



**N-Histofine<sup>®</sup>**

**DAB-2V**

Peroxidase Chromogen/Substrate solution

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**3,3'-DIAMINOBENZIDINE TETRAHYDROCHLORIDE (DAB) SUBSTRATE SOLUTION  
SUBSTRATE FOR HORSERADISH PEROXIDASE**

**Package/Code**     **500 tests/425312F, 1500 tests/425314F**

**FORM**

Liquid.

This product consists of followings:	[500 tests]	[1500 tests]
Reagent A: DAB solution concentrate	... 1.2mL×2	1.2mL×6
Reagent B: DAB buffer	... 30.0mL×2	30.0mL×6
Tube	... ×1	×1
Chip (For Reagent B)	... ×1	×1
Dropping bottle	... ×1	×1

**PREPARATION of Chromogen/Substrate solution**

Add 1 drop (approx. 20μL) of Reagent A Chromogen reagent per 500μL of Reagent B Chromogen substrate into graduated tube, and place its cap then mix carefully without foaming.

Remove full dosage of the substrate solution from the tube to dropping bottle using pipette as well for use.

Store in light shielded condition at 2 to 8°C and use within 2 weeks after the preparation.

- Note:** 1. Use the substrate solution removing foams or exclusive of foams when its surface is foamed by mixing.  
2. Do not use mixture of substrate solutions prepared different day.

**USAGE**

Brownish-red precipitates are produced by the reaction of this substrate solution with peroxidase. Such precipitates are insoluble in organic solvent.

Adjust the substrate solution to room temperature prior to use.

React the objective tissue section with peroxidase conjugate in advance.

At the same time, staining of negative control slide using such as PBS in place of primary antibody is recommended to be prepared for checking the staining result.

**Chip**

Reagent B is provided with translucent chip placed on its bottle. However the chip is not allowed for applying solution drop by drop, therefore it is not suitable for small volume preparation. For such preparation, replace the translucent chip by colored chip provided exclusively for dropping is recommended. The colored chip is suitable for applying solution drop by drop.

**Dropping bottle**

Dropping bottle is used for storage of reagents, not only for dropping of substrate solution.

**Reuse**

Tube and dropping bottle are reusable after wash with purified water and dry completely.

**STAINING PROCEDURE**

Apply the substrate solution using dropping bottle covering the tissue section on the slide completely and incubate at room temperature for 5 to 20 minutes after addition, reaction and washing of peroxidase conjugate.

For chromogenic condition, adjust reaction time by microscopic observation comparing with negative control slide.

After adequate chromogenic reaction, wash the slide well with tap water.

Mount the slide with permanent mounting media and fix with cover slip after dehydrated in graded series of alcohol and cleared in xylene.

**STORAGE**

Store at 2-8°C.

**REMARKS**

For research use only. Not for diagnostic use.

## **WARNING and PRECAUTIONS**

1. Avoid contact with eyes, skin and clothing as well as ingestion and inhalation.
2. Handle Reagent A with adequate attention due to its DAB evolves into mutagens is recognized.
3. Handle Reagent B with adequate attention due to hydrogen peroxide solution contained.
4. Dispose unused Reagent A and substrate solution properly in accordance with all applicable rules and regulations of local and country due to its DAB contained.