

STARCH

PRINCIPLE

Starch is solubilized by boiling the sample in aqueous calcium chloride. Interfering substances are removed by preliminary extraction with aqueous alcohol, and treatment of the calcium chloride extract with a suitable precipitant. Optical activity of the extract is measured with a polarimeter, and starch content is calculated therefrom.

SCOPE

The method is applicable to whole corn or to any of its component parts containing appreciable quantities of starch. With minor modification, the method is applicable to other grains and grain products. Standard deviation is 0.5% absolute.

SPECIAL APPARATUS

Polarimeter: Use a sensitive polarimeter or saccharimeter capable of reproducing within ± 0.01 circular degree, together with 1 dm and 2 dm observation tubes, and a source of monochromatic light such as a sodium vapor lamp or equivalent.

REAGENTS

1. Alcohol Solvent: Dissolve 1 g of mercuric chloride (HgCl_2) in 900 mL of purified water, add 100 mL of 95% ethyl alcohol, and mix thoroughly.
2. Calcium Chloride Solution: Dissolve 550 g of reagent grade calcium chloride dihydrate ($\text{CaCl}_2 \cdot 2\text{H}_2\text{O}$) in 760 mL of purified water and dilute with water to a specific gravity of 1.30 at 60 °F (33° Baumé at 60 °F). Adjust pH to 2.0 (± 0.1) by addition of glacial acetic acid.
3. Stannic Chloride-Phosphotungstic Acid Solution: Dissolve 2.5 g of stannic chloride ($\text{SnCl}_4 \cdot 5\text{H}_2\text{O}$) and 2.5 g of phosphotungstic acid ($\text{H}_3\text{PO}_4 \cdot 12\text{WO}_3 \cdot 14\text{H}_2\text{O}$) in 80 mL of purified water. Add 20 mL of glacial acetic acid, 100 mL of calcium chloride solution (sp g 1.30 at 60 °F), and mix (Note 1).

STARCH — continued**PROCEDURE**

Grind about 50 g of sample through a laboratory cutting mill to 20 mesh or finer and mix thoroughly (Notes 2 and 3). Determine moisture content of ground sample by the Standard toluene distillation method (Note 4) or alternate procedure giving equivalent results.

Weigh accurately about 2 g of sample and transfer quantitatively to a test tube. Add 10 mL of alcohol solvent, stopper tube and shake vigorously for 2 minutes. Filter with vacuum through a 9 cm hard paper supported on a filter cone in a 60 mm funnel. Rinse tube (transferring residue quantitatively) and wash residue with about 25 mL of alcohol solvent. Apply vacuum until residue is substantially dry (Note 5).

Transfer filter paper and residue to a 250 mL beaker, add 10 mL of purified water and macerate with a stirring rod. Add 60 mL of calcium chloride solution and mark liquid level on beaker. Place on a hot plate and bring to boil in about 5 minutes while stirring. Continue boiling vigorously for 30 minutes, stirring occasionally, and adding purified water as needed to maintain liquid level substantially constant. Cool to room temperature in a water bath.

Add 10 mL of stannic chloride-phosphotungstic acid solution to a 100 mL Kohlrausch flask. Transfer contents of the beaker quantitatively to the flask, rinse beaker and dilute to volume with calcium chloride solution (Note 6). Mix thoroughly and let stand about 5 minutes. Gravity filter extract through a folded 18.5 cm retentive paper into a dry flask, discarding the first portion of filtrate (Note 7).

Rinse and fill a clean 2 dm observation tube with the filtrate and determine the optical rotation with a polarimeter (Note 8).

Check zero point of polarimeter. Conduct a blank determination on new reagents or a new tube, and correct the sample rotation accordingly if necessary (Note 9).

STARCH — continued**CALCULATION**

% Starch (dry basis) =

$$= \frac{\text{Degrees Angular Rotation } 100 \times 100 \times 100}{2 \text{ dm} \times 203 \text{ Sample Wt. (g)} \times (100 - \text{Sample Moisture, \%})}$$

Where: 203 = specific rotation of corn starch

NOTES AND PRECAUTIONS

1. The stannic chloride-phosphotungstic acid mixture serves as a coagulating agent for soluble proteins. Alternatively, a 5% solution of uranyl acetate or a 2.5% solution of stannic chloride (prepared with purified water, acetic acid and calcium chloride) have been used successfully.
2. If moisture content is above about 18%, it is advisable to predry the sample prior to grinding. Place sample to be ground in an open dish (but protected from dust or other contamination) in a warm well-ventilated place so that the grain will dry reasonably fast and reach an approximate air-dried condition in from 14 to 24 hours. Moisture loss need not be recorded since moisture content of the ground sample will be determined.
3. If the corn sample is a high-amylose specimen or another type (e.g., flinty) from which starch is extracted with difficulty, grind sample to 60 mesh or finer prior to analysis. Mills suggested for this purpose are the Wiley Laboratory Mill, MIKRO-Samplmill, and the Spex Mixer/Mil
4. When moisture content of the ground sample is determined by the Standard toluene distillation method, the grinding loss is disregarded. The moisture calculation simplifies to:

$$\% \text{ Moisture} = \frac{(\text{mL H}_2\text{O in Trap} + \text{Blank}) \times 100}{\text{Sample Wt. (g)}}$$

5. The alcohol solvent removes optically active solubles while the mercuric chloride prevents enzyme action on the starch.

STARCH — continued

6. When analyzing samples that filter slowly or which tend to give cloudy filtrates, add 2.0 g of CELITE analytical filter aid after diluting to volume with calcium chloride solution. Mix thoroughly and let stand about 5 minutes prior to filtration. Incorporation of filter aid speeds filtration, yields filtrates with higher clarity, and does not affect measured starch contents.
7. Filtered starch extracts can be stored for future analysis when precautions are taken to prevent concentration change.
8. Hazy filtrates can sometimes be avoided by lowering the pH of the calcium chloride extracting solution by 0.2 unit with glacial acetic acid. If this technique and the use of filter aid does not provide adequate clarity for polarimeter readings, use a 1 dm observation tube and multiply the reading by 2.
9. 1° angular rotation = 2.888° Ventzke

REFERENCES

1. Earle and Milner, *Cereal Chemistry*, Vol. XXI, No. 6, Nov. 1944, pp 567-575
2. Greenberg and Shipe, *Journal of Food Science*, Vol. 44, 1979, pp 735-737