112588 Cellulose Content Assay Kit - Micro Method

Product Introduction

Cellulose is a macromolecular polysaccharide composed of glucose and is usually associated with hemicellulose, pectin, and lignin. It is the main structural component of plant cell walls, an important dietary fiber, and the most abundant polysaccharide in nature.

Cellulose consists of β-glucose residues. Under acidic heating conditions, it decomposes to β-glucose. In strong acid, β-glucose is dehydrated to form β-furfural compounds, which then undergo dehydration-condensation with anthrone to produce furfural derivatives. The color intensity is used for indirect quantitative determination of cellulose content.

Package Contents

Code	Item	Pack Size	Storage
112588.1	Reagent I	1 bottle	2-8°C
112588.2	Reagent II	1 bottle	2-8°C, protect from light
112588.3	Reagent III	1 bottle	2-8°C
112588.m	Instructions	1 copy	-

Package specification: 100T

Quality Standards and Safety Information

Raw Material and Packaging Name	Quality Standard	Main Toxicity
Reagent I	—	—
Reagent II	—	—
Reagent III	—	—

Transportation and Storage

Transportation: Transport with ice packs.

Storage: Store as instructed above. Valid for 180 days.

Instructions for Use

1. Sample Preparation

1. Dry the sample at 80°C to constant weight, grind, and pass through a 40-mesh sieve.

   Weigh 0.01 g of sample, add 1 mL of 80% ethanol, homogenize rapidly at room temperature, and heat in a 90°C water bath for 20 min. During heating, the tube may burst open; seal with tape or use an explosion-proof tube.

   Cool to room temperature, centrifuge at 6000g and 25°C for 10 min, and discard the supernatant. Wash the precipitate once each with 1.5 mL of 80% ethanol and acetone. For each wash, vortex for about 2 min, centrifuge at 6000g and 25°C for 10 min, and discard the supernatant. The remaining precipitate is the crude cell wall.

2. Add 1 mL of Reagent I and mix thoroughly. Heat in a 90°C water bath for 30 min. During heating, the tube may burst open; seal

with tape or use an explosion-proof tube.

Cool, centrifuge at 8000g and 25°C for 10 min, and discard the supernatant. Wash the precipitate 3 times with distilled water. For each wash, add 1 mL distilled water, mix well, vortex for about 2 min, centrifuge at 8000g and 25°C for 10 min, and discard the supernatant.

Add 1 mL acetone to the precipitate, mix, centrifuge at 8000g and 25°C for 10 min, discard the supernatant, and dry the precipitate for later use.

3. Add 0.5 mL distilled water to the dried precipitate. If the precipitate does not dissolve completely, use appropriate homogenization or ultrasonication to assist dissolution.

   Place in an ice-water bath, slowly add 0.75 mL concentrated sulfuric acid, mix well, and let stand in the ice-water bath for 30 min.

   Centrifuge at 8000g and 4°C for 10 min. Collect the supernatant and dilute it 20 times with distilled water before testing.

   For samples with low cellulose content, such as rice flour, chestnut, or sweet potato, dilution may not be needed or the dilution factor may be reduced.

2. Assay Procedure

1. Preheat the spectrophotometer or microplate reader for more than 30 min. Set the wavelength to 620 nm and zero with distilled water. Set the water bath to 95°C.
2. Add 4 mL of Reagent III to Reagent II and dissolve thoroughly to prepare the working solution. If dissolution is difficult, use heating and stirring. Unused working solution may be stored at 4°C for one week.
3. Add reagents according to the table below.

Reagent / μL	Blank Tube	Assay Tube
Sample	-	150
Distilled water	150	-
Working solution	35	35
Concentrated sulfuric acid	315	315

Mix well and heat in a 95°C water bath for 10 min. Tighten the cap securely to prevent water loss. After cooling to room temperature, transfer 200 μL to a microplate and measure the absorbance of the blank tube and assay tube separately at 620 nm.

Notes:

- Only one blank tube needs to be measured.
- Concentrated sulfuric acid is highly corrosive. Handle with caution.

3. Cellulose Content Calculation

a) Micro quartz cuvette

Cellulose content (mg/g, dry weight) = [((ΔA + 0.0043) ÷ 7.875 × V1) ÷ (W × V1 ÷ V2)] × 20 = 3.17 × (ΔA + 0.0043) ÷ W

b) 96-well plate

Cellulose content (mg/g, dry weight) = [((ΔA + 0.0043) ÷ 5.25 × V1) ÷ (W × V1 ÷ V2)] × 20* = 4.76 × (ΔA + 0.0043) ÷ W

ΔA = Ameasured - Ablank

V1 = added sample volume, 0.15 mL

V2 = added extraction solution volume, 1.25 mL

W = sample dry weight, 0.01 g

20* = sample dilution factor

Precautions

- The minimum detection limit is 1 mg/g dry weight or 10 ng/mg prot.

Visual Reference