Product Manual

DAB Horseradish Peroxidase Chromogenic Kit

Catalog No.: abx090660 **Revision date:** 28-Apr-20

Size: 20 ml / 100 ml

Storage: Store reagents B and C at 4 °C for frequent use or store at -20 °C for long term storage. Store reagent A at -20 °C for up to one year.

Introduction

The DAB Horseradish Peroxidase Chromogenic Kit can be used for color development in immunohistochemistry, in situ hybridization, western blot, southern blot, northern blot and EMSA. In immunohistochemistry or in situ hybridization applications, this kit can detect around 100 sections (20 ml, 0.2 ml per section) / 500 sections (100 ml, 0.2 ml per section).

Principle of the assay

DAB (3,3'-Diaminobenzidine Tetrahydrochloride) is a common substrate of HRP (Horseradish Peroxidase). DAB, when catalyzed by HRP, produces a brown precipitate that is insoluble in water and ethanol.

Kit components

1. Reagent A (DAB solution, 20 x): 1 ml / 5 ml
2. Reagent B (H_2O_2 solution, 20 x): 1 ml / 5 ml
3. Reagent C (TBS solution, 20 x): 1 ml / 5 ml

Material required but not provided

1. Distilled or deionized water
2. Ethanol
3. 1.5 ml tubes

Procedure

1. After incubating samples with HRP-conjugated antibody, wash with TBS or PBS thoroughly for 5 minutes. Repeat this 3 more times for a total of 4 washes.
2. Add 50 µl of Reagent A, 50 µl of Reagent B and 50 µl of Reagent C into 1 ml of distilled water. Mix thoroughly. Add this mixed solution on to the sample and allow the color to develop for 10-30 minutes.
3. Wash with distilled water to stop the reaction. If necessary, re-stain with haematoxylin for 30-60 seconds and wash with water.
4. Dehydrate with ethanol to ensure the tissues are embedded in the section. Use xylene to make the section transparent. Seal the section and observe under a microscope.