

**DESCRIPTION**

DAB (3, 3'-diaminobenzidine) HRP substrate produces a dark brown reaction product and can be used for both immuno-histochemical and blotting applications. This substrate kit is an optimized formula that is crisper and is 3-4 times more sensitive than conventional DAB substrate kit.

**PROTOCOL****PREPARATION OF SUBSTRATE WORKING SOLUTION:**

Add 1 drop (approximately 30 µl) of DAB Chromogen concentrate to 1 ml DAB Diluent. Mix well before use.

**INSTRUCTIONS FOR USE:**

After incubation with a peroxidase (HRP) detection system, rinse well. Incubate with the substrate working solution at room temperature for 2-10 minutes. Optimal development times should be determined by the investigator. Wash for 5 minutes in water or PBS. Counterstain if desired.

**NOTE:**

Unused working solution is stable up to 14 days if stored at 2-8 °C or up to 5 days if stored at room temperature (approximately 23 °C). Working solution may change color during storage but this will have no effect on the quality or intensity of the staining. No sample-obscuring precipitate will form. It is not necessary or recommended to add detergent to ImmPACT™ DAB working solution to reduce surface tension (e.g. for use in an automated stainer). The DAB reaction product can be intensified using a DAB Enhancing Solution after development.

**IMPORTANT:**

DAB is a suspected carcinogen. Appropriate care should be exercised when using this reagent including gloves, eye protection, lab coats, and good laboratory procedures. Dispose in accordance with local regulation.

**STORAGE**

Store reagents in original bottles at 2-8°C.

Avoid storing reagents or working solution in strong direct light.