

CD117/c-kit PolyDetector HRP/DAB Detection System

Intended Use *For Research Use Only*

**Summary And
Explanation**

The **CD117/c-kit PolyDetector HRP/DAB Detection System** is a non-biotin, 2-step polymeric detection system that allows for the Immunohistochemical detection of CD117 expression in normal and neoplastic formalin-fixed paraffin-embedded tissues.

The **CD117/c-kit PolyDetector HRP/DAB Detection System** has been developed using a proprietary tandem hyperlabelling technology used to directly label immunoglobulins with enzymes. This ensures consistent and reproducible immunostaining for CD117,

CD117 is a tyrosine kinase receptor for stem cell factor (SCF), also known as "steel factor" or "c-kit ligand". *C-kit* is a polypeptide that activates bone marrow precursors of a number of blood cells, but its receptor is also present on other cells. *C-kit* mutations in the interstitial cells of Cajal in the digestive tract are probably the key to gastrointestinal stromal tumors (GISTs).

CD117 is found on interstitial cells of Cajal, germ cells, bone marrow stem cells, melanocytes, breast epithelium and mast cells. This receptor is found on a wide variety of tumor cells (Follicular and Papillary carcinoma of Thyroid, Adenocarcinomas from endometrium, lung, ovary, pancreas, breast; malignant melanoma, endodermal sinus tumor, small cell carcinoma) but has been particularly useful in differentiating Gastrointestinal Stromal Tumors (GIST) from Kaposi's sarcoma and tumors of smooth muscle origin.

Presentation

The **CD117/c-kit PolyDetector HRP/DAB Detection System** contains a rabbit monoclonal CD117 prediluted antibody clone YR145, prediluted rabbit negative control, positive control slides with a normal and tumoral c-kit tissues, ImmunoRetriever with Citrate heat epitope retrieval solution, Peroxidase Blocker solution, an Anti-Rabbit Horseradish Peroxidase solution, a DAB Buffer and a DAB Chromogen solution. All the components are buffered and contain proteins, stabilizers and a non-azide anti-microbial.

Availability

Component Name	Catalog Number		
	BSB 0254 (70 Tests)	BSB 0255 (150 Tests)	BSB 0256 (500 Tests)
Rabbit Monoclonal CD117 Prediluted Antibody	7 mL	15 mL	50 mL
Rabbit Negative Control	7 mL	15 mL	50 mL
Positive Control Slides	5 Slides	10 Slides	30 Slides
20X ImmunoRetriever with Citrate	50 mL	100 mL	200 mL
PolyDetector Peroxidase Blocker	15 mL	30 mL	100 mL
PolyDetector HRP Label	15 mL	30 mL	100 mL
PolyDetector DAB Buffer	15 mL	50 mL	200 mL
PolyDetector DAB Chromogen	1 mL	3 mL	12 mL

Storage

Store at 2-8°C

Stability 3 years

The PolyDetector Solutions are stable at 2-8°C for up to 3 years from when originally produced (see expiration date on product label). These products are stable up to the expiration date on the label. Do not use this product after the expiration date. Adhere to all local laws when disposing of this product.

Protocol

Suggested protocol on reverse

Preparation of Working Solutions

The **Prediluted CD117 Rabbit Antibody**, **Prediluted Rabbit Negative Control**, **PolyDetector Peroxidase Blocker**, and **Anti-Rabbit Horseradish Peroxidase Label** are ready-to-use working solutions and require no further preparation. The **20X ImmunoRetriever with Citrate** needs to be diluted with distilled water to a 1X concentration before use by adding 50 mL of 20X ImmunoRetriever with Citrate to 950 mL of distilled water and mixing. The **DAB Chromogen** is concentrated and needs to be diluted and mixed into the **DAB Buffer** to produce the working DAB substrate-chromogen solution. For each 1 mL of working DAB substrate-chromogen solution required for the experiment, 1 drop of **DAB Chromogen** should be added and mixed into 1 mL of **DAB Buffer**.

Working DAB Substrate-Chromogen Required	1 mL	2 mL	3 mL
DAB Buffer	1 mL	2 mL	3 mL
DAB Chromogen	1 drop	2 drops	3 drops

Recommended Immunohistochemical Protocol

1. Deparaffinize and rehydrate tissues if necessary.
2. Place cut and dried slides in the prepared **1X ImmunoRetriever Citrate** and heat treat for 15 minutes in a Pressure Cooker or 45 Minutes in a Water Bath or Steamer at 95-98°C.
3. Wash with 5 changes of IHC Wash buffer.
4. Cover tissue with the **PolyDetector Peroxidase Blocker** for 5 min.
5. Wash with 3 changes of IHC wash buffer.
6. Cover tissue with the **CD117 Primary Antibody** or **Negative Control** and incubate for 45 minutes.
7. Wash with 3 changes of IHC wash buffer.
8. Cover tissue with the **PolyDetector HRP Label**, incubate for 45 min.
9. Rinse with 3 changes of IHC wash buffer.
10. Prepare DAB by adding one drop of **PolyDetector DAB Chromogen** per 1 mL of **PolyDetector DAB Buffer** and mix.
11. Cover tissue with the prepared DAB substrate-chromogen solution, incubate for 10 min.
12. Rinse with 5 changes of DI water
13. Counterstain and then dehydrate.
14. Coverslip

Abbreviated Immunohistochemical Protocol

Step	PolyDetector HRP
Peroxidase Blocker	5 minutes
Primary Antibody	45 minutes
HRP Label	45 minutes
DAB Substrate-Chromogen	10 minutes
Counterstaining	Time varies with counterstain

Precautions

For professional users. Minimize microbial contamination of reagents. Proper handling procedures should be used.



The following are the appropriate Risk and Safety requirements:

Harmful if swallowed. Irritating to eyes, respiratory system, and skin. Avoid contact with skin and eyes. In case of contact with eyes, rinse immediately with plenty of water and seek medical advice. Wear suitable gloves and eye/face protection. If swallowed seek medical advice immediately and show the product container



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