Starch Content Assay Kit

Note: Take two or three different samples for prediction before test.

Operation Equipment: Spectrophotometer/microplate reader

Catalog Number: BC0705

Size: 100T/96S

Product Composition: Before use, please carefully check whether the volume of the reagent is consistent with the volume in the bottle. If you have any questions, please contact Solarbio staff in time.

Reagent Name	Size	Preservation Condition
Reagent I	Liquid 65 mL×1	2-8℃
Reagent II	Liquid 65 mL×1	2-8℃
Reagent III	Powder ×2	2-8℃
Standard	Powder ×1	2-8℃

Solution Preparation:

1. Standard: Before use, add 1 mL distilled water to fully dissolve and prepare 10 mg/mL glucose standard solution for use; store the inexhaustible reagent at 2-8°C for 2 weeks.

2. Preparation of working liquid: Before use, take a bottle of Reagent III and add 2.625 mL distilled water, slowly add 14.875 mL concentrated sulfuric acid, stir continuously, fully dissolve, and wait for use, the reagent can be stored at 2-8℃ for 1 week.

Product Description:

Starch is the main storage form of sugar in plants. The determination of starch content has great significance in evaluating the nutritional value of food and researching the sugar metabolism in plants.

Separate soluble sugar from starch by 80% ethanol, then decompose starch into glucose by acid hydrolysis. The starch content can be calculated by measuring the glucose content using an anthrone colorimetric method.

Technical Indicators:

Minimum Detection limit: 0.0074 mg/mL

Linear Range: 0.008-0.7 mg/mL

Note: Before the experiment, it is recommended to select 2-3 sample with large expected differences for pre-experiment. If the absorption value of the sample is not within the measurement range, it is recommended to dilute or increase the sample size for detection.

Reagents and Equipment Required but Not Provided:

Visible spectrophotometer/microplate reader, water-bath, adjustable pipette, micro glass cuvette/96 well flat-bottom plate, mortar/homogenizer, ice, concentrated sulfuric acid (>95%, AR) and distilled water.

Operation procedure:

I. Sample preparation:(The sample size to be tested can be adjusted appropriately, and the specific proportion can be referred to the literature.)

1. Take 30mg of sample, grind in mortar and add 0.6 mL of Reagent I. After homogenization, transfer to a centrifuge tube, place in a water bath at 80℃ for 30 minutes, then centrifuge at room temperature and 3000 ×g for 5 minutes, discard the supernatant, retain precipitation.
2. Add 0.3 mL of distilled water to the precipitate. Place in boiling water bath for 15 minutes (Wrap the sealing film to prevent bursting).
3. After cooling, add 0.6 mL of Reagent II, place in boiling water bath for 15 min (Wrap the sealing film to prevent bursting), shake 3~5 times.
4. After cooling, centrifuge at room temperature and 8000 ×g for 15 minutes, take the supernatant for test. If there is still turbidity after centrifugation, the centrifugation can be repeated and the supernatant can be taken.

II. Determination procedure:

1. Preheat the spectrophotometer/microplate reader for 30 minutes, adjust wavelength to 620 nm, set zero with distilled water.
2. Adjust the water-bath to 95°C. Wrap the sealing film to prevent bursting.
3. Standard working solution: dilute the 10 mg/mL standard solution with distilled water to 0.4, 0.2, 0.1, 0.05, 0.04, 0.03, 0.02 mg/mL.
4. Standard test: Take 50 μL of Standard solution or 50 μL of distilled water (Blank Tube) and 250 μL of Working solution to EP tube. Place in 95℃ water bath for 10 minutes (tighten the lid to prevent water loss), natural cool to room temperature, take 200 μL to micro glass cuvette/96 well plate, determine absorbance of Standard solution (A_S) and Blank control (A_B) at 620 nm. Calculate $\Delta A=A_S- A_B$. Blank tube and standard curve only need to be test one or two times.
5. Sample test: Take 50 μL of sample and 250 μL of Working solution to EP tube. Place in 95℃ water bath for 10 minutes (tighten the lid to prevent water loss), naturally cool to room temperature, take 200 μL to micro glass cuvette/96 well flat-bottom plate, determine absorbance of Test tube (A_T) at 620 nm. Calculate $\Delta A' = A_T - A_B$.

III. Calculations:

1. Create standard curve

Take glucose standard solution (0.4, 0.2, 0.1, 0.05, 0.04, 0.03, 0.02 mg/mL) as x-axis, ΔA as y-axis, to draw the standard curve and obtain the linear regression equation y=kx+b, substituting ΔA′ into the equation yields x(mg/mL).

2. Calculation of starch content

Calculated by the mass of sample

Starch content (mg/g mass) = $x\times F\times V_E\div W\div 1.11=0.811x\div W\times F$

V_E: volume after extraction, 0.9 mL;

W: sample mass, g;

F: dilution ratio;

1.11:It is the constant of converting glucose content measured by this method into starch content, that is, 111 μg glucose is colored by anthrone reagent, which is equivalent to 100 μg starch by anthrone reagent.

Note:

1. As the working fluid is highly corrosive, please operate with caution.
2. If the absorbance value exceeds the linear range, the sample size can be increased or diluted before determination.

Experimental example:

1. Take 0.03g of corn for sample treatment, take the superneant, dilute it with distilled water 128 times, and then follow the measurement procedure. Use 96 well plate to measure and calculate $\Delta A' = A_T - A_B = 0.728 - 0.106 = 0.622$, standard curve y = 3.1829x + 0.002, calculate x = 0.195.

Starch content (mg/g mass)=0.811x÷W×F =0.811×0.195÷0.03×128 = 674.8mg /g mass.

Recent Products Citations:

[1] Wu HM, Xie DJ, Jia PF, Tang ZS, Shi DQ, Shui GH, Wang GD, Yang WC. Homeostasis of flavonoids and triterpenoids most likely modulates starch metabolism for pollen tube penetration in rice. Plant Biotechnol J. 2023 Sep;21(9):1757-1772. doi: 10.1111/pbi.14073. Epub 2023 May 23. PMID: 37221659; PMCID: PMC10440988.

[2] Sun C, Wang Y, Yang X, Tang L, Wan C, Liu J, Chen C, Zhang H, He C, Liu C, Wang Q, Zhang K, Zhang W, Yang B, Li S, Zhu J, Sun Y, Li W, Zhou Y, Wang P, Deng X. MATE transporter GFD1 cooperates with sugar transporters, mediates carbohydrate partitioning and controls grain-filling duration, grain size and number in rice. Plant Biotechnol J. 2023 Mar;21(3):621-634. doi: 10.1111/pbi.13976. Epub 2022 Dec 29. PMID: 36495424; PMCID: PMC9946139.

[3] Wang K, Cai S, Xing Q, Qi Z, Fotopoulos V, Yu J, Zhou J. Melatonin delays dark-induced leaf senescence by inducing miR171b expression in tomato. J Pineal Res. 2022 Apr;72(3):e12792. doi: 10.1111/jpi.12792. PMID: 35174545.

[4] Li M, Li H, Zhu Q, Liu D, Li Z, Chen H, Luo J, Gong P, Ismail AM, Zhang Z. Knockout of the sugar transporter OsSTP15 enhances grain yield by improving tiller number due to increased sugar content in the shoot base of rice (Oryza sativa L.). New Phytol. 2024 Feb;241(3):1250-1265. doi: 10.1111/nph.19411. Epub 2023 Nov 27. PMID: 38009305.

[5] Lv P, Liu J, Wang Q, Zhang D, Duan X, Sun H. Influence of accelerating storage of foxtail millet on the edible and cooking quality of its porridge: An insight into the structural alteration of the in-situ protein and starch and physicochemical properties. Int J Biol Macromol. 2023 Jun 15;240:124375. doi: 10.1016/j.ijbiomac.2023.124375. Epub 2023 Apr 6. PMID: 37028630.

References:

[1] Clegg K M. The application of the anthrone reagent to the estimation of starch in cereals[J]. Journal of the Science of Food and Agriculture, 1956, 7(1): 40-44.

[2] Viles Jr F J, Silverman L. Determination of starch and cellulose with anthrone [J]. Analytical Chemistry, 1949, 21(8): 950-953.

Related Products:

BC0610/BC0615	Soil β-glucosidase (β- GC) Activity Assay Kit
BC2040/BC2045	β-amylase Activity Assay Kit
BC1850/BC1855	Soluble Starch Synthase(SSS) Activity Assay Kit