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SZABO-SCANDIC HandelsgmbH

Quellenstraße 110, A-1100 Wien

T. +43(0)1 489 3961-0

F. +43(0)1 489 3961-7

mail@szabo-scandic.com

www.szabo-scandic.com

linkedin.com/company/szaboscandic

Alkaline Phosphatase Mouse Kit (Fast Red)

CLSG80130

Intended Use:
This product is intended for qualitative immunohistochemistry with normal and neoplastic formalin-fixed, paraffin-embedded tissue sections, to be viewed by light microscopy.

Introduction:
This kit is designed to label specific primary antibodies immunohistochemically on tissue sections. The immunohistochemical protocol defined in this brochure is just a guideline. We encourage the individual laboratory to optimize its own protocol based upon tissue fixation conditions, primary antibodies employed, and the user's experience. These reagents were tested and quality controlled using tissue sections, however, they can also be optimized for cell smears and cytospin preparations. Sufficient reagents are provided to run 50 to 100 tests.

All immunohistochemical techniques require the specific antibody employed to be labeled that they can be easily seen when attached to cellular antigens. At the same time the sensitivity of the immunoperoxidase techniques are central to wide variety of specific antigen localization. Alkaline phosphatase based kits are of special value for staining tissues that have high endogenous peroxidase activity. Alkaline phosphatase is unaffected by endogenous peroxidase and therefore results in cleaner background. Our kit is based on direct SA-ALP conjugate technology. The linker reagent is biotinylated anti-mouse immunoglobulins, capable of labeling mouse primary antibodies.

Reagents Supplied:
Bottle 1 - Alkaline Phosphatase Enhancer: 10mL clear solution.
Used to increase the intensity of signal

Bottle 2 - Linker Reagent: 10mL clear yellow solution of biotinylated anti-mouse immunoglobulins.
Bottle 3 - ALP Tracer Reagent: 10mL clear cherry red solution of conjugated streptavidin alkaline phosphatase.
Detector Reagents - PermaRed/AP Buffer 15ml
PermaRed/AP Chromogen 1ml

Storage: All the reagents should be stored at 2-8°C. Do not freeze. Do not use beyond the expiration date stated on the label.

Specimen Preparation:
Tissues fixed in 10% formalin are suitable for use prior to paraffin embedding. The user is advised to validate the use of the products with their tissue specimens prepared and handled in accordance with their laboratory practices.

Reagents Preparation:
Except for the chromogen, all reagents are provided in ready to use form. Aliquot 1mL of PermaRed/AP Substrate Buffer in a mixing bottle. Add one drop (~20µL) of PermaRed/AP Chromogen. Replace tip, mix, and allow solution to reach room temperature before using.

In CANADA: Toll Free: 1-800-268-5058
4410 Paletta Court, Burlington, ON L7L 5R2 ph: (289) 288-0001, fax: (289) 288-0020
e-mail: general@cedarlanelabs.com

In the USA: Toll Free: 1-800-721-1644
1210 Turrentine Street, Burlington, NC 27215 ph: (336) 513-5135, fax: (336) 513-5138
e-mail: service@cedarlanelabs.com

Note: The working chromogen-substrate solution should be prepared fresh and used within 20-30 minutes of preparation. Any solution not used during this period should be discarded.

Positive and Negative controls:
Each immunostaining run should include a known positive and a negative control to assure the proper functioning of staining system and valid interpretation of the results.

Staining Protocol:
Step I Removal of paraffin wax: Deparaffinize tissue sections according to the established procedure in your lab and bring tissues to wash buffer.
Step II Endogenous Alkaline Phosphatase Blocking: This is an optional step and should be performed only if tissue is suspected to have high endogenous alkaline phosphatase activity. Apply enough drops of Alkaline Phosphastase Blocker to cover tissue. Incubate for 5-10 minutes at room temperature.
Step III Washing: Drain off excess reagent. Rinse with wash buffer three times for 1 minute each time. Drain off excess buffer and carefully wipe slide around the tissue to remove excess buffer from the glass leaving the tissue wet.

Immunostaining Protocol I:
This protocol is recommended for optimally fixed tissues with abundant antigens in the tissue and for high affinity primary antibodies.
Step I Primary antibody: Apply enough drops of primary antibody to cover the tissue section. Incubate according to the manufacture's recommended conditions. Wash and wipe slides as described above.
Step II Linker reagent: Apply enough drops of linker reagent to cover the tissue section. Incubate for 10 minutes at room temperature. Wash slides as described previously.
Step III Tracer Reagent: Apply enough drops of tracer reagent to cover the tissue section. Incubate for 10 minutes at room temperature. Wash slides as described previously.
Step IV Substrate/Chromogen: Apply working chromogen solution for 5-10 minutes at room temperature for color development. For best results, look under the microscope for signal development. Once desired signal to noise ratio is achieved, stop the reaction by washing slides in wash buffer. Note development time and follow it during subsequent incubations.

Immunostaining Protocol II:
This protocol is recommended for less than optimally fixed tissues with low antigenic density in the tissue and for low affinity primary antibodies.
Step I Primary antibody: Apply enough drops of primary antibody to cover the tissue section. Incubate according to the manufacture's recommended conditions. Wash slides as described previously.
Step II Linker reagent: Apply enough drops of linker reagent to cover the tissue section. Incubate for 20 minutes at room temperature. Wash slides as described previously.
Step III Tracer Reagent: Apply enough drops of tracer reagent to cover the tissue section. Incubate for 20 minutes at room temperature. Wash slides as described previously.
Step IV Substrate/Chromogen: Apply working chromogen solution for 5-10 minutes at room temperature for color development. For the best results, look under the microscope for the signal development. Once desired signal to noise ratio is achieved, stop the reaction by washing the slide in wash buffer. Note development time and follow it during subsequent incubations.
Step V Wash slides and counter stain with an appropriate counter stain. Mount and observe staining under the microscope.

Enhanced Staining
If additional signal is desired, treat tissue sections with Alkaline Phoshpatase Enhancer for 1 minute after Step III. Drain off excess enhancer and add substrate/chromogen solution without any wash.

References:
(i) Bayer et al., Method Enzymol. 62, 308 (1979).
(ii) Chaiet & Wolf, Arch. Biochem. Biophys. 106, 1 (1964).
(iii) Giorno, Diag. Immuno. 2, 161 (1984).
(iv) Hsu et al., J Histo. Cyto. 29, 577 (1981).
(v) Nadji & Morales, Anat. pathol. 14, 767 (1983).
(vi) Nagle et al. J Histo. Chem. 31, 1010 (1983).
(vii) Larsson, Applied Immunohistochem. 1, 2, (1993).
(viii) Petrusz & Ordronneau, In Polak & Van noorden eds., Immunocytochemistry: Practical applications in pathology and biology. Bristol wright-PSG, 212, (1983).

Laboratory Reagent For Research Use Only