Sucrose Phosphoric Acid Synthetase (SPS) Activity Assay Kit

Detection instrument: Spectrophotometer/microplate reader

Catalog Number: BC0605

Size: 100T/48S

Components:

Extract Solution: 50 mL ×1. Storage at 4℃.

Reagent I: 2.5 mL×1. Storage at -20℃.

Reagent II: powder 10 mg×1. Storage at 4℃. Add 1 mL of distilled water to form 10 mg/mL sucrose solution. Dilute to 500 μg/mL with distilled water when the solution will be used.

Reagent III: 2 mL ×1. Storage at 4℃.

Reagent IV: 25 mL×1. Storage at 4℃.

Reagent V: 10 mL×1. Storage at 4℃.

Product Description

Sucrose is not only an important photosynthetic product, but also a major transport material in plants. Moreover, it is one of the storage forms of carbohydrates. Sucrose phosphate synthase (SPS) takes fructose-6-phosphate as the receptor, the sucrose produced by the reaction forms sucrose phosphate under the action of sucrose phosphatase. Sucrose phosphate synthase-sucrose phosphatase system is generally regarded as the main route of sucrose synthesis.

Sucrose phosphate synthase catalyzes fructose-6-phosphate to form sucrose phosphoric acid. The reaction between sucrose and resorcinol can present color change, which has a characteristic absorption peak at 480 nm and the enzyme activity is proportional to the depth of color.

Reagents and Equipment Required but Not Provided

Spectrophotometer/microplate reader, water-bath, centrifuge, adjustable pipette, micro glass cuvette/96 well plate, mortar/homogenizer and ice.

Procedure

I. Sample Extraction:

The tissue mass (g): Extract solution volume (mL) is 1:5-10 (We recommend weigh about 0.1 g of tissue and add 1 mL of Extract solution). conduct ice bath homogenate. Centrifuge at 8000×g for 10 minutes at 4℃, take the supernatant and placed on the ice for test.

II. **Determination procedure:**

1. Preheat the spectrophotometer 30 minutes, adjust the wavelength to 480 nm and set zero with distilled water

2. Add reagents into 1.5 mL centrifuge tube with the following list:

Reagent Name (μL)	Test tube (T)	Control tube (C)	Standard tube (S)	Blank tube (B)
Sample	10	10	-	-

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Distilled water	-	45	45	45
Reagent I	45	-	-	-
Reagent II	-	-	10	-
Blending, water bath for 10 minutes at 25℃.				
Reagent III	15	15	15	15
Boil in boiling water bath for about 10 minutes (entangling sealing film to prevent explosion cover) and cool.				
Reagent IV	210	210	210	210
Reagent V	60	60	60	60

Mix thoroughly, react in water bath for 20 minutes at 80℃(entangling sealing film to prevent explosion cover). After cooling, measure the absorption value of each tube at 480 nm. Calculate $\Delta A_T = A_T - A_C$, $\Delta A_S = A_S - A_B$.

Note: (1) as far as possible within 30 minutes to complete the determination;

(2) Blank tube and standard tube only need to determine 1-2 times

III. Calculation of SPS Activity Unit

1. Calculate by the concentration of protein

Unit definition: One unit of enzyme activity is defined as the amount of enzyme catalyzes the production of 1 μg of sucrose per minute every milligram of tissue protein.

SPS activity(μg/min/mg prot)=$(C_S \times V1 \times \Delta A_T \div \Delta A_S) \div (V1 \times Cpr) \div T = 50 \times \Delta A_T \div \Delta A_S \div Cpr$

2. Calculate by the sample fresh weight

Unit definition: One unit of enzyme activity is defined as the amount of enzyme catalyzes the production of 1 μg of sucrose per minute every gram of tissue.

SPS activity (μg/min/g fresh weight)=$(C_S \times V1 \times \Delta A_T \div \Delta A_S) \div (W \times V1 \div V2) \div T = 50 \times \Delta A_T \div \Delta A_S \div W$

C_S: Concentration of standard tube, 500 μg /mL;

V1: Add the sample volume into the reaction system, 0.01 mL;

V2: Add the extraction liquid volume, 1 mL;

Cpr: Concentration of sample protein, mg/mL;

W: Sample fresh weight, g;

T: Reaction time, 10 minutes.

References:

[1] Schrader S, Sauter J J. Seasonal changes of sucrose-phosphate synthase and sucrose synthase activities in poplar wood (Populus× canadensis Moench ‘robusta’) and their possible role in carbohydrate metabolism[J]. Journal of Plant Physiology, 2002, 159(8): 833-843.

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[1] Han W, Wang Y, Li H, Diao S, Suo Y, Li T, Sun P, Li F, Fu J. Transcriptome and Metabolome Reveal Distinct Sugar Accumulation Pattern between PCNA and PCA Mature Persimmon Fruit. Int J Mol Sci. 2023 May 11;24(10):8599. doi: 10.3390/ijms24108599. PMID: 37239943; PMCID: PMC10217969.

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[3] Shi Y, Zhao Y, Yao Q, Liu F, Li X, Jin X, Zhang Y, Ahammed GJ. Comparative Physiological and Transcriptomic Analyses Reveal Mechanisms of Exogenous Spermidine-Induced Tolerance to Low-Iron Stress in Solanum lycopersicum L. Antioxidants (Basel). 2022 Jun 27;11(7):1260. doi: 10.3390/antiox11071260. PMID: 35883751; PMCID: PMC9312307.

[4] Wu J, Chen H, Chen W, Zhong Q, Zhang M, Chen W. Effect of ultrasonic treatment on the activity of sugar metabolism relative enzymes and quality of coconut water. Ultrason Sonochem. 2021 Nov;79:105780. doi: 10.1016/j.ultsonch.2021.105780. Epub 2021 Oct 6. PMID: 34628309; PMCID: PMC8501503.

[5] Kang L, Wu Y, Zhang J, An Q, Zhou C, Li D, Pan C. Nano-selenium enhances the antioxidant capacity, organic acids and cucurbitacin B in melon (Cucumis melo L.) plants. Ecotoxicol Environ Saf. 2022 Aug;241:113777. doi: 10.1016/j.ecoenv.2022.113777. Epub 2022 Jun 20. PMID: 35738099.

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[1] Schrader S, Sauter J J. Seasonal changes of sucrose-phosphate synthase and sucrose synthase activities in poplar wood (Populus× canadensis Moench ‘robusta’) and their possible role in carbohydrate metabolism[J]. Journal of Plant Physiology, 2002, 159(8): 833-843.

[2] Steven C Huber. Interspecific Variation in Activity and Regulation of Leaf Sucrose Phosphate Synthetase [J]. Zeitschrift für Pflanzenphysiologie, 1981, 102(5): 443-450.

Related Products:

BC0580/BC0585	Sucrose Synthetase(SS) Activity Assay Kit
BC2460/BC2465	Plant Sucrose Content Assay Kit
BC0560/BC0565	Acid Invertase(AI) Activity Assay Kit
BC0570/BC0575	Neutral Invertase (NI) Activity Assay Kit
BC4310/BC4315	Sucrose Synthetase (SS, Cleavage Direction) Activity Assay Kit
BC4320/BC4325	Solid-Acid Invertase (B-AI) Activity Assay Kit

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