

DAB Staining Solution (2.0 mg/mL, pH3.8, for plant)

Cat. No.: D1509203 | Pack size: 100 mL | Storage: 2-8°C, protected from light

Overview

Under environmental stress conditions, plant tissues generate various reactive oxygen species (ROS). ROS are highly active and unstable, so their detection is often based on their end products. Hydrogen peroxide (H_2O_2) is one type of ROS. Catalyzed by peroxidase, hydrogen peroxide can rapidly react with DAB (3,3-diaminobenzidine tetrahydrochloride) to produce a brown-red precipitate, thereby localizing hydrogen peroxide in the tissue.

Aladdin DAB Staining Solution (2 mg/mL, pH 3.8) is composed of DAB and phosphate buffer, used for hydrogen peroxide staining in living plant tissues. It is generally applied for whole-mount staining of tender tissues like root tips and leaves. After staining, areas with hydrogen peroxide accumulation appear brown to dark brown. This product is for research use only, not for clinical diagnosis or other purposes.

Component

D1509203	Component	100 mL	Storage
D1509203A	DAB	200 mg	2-8°C. Store in the dark.
D1509203B	Phosphate Buffer(pH 3.8)	100 mL	2-8°C.

Reagents, consumables and Equipment not provided

- Fresh plant leaves or roots, tap water, distilled water, 95% ethanol.
- Ultrasonic cleaner, magnetic stirrer, electronic balance, measuring cylinder, filter paper, camera.
- DAB staining sample preservation solution (optional).

Procedure (For Reference Only)

1. Reagent Preparation: Fully dissolve 200 mg of Component D1509203A (DAB) into 100 mL of Component D1509203B (Phosphate Buffer) to obtain the DAB staining working solution. Store at 4°C protected from light; it is stable for one week. Storage at -20°C can appropriately extend the shelf life.

Note: DAB is light-sensitive. The dissolving process must be protected from light. If dissolution is difficult, methods like sonication or magnetic stirring can be used to promote dissolution.

2. Sample Preparation: Collect stressed (e.g., by heavy metals) plant seedlings or root tips. Rinse briefly with tap water and place on filter paper to absorb excess moisture.

3. Staining: Immerse the plant seedlings or root tips in the DAB staining working solution. Stain at room temperature protected from light for 2 to 6 hours, until positive areas turn deep brown and other areas are nearly colorless or show the plant's natural color. (Adjust staining time based on tissue tenderness and degree of color development.)

4. Decolorization: Carefully remove the seedlings or leaves with forceps. Rinse by immersing in distilled water 3-5 times. Place on filter paper to absorb excess moisture, then immerse in 95% ethanol and treat at 40°C for 3 to 16 hours. The purpose is to remove the plant's own chlorophyll. Replace with fresh 95% ethanol several times during decolorization.

5. Observation: Remove the seedlings or leaves with forceps. Rinse by immersing in distilled water 3-5 times. Place on filter paper to absorb excess moisture, then photograph. (After absorbing moisture, samples can be transferred into DAB staining sample preservation solution, soaked for 30 minutes, and then stored in this solution at room temperature for up to one week.)

Precautions

1. The prepared DAB working solution must be stored at 4°C in the dark and used within one week. Prolonged storage affects chromogenic performance.

2. Since hydrogen peroxide decomposes easily and any external factors may stimulate plants to produce it stressfully, plant samples must be freshly collected and stained as soon as possible. It is recommended to include negative and positive blank control groups.

3. Photograph the stained samples promptly to preserve results.

4. DAB is potentially carcinogenic. Handle with care and avoid direct contact.

5. An alternative for staining and decolorization: Place tissue in staining solution, apply vacuum (-0.1 MPa, maintain negative pressure for 20-30 min), then let stand at room temperature for 60 min. Discard staining solution. Add 95% ethanol and decolorize in a 70-80°C water bath, changing the 95% ethanol every 10 minutes until the sample's green color is completely removed.

6. For your safety and health, please wear a lab coat and disposable gloves during operation.
7. After opening the reagent, please use it as soon as possible to prevent affecting subsequent experimental results.

Appendix: Differences Between DAB Working Solutions at Different pH Values

Feature	pH 3.8	pH 5.5
Reaction Condition	The acidic condition favors the DAB-peroxidase reaction, producing a darker precipitate with higher contrast. Suitable for experiments requiring high sensitivity and clear contrast.	Relatively neutral; may be more suitable for certain specific tissues or cell types, reducing non-specific staining and improving specificity.
Staining Effect	Under acidic conditions, the DAB reaction precipitate is typically brown, with a more pronounced staining effect, suitable for detecting low levels of peroxidase activity.	Under near-neutral conditions, the DAB reaction precipitate color is relatively lighter but more uniform, suitable for detecting high levels of peroxidase activity or samples requiring fine resolution.
Experimental Application	Commonly used for quantitative determination of hydrogen peroxide content in plant tissues, especially showing high sensitivity in detection of leaves and other parts.	May be more applicable for staining certain animal tissues or cells, especially when background staining reduction is needed.
Notes	Highly acidic; requires caution during operation to avoid contact with skin and eyes.	Relatively mild, but still follow lab safety rules and wear appropriate protective equipment.

Specifications

Property	Information
Specifications & Purity	BioReagent, Biological Stain, 2.0 mg/mL
Grade	Biological Stain, BioReagent
Application	Cell Stain

Storage and Shipping

Property	Information
Storage	Store at 2-8°C, Protected from light
Shipped In	Wet ice
Stability And Storage	Store at 2-8°C long term (12 months). Store in the dark.

Contents & Storage

D1509203	Component	100 mL	Storage
D1509203A	DAB	200 mg	2-8°C. Store in the dark.
D1509203B	Phosphate Buffer (pH 3.8)	100 mL	2-8°C.

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