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CHAPTER 4

SAMPLE PREPARATION AND ANALYSIS

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CHAPTER 4

SAMPLE PREPARATION AND ANALYSIS

4.1 BASIS OF DETERMINATION

NIRT wheat protein is determined on a representative sample portion of approximately 500 grams up to the work portion size of wheat after the removal of dockage using a standard dockage tester. NIRT soybean protein and oil are determined on a representative portion of approximately 500 grams up to the work portion size of cleaned soybeans. NIRT corn protein, oil, and starch are determined on a representative portion of approximately 500 grams up to the work portion size after the removal of broken corn and foreign material. An NIRT instrument equipped with a flow through sample cell is approved equipment for official testing. An NIRT instrument equipped with the sample transport mechanism can be approved (ISE will approve these instruments on a case-by-case basis) for use in analyzing an approximately 80-gram wheat sample and 175-gram samples of corn and soybeans. Contact ISE for more information on the use of the sample transport mechanism.

The official NIRT calibrations were developed to automatically provide protein percentages corrected to a 12 percent moisture basis for wheat, protein and oil in soybeans to a 13 percent moisture basis, and protein, oil, and starch in corn on a dry matter basis. This eliminated the need for mathematical calculation.

Since ISE provides separate calibrations for each wheat class, it is essential that the correct calibration is used. A qualified inspector must review the wheat sample to determine class to ensure that the appropriate calibration is used. Use the calibration for the predominant class when performing an NIRT protein determination on a mixed wheat sample.

As the sample passes through the light path, multiple measurements of the transmitted light takes place. Each time a measurement is taken, the portion of the sample within the light path is considered a subsample for NIRT purposes.

4.2 CLEANING SAMPLES

a. Wheat.

Samples shall be cleaned by a mechanical process (dockage tester) that removes dockage. The dockage tester uses aspiration (air) and a combination of riddles and sieves to remove any readily separable foreign matter. Generally, the foreign matter removed consists of all matter lighter, larger, or smaller than grain. If excessive quantities of wild buckwheat, cob joints, chess and similar types of seeds, and flaxseed are found additional sieving procedures are required. Refer to the Wheat Chapter in Book II of the Grain Inspection Handbook for detailed instructions.

b. Soybeans.

Samples shall be cleaned by hand sieving over an 8/64-inch round hole sieve to remove any fine foreign material (FM). **The portion remaining on top of the sieve shall be handpicked to remove any coarse FM (refer to chapter 1, section 1.3 for the definition of coarse FM).** The portion remaining on top of the sieve after the removal of fine and coarse FM shall be used for the analysis.

c. Corn.

Broken corn and foreign material (BCFM) shall be removed by sieving the sample (machine sieving with a dockage tester or hand sieving) over an 12/64-inch round hole sieve **and handpicking any material (e.g., soybeans, pieces of corn cob) other than corn that remains on top of the sieve.** The portion remaining on top of the sieve after the removal of BCFM shall be used for the analysis.

d. Mechanical/Hand Sieving Procedures.

Use the following procedures to mechanically/hand sieve foreign material (FM in soybeans, BCFM in corn).

(1) For mechanical sieving:

(a) Place the appropriate sieve on top of a bottom pan and position it in the mechanical grain sizer.

(b) Pour the sample portion into the center of the sieve. (Do not sieve more than 500 grams at a time.)

(c) Set the sizer's counter to the appropriate number of strokes (20 for corn, and 5 for soybeans) and then turn it on.

(2) For hand sieving:

(a) Place the appropriate sieve on top of a bottom pan.

(b) Pour the sample portion into the center of the sieve. (Do not sieve more than 500 grams at a time.)

(c) Hold the sieve and bottom pan level and, using a steady motion, move the sieve from right to left approximately 10 inches. Return from left to right to complete one sieving operation. Repeat this operation 20 times for corn, and 5 times for soybeans.

4.3 NIRT ANALYSIS

Operators must read the user's manual and familiarize themselves with the instructions before operating the NIRT instrument.

a. Locations Using the Flow Through Sample Cell.

(1) For wheat protein determinations, use the 18-millimeter cell in the NIRT analyzer. The protein portion shall be a representative amount (dockage-free), weighing between 500 grams and the work portion size. For soybean protein and/or oil determinations, use the 30-millimeter cell in the NIRT analyzer. The analytical portion shall be a representative amount weighing between 500 grams and the work portion size that has been cleaned using the procedures found in section 4.2 of this chapter. For corn protein, oil, and/or starch determinations, use the 30-millimeter cell in the NIRT analyzer. The analytical portion shall be a representative amount (broken corn and foreign material free) weighing between 500 grams and the work portion size. **The operator must ensure that the sample's temperature is between 60°F and 80°F (16°C and 27°C).** If the operator has a reason to suspect that the sample may not be within the acceptable temperature range, use a liquid-in-glass or digital thermometer specified to $\pm$ 2°F ($\pm$ 1°C) accuracy or better to measure the sample temperature. If the sample is not in the acceptable temperature range, allow it to cool or warm to within the limits before testing.

(2) Ensure that the displayed calibration ID is the ISE approved calibration for the type of grain and wheat class being tested and that the SRS for that grain type or wheat class have been tested that day.

(3) Pour the entire sample into the NIRT hopper, enter the sample identification number, and press the "RUN" button. The instrument provides a display (and printout, if a printer is attached) of percent protein for wheat, percent protein and oil for soybeans, and percent oil, protein, and starch for corn.

(4) Retain the sample for further analysis if needed.

b. Locations Using a Sample Transport Mechanism. The sample transport mechanism is needed when samples do not meet the requirement for the flow through sample cell. Contact ISE for the sample transport mechanism procedures.

4.4 OUTLIER INDICATIONS

An outlier indication is a warning that the result may be in error. Outlier indications should rarely occur. If frequent outlier indications are observed, contact ISE.

a. Types of Outliers. There are four possible outlier indicators (A, B, C, and D). The values for each indicator can range in degree from 0 (no outlier) to 5 (very extreme). The greater the value, the less reliable the predicted result. Review the user's manual for more information.

(1) "A" Outlier (Residual). The sample's scan doesn't fit the calibration.

(2) "B" Outlier (Leverage). The sample's scan doesn't fit the calibration and, if added, will have a strong influence.

(3) "C" Outlier (Standard Deviation). The standard deviation (variability) between subsamples is beyond the specified limit.

(4) "D" Outlier (Outside Range Limits). One or more of the subsamples have been predicted above or below the constituent value limits for the calibration.

b. Causes of Outliers. The most likely causes for outliers are:

(1) Wrong calibration selected. Testing a wheat sample with the wrong wheat calibration or the soybean calibration;

(2) Wrong sample cell. Testing a wheat sample using the soybean/corn (30 mm) cell or vice versa;

(3) Extremely dirty samples;

(4) Poorly mixed samples;

(5) Sample packing or settling problems;

(6) Electrical interference;

(7) Measurement of samples not represented in the calibration set; or

(8) Improper instrument warm up.

Some of these conditions will cause intermittent outlier indications (not repeated on reruns) and others will cause consistent outlier indications. In general, an outlier indication is a warning that the result may be in error.

c. What to do when an outlier occurs? When any outlier indication occurs, the operator must perform the following:

(1) Verify that the correct calibration and sample cell was used (18 mm for wheat and 30 mm for soybeans and corn).

(2) Make sure the sample is cleaned per official procedures.

(3) Make sure the room and sample temperature and the room relative humidity are within the specified ranges.

(4) Mix the sample thoroughly before retesting.

(5) Rerun the sample through the Infratec.

(6) If the second run does not show an outlier indication, certify the results of the second run.

(7) If the second run shows an outlier indication, certify on the basis of the average of the two runs.

(8) Never certify a result with a level "5" outlier in the "A" or "B" positions.

NOTE: The NIRT determines both protein and oil in soybeans, and protein, oil, and starch in corn on each run. Use the second run result for the constituent in question only. Use the first run result for the other constituent.

4.5 TESTING INDIVIDUAL AND SUBLOT SAMPLES

a. Individual Sample. Official sample-lot or submitted samples of wheat, corn, or soybeans are tested and certificated for protein, oil, or starch as applicable, in conjunction with official grade determinations or as a separate testing service.

b. Shiplot and Lash Barges. Testing for protein in wheat is based on sublot results in accordance with the cu-sum loading plan. Testing for protein and/or oil in soybeans and for protein, and/or oil, and/or starch in corn can be determined on an average of individual sublot results or on the basis of a composite sample representing the entire lot. Refer to the Grain Inspection Handbook, Book III, for details concerning the cu-sum loading plan.

c. Unit Trains. Testing for protein in wheat is determined on individual or sublot (up to five carriers per sublot) basis in accordance with the cu-sum loading plan. Testing for protein and/or oil in soybeans and for protein, and/or oil, and/or starch in corn can be determined on individual, sublot (up to five carriers per sublot) or composite sample basis. Applicants can request corn protein, oil, and/or starch, and soybean protein and/or oil on a sublot basis while requesting inspection for grade on an individual carrier basis. When articulated railcars are used, each car is tested as a sublot.

4.6 CERTIFYING OFFICIAL RESULTS

a. Moisture Basis. NIRT instruments are programmed to determine official criteria results (i.e., protein, oil, starch) on a moisture basis that is commonly used for trading purposes. The instruments will automatically report wheat protein results on a 12.0 percent moisture basis, soybean protein and oil results on a 13.0 percent moisture basis, and corn protein, oil, and starch results on a dry matter basis.

b. General Procedures. All official criteria results (e.g., protein, oil, starch) shall be recorded on the work record and in the "Remarks" section of the official inspection certificate to the nearest tenth percent on the applicable moisture basis (wheat 12.0 percent, soybeans 13.0 percent, corn - dry matter basis). Upon request, an applicant can request an alternate moisture basis be used in lieu of the standard moisture basis for certifying soybean protein and oil, or corn protein, oil, and starch.

If an applicant requests an alternate moisture basis for a wheat protein result, show both results (12.0 percent basis and the specified moisture basis) on the work record and certificate.

Show the official results on the inspection certificate using an approved statement as shown in Grain Inspection Handbook, Book IV. Upon request of the applicant for service, official criteria results may be stated on letterhead stationery in lieu of an official certificate.

4.7 CONVERTING RESULTS TO AN ALTERNATE MOISTURE BASIS

a. Conversion Formulas.

Examples of alternate moisture basis specifications and formulas for correcting the NIRT results are:

(1) Converting a wheat protein, soybean protein/oil, or corn protein/oil/starch result from the standard moisture basis to an "as is" moisture basis.

$$A = \frac{P \times (100 - M)}{C}$$

Where A = Percent protein/oil/starch on an "as is" moisture basis.
P = NIRT protein/oil/starch result on the standard moisture basis (based on NIRT result rounded to the nearest tenth percent).
M = Official moisture result for the sample/lot (as applicable).
C = 87 for soybeans, 88 for wheat, or 100 for corn.

(2) Converting a corn protein, oil, or starch result from the dry matter basis to another specified moisture basis.

$$A = \frac{P \times (100 - M)}{100}$$

Where A = Percent protein/oil/starch on a specified moisture basis.
P = NIRT protein/oil/starch result on a dry matter basis (based on NIRT result rounded to the nearest tenth percent).
M = Moisture basis specified by applicant.

(3) Converting a soybean protein or oil result from the 13.0 percent moisture basis to another specified moisture basis.

$$A = \frac{P \times (100 - M)}{87}$$

Where A = Percent protein/oil on a specified moisture basis.
P = NIRT protein/oil result on the 13.0 percent moisture basis (based on NIRT result rounded to the nearest tenth percent).
M = Moisture basis specified by applicant.

(4) Converting a soybean protein result to an oil-free and moisture-free basis.

$$B = \frac{P \times 100}{(100 - (O + 13))}$$

Where B = Percent protein on an oil-free and moisture-free basis.
P = NIRT protein result on a 13 percent moisture basis (based on NIRT result rounded to the nearest tenth percent).
O = Percent oil on a 13 percent moisture basis (based on NIRT result rounded to the nearest tenth percent).

(5) Converting a soybean protein result to an oil-free and specified moisture basis.

$$C = \frac{P \times (100 - M)}{100 - (13 + O)}$$

Where C = Percent protein on an oil-free and specified moisture basis.
P = NIRT protein result on a 13 percent moisture basis (based on NIRT result rounded to the nearest tenth percent).
M =Moisture basis specified by the applicant.
O = NIRT oil result on 13 percent moisture basis (based on NIRT result rounded to the nearest tenth percent).

(6) Converting a wheat protein result from the 12.0 percent moisture basis to another specified moisture basis.

$$A = \frac{P \times (100 - M)}{88}$$

Where A = Percent protein on a specified moisture basis.
P = NIRT protein result on the 12.0 percent moisture basis (based on NIRT result rounded to the nearest tenth percent).
M = Moisture basis specified by applicant.

b. Conversion Guidelines for Submitted Samples and Single Lot Inspections.

For submitted samples and single lots, use the official moisture result for the lot if the applicant requests an "as is" moisture basis.

c. Conversion Guidelines for Sublot Testing.

(1) When sublot testing is performed on unit trains, lash barges, or ships inspected as single lots, and the applicant **does not** specify limits for protein, oil, or starch content on an alternate moisture basis, record individual sublot results and calculate CuSum values on the basis of the standard moisture basis. Upon completion of loading, convert the final average protein/oil/starch result to the specified moisture basis. For an "as is" moisture basis, use the official average moisture result in the conversion formula.

(2) If a load order specifies limits on the protein, oil, or starch content on a specified moisture basis, convert the individual sublot protein/oil/starch results to the desired moisture basis. For an "as is" moisture basis, use the official sublot moisture result in the conversion formula.

Using two separate columns on the loading log, record the individual sublot results (from the standard moisture basis and specified moisture basis) and apply the CuSum loading plan information (e.g., breakpoints, starting values) to the specified moisture basis results column.

Upon completion of the lot, average the protein results in the standard moisture basis column and convert this value to the applicable protein/oil/starch value on the specified moisture basis. For an "as is" moisture basis, use the official average moisture result in the conversion formula.

Enter the converted protein/oil/starch value as the final "average" under the specified moisture basis column (protein/oil/starch results) on the log. **Do not average results from the specified moisture column to obtain a "final" specified moisture basis result.**