: _________

(Dry matter % = Final weight ÷ Initial weight x 100) Dry matter % ____

— —

Forage:________________________ Initial weight: ________

Final weight: _________

(Dry matter % = Final weight ÷ Initial weight x 100) Dry matter % ____

— —

Forage:________________________ Initial weight: ________

Final weight: _________

We will illustrate the significance of how DM changes in forage can affect the pounds of forage DMI.

A herd was feeding TMR consisting of haylage as forage source and a grain mix as the concentrate source. The ration was 50:50 forage to concentrate ratio on a DM basis. For cow, the program called for 22 Kg of haylage and 14 Kg of grain mix as-fed basis. This herd was part of field trial, where forage DM was determined on a weekly basis. Assume this rancher did not account for DM changes in the haylage over the next three weeks and use the same formula of 22 Kg of haylage as-fed. Compare the differences in haylage dry matter intake.

Week 1

22 Kg. X____.48____% haylage DM as a decimal =_____haylage DM Kg.

Week 2

22 Kg. X____.41____% haylage DM as a decimal =_____haylage DM Kg.

Week 3

22 Kg. X____.36____% haylage DM as a decimal =_____haylage DM Kg.

Week 4

22 Kg. X____.32____% haylage DM as a decimal =_____haylage DM Kg.

1. What problems would you supposed to happen in animal performance with these changes in forage DMI?

---

2. If the recommendation of program were for cows to consume 15 Kg of haylage dry matter, how much haylage would need to be fed on a wet basis during weeks 2 & 4 to cover the required forage DMI?

Task 1……

Task 2…….

Task 3……

Task 4……

Task 5……

Task 6……

Task 7……

Task 8……