BIOMEDICAL RESEARCH SERVICE CENTER
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DAB Substrate Kit, HRP-based In Situ Immunostaining (Cat #: A-122)

COMPONENTS: DAB Substrate: 2 ml, store at room temperature shielded from light
Peroxide Solution: 200 ml, store at room temperature shielded from light

PRODUCT DESCRIPTION: One of the most widely used enzymes for chromogenic detection in immunoassays is horseradish peroxidase (HRP), a highly stable enzyme (Clin. Chim. Acta 81:1,1977). The compound 3,3'-diaminobenzidine (DAB) is a highly sensitive HRP substrate and is thus widely used in immunohistological and immunoblotting assays (J Histochem Cytochem 36, 317, 1988). HRP in the presence of peroxide converts DAB to a brownish water-insoluble polymeric product, which is suitable for in situ immunohistochemical staining on cell or tissue sections. The working substrate solution needs to be prepared fresh by mixing DAB Substrate solution and Peroxide solution. The kit should be used within one year upon purchase for best performance.

PROTOCOL:

Preparation of working substrate solution

Mix one part of DAB Substrate with 100 parts of Peroxide Solution. Use the working solution immediately.

Immunohistochemical staining

1. Cells or tissue sections are processed with a peroxidase (HRP) detection system and washed well.
2. Add enough freshly prepared working substrate solution to cover sample. Incubate sample shielded from light at room temperature for 10 − 30 min. Optimal color development time should be empirically determined by the investigator.
3. To stop staining, rinse sample with dH_2O several times for 5 min.

NOTE:

DAB is a suspected carcinogen. Care should be taken to avoid skin contact. Please visit the product webpage or contact us for MSDS information on DAB.