For Research Use Only.
Not for use in diagnostic procedures.

POLYCLONAL ANTIBODY

Anti-CD117 (c-Kit) (Human) pAb

Code No.	Quantity	Form
566-H	6 mL	Ready for use

BACKGROUND: c-Kit, also known as stem cell factor receptor, steel factor receptor or CD117 is classified as a type III receptor tyrosine kinase (RTK) belonging to the platelet-derived growth factor receptor subfamily. Binding of stem cell factor (SCF), known as c-Kit ligand to c-Kit, initiates autophosphorylation of the receptor, subsequently leading to promotes a signal transduction cascade through Ras-Raf-MAP kinase cascade, phosphatidylinositol-3-kinase, src family kinases, and JAK/STAT pathways. The roles of c-Kit include maturation of hematopoietic and primordial germ cells precursors and melanocytes during embryonic development. In acute myeloid leukemia (AML), c-Kit has been proposed to play a functional role, and becomes target molecule for drug development.

SOURCE: This antibody was purified from rabbit serum using the synthesized peptides (C-terminal of c-Kit gene product; K963) coupled protein A agarose column. The rabbit was immunized with carrier protein conjugated synthesized peptides (K963).

FORMULATION: 6 mL volume of pre-diluted antibody in 20 mM HEPES (pH 7.2), containing 135 mM NaCl, 1% BSA and 0.09% NaN_3.
*Azide may react with copper or lead in plumbing system to form explosive metal azides. Therefore, always flush plenty of water when disposing materials containing azide into drain.

STORAGE: This antibody solution is stable for 3 years from the date of purchase when stored at 4°C.

REACTIVITY: This antibody reacts with c-Kit on Immunohistochemistry.

APPLICATIONS:
Immunohistochemistry; Ready for use
Heat treatment is necessary for paraffin embedded sections.
Microwave oven; 2 times for 10 minutes each in 10 mM citrate buffer (pH 6.5)

*Please refer to the data sheet (MBL code no. 566) for other applications.

Detailed procedure is provided in the following **PROTOCOL**.

SPECIES CROSS REACTIVITY:

Species	Human	Mouse	Rat
Cell and Tissue	HEL, pancreas	Not tested	Not tested
Reactivity on IHC	+		

INTENDED USE:
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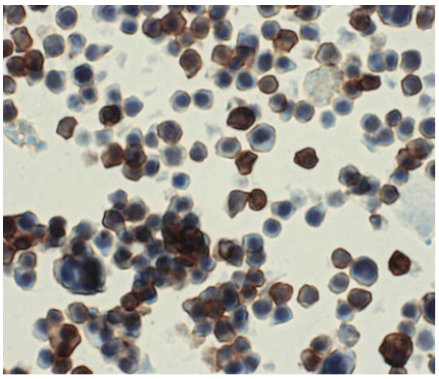
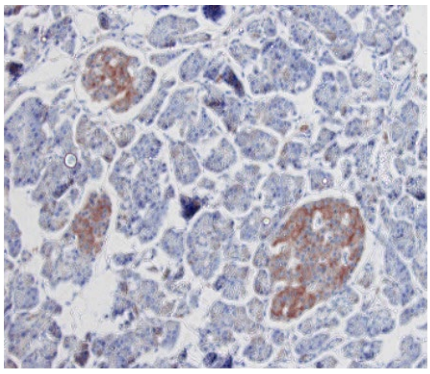


Immunohistochemical detection of CD117/c-Kit on paraffin embedded section of HEL cells with 566-H.



Immunohistochemical detection of CD117/c-Kit on paraffin embedded section of human pancreas with 566-H.

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REFERENCES:

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2) Lyford, G. L., *et al*., *Gut* **51**, 496-501 (2002)
3) Sakurai, S., *et al*., *Am. J. Pathol*. **154**, 23-28 (1999)
4) Tsuura, T., *et al*., *Virchows Archiv.* **424**, 135-141 (1994)
5) Hidi, K., *et al*., *Oncogene* **6**, 2291-2296 (1991)
6) Yarden,Y., *et al*., *EMBO J.* **6**, 3341-3351 (1987)

This antibody is used in the reference number 1) - 3).

The descriptions of the following protocols are examples.
Each user should determine the appropriate condition.

PROTOCOL:

Immunohistochemical staining for paraffin-embedded sections

1) Deparaffinize the sections with Xylene 3 times for 3-5 minutes each.
2) Wash the slides with Ethanol 3 times for 3-5 minutes each.
3) Wash the slides with PBS 3 times for 3-5 minutes each.
4) Heat treatment
   Heat treatment by microwave oven:
   Place the slides put on staining basket in 500 mL beaker with 500 mL citrate buffer (pH 6.5). Cover the beaker with plastic wrap, then process the slides 2 times for 10 minutes each at 500 W with microwave oven. Let the slides cool down in the beaker at room temperature for about 40 minutes.
5) Remove the slides from the citrate buffer and cover each section with 3% H_2O_2 for 10 minutes at room temperature to block endogenous peroxidase activity. Wash 3 times in PBS for 5 minutes each.
6) Remove the slides from PBS, wipe gently around each section and cover tissues with 1% BSA in PBS for 5 minutes to block non-specific staining. Do not wash.
7) Tip off the blocking buffer, wipe gently around each section and cover tissues with one or two drops of primary antibody (Ready for use).
8) Incubate the sections for 1 hour at room temperature.
9) Wash the slides 3 times in PBS for 5 minutes each.
10) Wipe gently around the section and incubate with Histostar™ (Rb) (MBL; code no. 8466) for 30 min. at room temperature. Wash as in step 9).
11) Visualize by reacting for 5 min. with Histostar™ DAB Substrate Solution (MBL; code no. 8469). *DAB is a suspect carcinogen and must be handled with care. Always wear gloves.
12) Wash the slides in water for 5 minutes.
13) Counter stain in hematoxylin for 1 minute, wash the slides 3 times in water for 5 minutes each, and then immerse the slides in PBS for 5 minutes. Dehydrate by immersing in Ethanol 3 times for 3 minutes each, followed by immersing in Xylene 3 times for 3 minutes each.
14) Now ready for mounting.

(Positive controls for immunohistochemistry; HEL and human pancreas)

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