

Methods of sampling and analysis

Section 5 - Methods of sampling and analysis

Sampling

Sampling protocols should meet scientifically recognized principles and procedures

Analysis

Laboratory methods developed and validated using scientifically recognized principles and procedures should be used.¹⁵ When selecting methods, consideration should also be given to practicability, with preference given to those methods which are reliable and applicable for routine use. Laboratories conducting routine analyses of feed and feed ingredients should ensure their analytical competency with each method used and maintain appropriate documentation.¹⁶

Source: Code of practice on good animal feeding (CAC/RCP 54-2004).



INTRODUCTION

Important factors that determine the design and implementation of a sampling programme involve shipment size, ingredient variability, laboratory accuracy, cost of the assay and value of the ingredient. Therefore, when defining the sampling procedures one should consider the purpose of sampling, the laboratory analysis through which samples will undergo and the characteristic of the ingredients and finished products.

Sampling protocols should meet scientifically recognized principles and procedures.

Laboratory methods should be developed and validated according to scientifically recognized principles.

Sampling procedures will depend on the nature of the raw material, in process or finished product lots, conveying and sampling equipment. Prior knowledge of the product data and sampling resources allows the assignment of the appropriate sampling procedures.

The use of recognized international sampling methods will ensure a standardized administrative and technical approach and will facilitate the interpretation of results of analysis related to lots or consignments of feed.

GUIDELINES ON SAMPLING

The objectives and sampling purposes to be achieved should be clear when developing the sampling procedures to be adopted. Examples of objective that should be taken into consideration are the following:

- Acceptance of consignments;
- Testing for batch release;
- Control of raw materials;
- Control of in process products;
- Finished products controls;
- Release of non-conforming products;
- Obtaining of retention sample;
- Legal disputes;
- Inter-laboratory trials;
- Validation of analytical methods;
- Validation of control measures.

Sampling should be done in a well defined area in order to avoid difficulties in the executing of procedures, reduce the risk of contamination and cross contamination, enable the proper execution of laboratory analysis and include all necessary safety and health precautions to the sampler and environment.

Personnel responsible for the sampling activities should be trained on the applicable pro-

cedures and have the necessary knowledge of products to be sampled, tools used in the sampling process, adequacy and cleanliness of the environment and sample storage container not to allow contamination or deterioration of the sample.

Sampling process and equipment

For the execution of the sampling procedures proper tools and materials need to be available to allow:

- The opening of bags, packages, barrels, drums, containers, trucks, etc;
- The re-closing of containers;
- The labeling to indicate that a sample has been removed;
- The storing, retaining and preservation of the sample;
- The labeling of the storage and retention container;
- The sampling precautions required by the chemical and microbiological methods of analysis.

All tools and auxiliary materials should be inert, and in a clean condition before and after their use. In the same manner, cleaning of the containers to be sampled is to be considered prior to sampling.

The feed industry uses a combination of tools for collecting samples. Bulk trucks and rail shipments of grains or soybean meal are frequently sampled using a hand probe. Bulk containers may be stratified and multiple samples collected if different portions of the grain are to be sampled.

Slotted grain probes may be used to collect a representative sample from grain, soybean meal or finished feed. The grain probe should be long enough to penetrate at least $\frac{3}{4}$ of the depth of the feedstuff. Official grain samples are collected using a 4.13cm diameter probe that consists of two tubes, one inside the other. The inner tube is divided into compartments that enable the individual collecting the sample to detect inconsistencies in grain quality across the profile of the carrier. This procedure is more labor intensive since the contents of the probe must be emptied onto a tarp or trough and inspected before the grain is transferred into a container. Open handled grain probes, in which the inner-tube is not divided into compartments may be used for sampling feed ingredients including grain. The probe's contents are emptied from the handle and mixing will occur, making it difficult to perform a visual inspection for load inconsistencies by depth. An open handled spiral probe

is designed such that openings on the inside tube rotate around so it opens first at the bottom and then in gradual steps to the top. This assures a fair portion of the sample is collected across the profile of the material.

However, incorrect use of this probe can result in the opposite effect if the inside tube is rotated in the opposite direction, resulting in a disproportionate amount of sample collected from the top. The probe should be inserted into the grain or feed ingredient at a 10-degree angle from the vertical, with the slots facing upward and completely closed. A 10-degree angle is used to obtain a cross section of the material, while placing the end of the probe as close to the bottom of the carrier as possible. The slots must be kept closed until the probe is inserted as far as it will go. If the probe's slots are open as it enters the grain, a disproportionate amount of material from the top will fill the probe. After the probe is fully inserted, the slots should be opened and the probe moved up and down quickly in two motions. The slots are then closed completely, the probe grasped by the outer tube and withdrawn from the grain.

FIGURE 3. Manual Quartering



Source: Compêndio Brasileiro de Alimentação Animal, 2005, SINDIRAÇÕES

The Pelican grain sampler is used for on-line grain sampling. The probe is a leather pouch, approximately 0.46m long, with a band of iron inserted along the edge to hold the pouch open. The pouch is attached to a long pole. Pelicans are designed to catch grain as the pouch is swung or pulled through a falling stream of grain. The Pelican grain sampler is useful for sampling grain, soybean meal or complete feed samples while a truck is unloading.

Bag shipments of base mixes, premixes and medicated feeds should be sampled with a bag probe. Tapered bag triers are used to sample closed bags of powdered and granular commodities. Double-tube bag triers are constructed of stainless steel or chrome plated brass. These triers are available in various lengths and diameters, in both close ended and open ended models and may be used to sample closed and opened bags of powdered and granular ingredients. Single tube, open ended bag triers are constructed of stainless steel tubing and are used to sample opened bags of dry, powdery commodities when removal of a core material is desired.

Fat, molasses and other liquid ingredients stored in drums or barrels can be sampled using a tube of glass or stainless steel. Bulk shipments of liquid ingredients may require a pump sampler. In all cases, the liquid should be subject to stirring prior to the withdrawal of the sample to ensure ingredient distribution.

Forage samples should contain substantial amount of material. The sampling procedure and sample preparation will vary depending on whether the material is a dry forage, silage, pasture, green chopped forage or forage in the field. Sample should be collected in twenty different locations using a core sampler. If this tool is not available, hand sampling can be used. Care should be taken to avoid leaf loss when using this latter procedure.

Collecting silage samples should be performed by removing a column of 0.15m deep by 0.30m wide on the open face. Silage should be mixed, placed in a plastic bag, tightly packed and sealed to exclude the air.

Pasture and field storage sampling is subject to variations in soil fertility and moisture content, therefore should be exercised with care. Eight to ten locations should be selected for sampling, removing approximately 0.1m square of forage at grazing height at each location. The composite of the sub samples should be mixed and material reduced to 1kg of working sample. Samples of

green pasture should be immediately dried to prevent chemical changes.

Water samples may be collected directly into a clean sample container from ponds, lakes, tanks or other sources. The container should be immersed, holding it neck down to 0.30m below the surface. Then, turned mouth up to be filled with sample. Water after extended pumping should be sampled for two to four minutes to ensure it has not been standing in pipes. When bacterial examination is performed, a sterile container should be used for the water sampling.

Finished feed can be sampled as it is transferred to the delivery vehicle if the feed is in the bulk form. In the case of cattle feed that is mixed during transport, collecting the sample from the feed bunk is an acceptable practice.

Any sign of non uniform material, which includes differences in shape, size or color of particles in crystalline, granular or powdered solid substances, moist crust on hygroscopic substances, deposits of solid material or stratification in liquid products should be detected during the sampling procedure. Portions of the material that are non-homogeneous should be sampled separately and should not make a composite as it can mask quality problems.

Sample reduction

Sample reduction may be performed by quartering the sample to a convenient amount for analysis. The mixed composite sample should be spread on clean plastic or paper to form an even layer. The paper is marked into quarters and the two opposite quarters are taken and mixed. The process is repeated until the two quarters selected give the desired sample size. The end result of this process should produce a working sample of 0.5 to 1kg.

Complete feed and feed ingredients may be partitioned into uniform sub samples using a riffler. The sample is poured into the hopper, which is divided into equal portions by two series of chutes that discharge alternately in opposite directions into separate pans.

Heavy plastic bags, zip lock bags, plastic bags or plastic containers make excellent sample containers for dry ingredients or finished feed. The container should protect the sample from light, air, moisture as required by the storage conditions.

Sampling frequency and retention

With few exceptions, all incoming ingredients should be sampled upon arrival and inspected

for identity, physical purity and compared with a reference sample and standard specifications. The sampling procedure should include inspection of the carriers paperwork to ensure the correct material is being delivered and documentation of receipt of the ingredients, which may include a certificate of analysis. When receiving bulk materials, the shipping documents should be inspected for identification of mill, supplier and the name of the individual hauling the cargo. A receiving report that documents receipt of raw materials will augment a sampling programme. This report should include the date, raw material identification, supplier name, carrier name, bill of lading, purchase order, invoice number, time of receiving, weight, bin number where the ingredient was placed, number of the supplier certificate of analysis, sensory and physical properties verified on the receipt of goods and signature of the person responsible for the receiving inspection.

Samples should be retained until the complete feed has been consumed by the animal or as long as liability exists. Commercial feed mills should collect and retain a sample of complete feed for each run of a given product. Medicated feed sampling and evaluation must conform to regulatory requirements.

Sampling plans for raw materials and finished products

International methods of sampling should be used to ensure that valid sampling procedures are applied when feed is being tested for compliance to a particular standard or objective. The Codex General Guidelines on Sampling – CAC/GL 50-2004 (FAO/WHO, 2004) provides information to facilitate the implementation of these goals (Box 21).

Numerous sampling plans are available and none can ensure that every item in a lot conforms to the studied parameters. They are nevertheless useful for guaranteeing an acceptable quality level agreed by the parties for the specified controls.

A sampling procedure should stipulate the conditions based on which a lot should be inspected and classified. These conditions include the inspection procedures (normal, tightened or reduced inspection), switching procedures (normal to tightened, tightened to normal and normal to reduced) inspection level (I, II and III, S-1, S-2, S-3, S-4), Acceptance Quality Levels (AQLs), number of items to be randomly selected from the lot and that will comprise the sample, acceptance and rejection numbers.

BOX 21**Recommendations for the selection of sampling plans**

The following enumerates the essential points that users should address for the selection of appropriate sampling plans

1. Existence (or not) of international reference documents on sampling of the considered products.
2. Nature of the control
 - Characteristic applicable to each individual item of the lot;
 - Characteristic applicable to the whole lot (statistical approach).
3. Nature of characteristic to control
 - Qualitative characteristic (characteristic measured on a pass/failed or similar basis, i.e. presence of pathogen micro-organism);
 - Quantitative characteristic (characteristic measured on a continuous scale, for example a compositional characteristic).
4. Choice of the quality level (AQL or LQ)
 - In accordance with the principles laid down in the Codex Manual of Procedures and with the type of risk: critical/non-critical non-conformities.
5. Nature of the lot
 - Bulk or pre-packed commodities;
 - Size, homogeneity and distribution concerning the characteristic;
 - control.
6. Composition of the sample
 - Sample composed of a single sampling unit;
 - Sample composed of more than one unit (including the composite sample).
7. Choice of the type of sampling plan
 - Acceptance sampling plans for statistical quality control;
 - For the control of the average of the characteristic;
 - For the control of per cent non-conforming items in the lot
 - Definition and enumeration of non-conforming items in the sample (attribute plans)
 - Comparison of the mean value of the items forming the sample with regards to an algebraic formula (variable plans)

Source: The Codex General Guidelines on Sampling – CAC/GL 50-2004 (FAO/WHO, 2004)

A number of ISO standards are available in the case of control situations not dealt with by The Codex General Guidelines on Sampling – CAC/GL 50-2004 (FAO/WHO, 2004). The standards provided are:

ISO 2854:1976: Statistical interpretation of data
- Techniques of estimation and tests relating to means and variances

ISO 2859-1:1999: Sampling procedures for inspection by attributes
- *Part 1: Sampling schemes indexed by acceptance quality limit (AQL) for lot-by-lot inspection*

ISO 2859-2:1985: Sampling procedures for inspection by attributes
- *Part 2: Sampling plans indexed by limiting quality (LQ) for isolated lot inspection*

ISO 2859-3:2005: Sampling procedures for inspection by attributes
- *Part 3: Skip-lot sampling procedures*

ISO 2859-4:2002: Sampling procedures for inspection by attributes
- *Part 4: Procedures for assessment of declared quality levels*

ISO 2859-5:2005: Sampling procedures for inspection by attributes
- *Part 5: System of sequential sampling plans indexed by acceptance quality limit (AQL) for lot-by-lot inspection*

ISO 2859-10:2006: Sampling procedures for inspection by attributes
- *Part 10: Introduction to the ISO 2859 series of standards for sampling for inspection by attributes*

ISO 3494:1976: Statistical interpretation of data
- Power of tests relating to means and variances

ISO 3951-1:2005: Sampling procedures for inspection by variables
- *Part 1: Specification for single sampling plans indexed by acceptance quality limit (AQL) for lot-by-lot inspection for a single quality characteristic and a single AQL*

ISO 3951-2:2006: Sampling procedures for inspection by variables
- *Part 2: General specification for single sampling plans indexed by acceptance quality limit (AQL) for lot-by-lot inspection of independent quality characteristics*

ISO 3951-3:2007: Sampling procedures for inspection by variables
- *Part 3: Double sampling schemes indexed by acceptance quality limit (AQL) for lot-by-lot inspection*

ISO/WD 3951-4: Sampling procedures for inspection by variables

- *Part 4: Procedures for assessment of declared quality levels*

ISO 3951-5:2006: Sampling procedures for inspection by variables

- *Part 5: Sequential sampling plans indexed by acceptance quality limit (AQL) for inspection by variables (known standard deviation)*

ISO 5725-1:1994: Accuracy (trueness and precision) of measurement methods and results

- *Part 1: General principles and definitions*

ISO 7002:1986: Agricultural food products

- Layout for a standard method of sampling from a lot

ISO 8422:2006: Sequential sampling plans for inspection by attributes

ISO 8423:1991: Sequential sampling plans for inspection by variables for percent nonconforming (known standard deviation)

ISO/TR 8550-1:2007: Guidance on the selection and usage of acceptance sampling systems for inspection of discrete items in lots

- *Part 1: Acceptance sampling*

ISO/TR 8550-2:2007: Guidance on the selection and usage of acceptance sampling systems for inspection of discrete items in lots

- *Part 2: Sampling by attributes*

ISO/TR 8550-3:2007: Guidance on the selection and usage of acceptance sampling systems for inspection of discrete items in lots

- *Part 3: Sampling by variables*

ISO 10725:2000: Acceptance sampling plans and procedures for the inspection of bulk materials

ISO 11648-1:2003: Statistical aspects of sampling from bulk materials

- *Part 1: General principles*

ISO 11648-2:2001: Statistical aspects of sampling from bulk materials

- *Part 2: Sampling of particulate materials*

ISO 14560:2004: Acceptance sampling procedures by attributes

- Specified quality levels in nonconforming items per million

BOX 22

Selection of methods

- Methods that have been applied to the matrix of interest should be preferred over methods that have been applied to other matrices or methods that apparently have not been tested on authentic samples.
- Methods documented by published inter-laboratory validation data should be selected over those that are not.
- Methods that have been tested and validated over the concentration range of interest should be chosen over methods tested at other levels. Methods that perform quite well at one level may be totally inadequate at a lower level.
- Methods that are widely used should be chosen over methods not in wide use.
- Methods that are simple, low cost, or rapid should be chosen over methods that are complex, more costly, or slower.
- Preference should be given to methods for which reliability has been established in collaborative studies in several laboratories.
- Preference should be given to methods that have been recommended or adopted by relevant international organizations.
- Preference should be given to methods of analysis that are uniformly applicable to various substrates over those that apply to individual substrates.

Source: Garfield, F.M. Quality Assurance Principles for Analytical Laboratories, 1994.

ANALYSIS

Methods of analysis

Knowledge of feed composition is of utmost importance for determining the nutritional requirements of livestock, to produce balanced compound feeds, to control production process and to manage the final quality of products.

Accuracy, precision, specificity, sensitivity, dependability and practicality should be considered when choosing the most appropriate method. Furthermore, the selection of appropriate methods must take into account matters other than the listed attributes. Depending on their purpose and administrative propriety, methods can be classified in (Garfield, 1994):

- Official methods
- Reference methods
- Screening or rapid methods
- Routine methods
- Automated methods
- Modified methods

Official methods are those required by law or regulation and used in regulatory analyses by a government agency or an industry regulated by a government agency.

Reference methods are developed by organizations or groups that use collaborative studies to validate them.

Screening or rapid methods are used as expedient means to determine, for a large number of samples, whether any of them should be subjected to additional testing by a more accurate method.

Routine methods are used on routine analysis and can be official or standard or even modified to be more convenient when a large number of samples need to be processed.

Automated methods use automated equipment and may be official or screening methods.

Modified methods are usually official or standards methods that were modified for simplification, to remove interfering substances or to be applicable to different types of samples.

Laboratory quality assurance program

One of the main goals of the laboratory is the production of high quality analytical data obtained through analytical measurements that are accurate, reliable and adequate for the intended purpose. This can be achieved with the implementation of a well established quality assurance program ensuring analytical competency and maintenance of proper documentation.

Quality assurance programmes will require the implementation of elements such as: management quality policy statement, program objectives, control of samples and records, equipment maintenance, methods evaluation, measurement principles, training, methods selection, intra- and

inter-laboratory testing, reference standards, field and lab sampling, statistical considerations, audits, corrective actions, program revision and update.

Laboratories operating under a recognized quality standard should seek independent approval of their quality assurance arrangements preferably by accreditation which will allow them to demonstrate competency and reliability. Quality standards are available such as ISO/IEC 17025 – General Requirements for the Competence of Testing and Calibration Equipment and applied by the accreditation organization on the evaluation of the compliance of the laboratory.

References

- FAMI-QS. 2007. EU Guide to Good Practice for Feed Additives and Premixtures Operators, Version 2, 17 January 2007;
- FAO. 2004. Assessing Quality and Safety of Animal Feeds;
- FAO/WHO. 2004. The Codex General Guidelines on Sampling – CAC/GL 50-2004;
- FAO/WHO. 2006. Guidelines for Food Import Control Systems;
- Garfield, F.M. 1994. Quality Assurance Principles for Analytical Laboratories;
- ISO/IEC 17025:2005. General Requirements for the Competence of Testing and Calibration Equipment;
- SINDIRAÇÕES. 2005. Compêndio Brasileiro de Alimentação Animal. Guia de Métodos Analíticos, pages 5 - 9.

TABLE 4. **RECOMMENDATIONS FOR SAMPLING AND ANALYSING**

When defining the sampling procedures one should consider the purpose of sampling, the laboratory analysis through which samples will undergo and the characteristic of the ingredients and finished products.
The objectives and sampling purposes to be achieved should be clear when developing the sampling procedures to be adopted.
Sampling should be done in a well defined area in order to avoid difficulties in the executing of procedures, reduce the risk of contamination and cross contamination, enable the proper execution of laboratory analysis and include all necessary safety and health precautions to the sampler and environment.
Personnel responsible for the sampling activities should be trained on the applicable procedures.
All tools and auxiliary materials should be inert, and in a clean condition before and after their use.
Portions of the material that are non homogeneous should be sampled separately and should not make a composite as it can mask quality problems.
With few exceptions, all incoming ingredients should be sampled upon arrival and inspected for identity, physical purity and compared with a reference sample and standard specifications.
International methods of sampling should be used to ensure that valid sampling procedures are applied when feed is being tested for compliance to a particular standard or objective.
A sampling procedure should stipulate the conditions based on which a lot should be inspected and classified.
Accuracy, precision, specificity, sensitivity, dependability and practicality should be considered when choosing the most appropriate method.
Laboratories operating under a recognized quality standard should seek independent approval of their quality assurance arrangements preferably by accreditation which will allow them to demonstrate competency and reliability.