

2.3.1 Toluene Determination of Moisture

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References:

Moisture in Animal Feed by Distillation with Toluene (925.04) Official Methods of Analysis. 1990. Association of Official Analytical Chemists. 15th Edition.

Dewar, W.A. and P. McDonald. 1961. Determination of dry matter in silage by distillation with toluene. J. Sci. Agric. 12:790-795.

Fenner, Heinrich. 1984. Methods for determining fermentation products in acid preserved feeds and forages. Massachusetts Agricultural Experiment Station. Res. Bull. 691. 14pp.

Scope:

This procedure is recommended for determination of moisture in fermented feeds (haylages and silages) which contain high levels of volatile acids. Volatile acids and alcohols have low vaporization points and are lost when the sample is oven dried.

Basic Principle:

Water is distilled from sample and trapped under a layer of toluene.

Equipment:

250 mL pyrex flask Bidwell-Sterling Moisture Receiver, calibrated to 0.01 mL by distilling known amounts of water into graduated column and estimating column of water to 0.01 mL. Clean tube and condenser with chromic acid cleaning mixture, rinse thoroughly with water, then alcohol, and dry to prevent undue amount of water from adhering to inner surface.

- 500 mm Liebig condenser
- Toluene distillation hood

Reagents:

Toluene, reagent grade

Personal Protective Equipment:

- Lab coat
- Safety glasses/goggles
- Designated toluene specific gloves

Safety Precautions:

- Use standard precautions when working with electrical equipment or glassware.
- Toluene is flammable. Observe proper precautions for flammable solvents.
 - Avoid inhaling vapors.
 - Avoid skin contact.

Procedure:

1. Weigh sufficient wet sample (at least 25g) to yield a minimum of 5 mL water, record weight (W1) to nearest 1 mg, and transfer to 500 mL flask.
2. Add sufficient toluene to cover sample completely.
3. Immediately fill receiving tube with toluene, pouring it through top of condenser.
4. Bring to boil and distill slowly, ca. 2 drops/sec, until most of the water passes over, then increase rate of distillation to about 4 drops/sec. Distill 1hr (or longer if necessary for wetter samples) to obtain clearing at the top of the condenser.
5. When all water has apparently been distilled, wash down condenser by pouring toluene in at top and continue distillation for a short time (approximately 15 min) to ensure all water is distilled. If 0.1 mL of additional water is distilled in 15 minutes, repeat this step.
6. If water remains in the condenser after distillation is completed, wash it down with toluene.
7. Let receiving tube come to room temperature and read volume of water in lower layer of receiver and record volume (V) to nearest 0.01 mL.

Comments:

- To be accurate, distilled water should be analyzed for volatile acids and alcohol that can co-distill with water (see references).
- Toluene dry matter is approximately equivalent to partial drying (section [2.2.1.1](#) or [2.2.1.2](#)) which leaves 3 to 5% moisture in the sample (which is roughly equivalent to volatile acids lost during drying) and is adequate for dry matter adjustment of crude protein and fiber.
- Refer to EHS for proper disposal of Toluene

Calculation: Percent Total Dry Matter

$$\% \text{ Total DM} = 1 - \frac{V}{W1} \times 100$$

- Where V = Volume of water in mL
- W1 = weight of sample in grams

Calculation: Percent Total Moisture:

$$\% \text{ Total Moisture} = 100 - \% \text{ Total DM}$$

Quality Control:

Include at least one set of duplicates in each run if single determinations are being made.

An acceptable average standard deviation among replicated analyses for moisture or dry matter is about \diamond 0.10, which results in a warning limit (2s) of about \diamond 0.20 and a control limit (3s) of

about ± 0.30 . Plot the results of the duplicate analyses on an R-control chart (Appendix D) and examine the chart for trends. Results outside the 95 percent confidence limits warn of possible problems with the analytical system. Results outside the 99 percent confidence limits indicate loss of control, and results of the run should be discarded. If more than five or six points in succession fall on one side or the other of the 50 percent line, it is a strong indication that something has changed and is cause for investigation.