Novocastra™ Lyophilized Mouse Monoclonal Antibody Lambda Light Chain

Product Code: NCL-LAM

Intended Use	FOR RESEARCH USE ONLY.
Specificity	Human lambda immunoglobulin light chain (free or bound to heavy chain isotypes).
Clone	HP-6054
Ig Class	IgG2a
Antigen Used for Immunizations	Purified human IgG myeloma proteins covalently coupled to polyaminostyrene micro beads.
Hybridoma Partner	Mouse myeloma (p3-NS1-Ag4-1).
Preparation	Lyophilized tissue culture supernatant containing 15 mM sodium azide. Reconstitute with the volume of sterile distilled water indicated on the vial label.
Effective on Frozen Tissue	Yes
Effective on Paraffin Wax Embedded Tissue	Yes. It is recommended that tissue be fixed in either primary formol sublimate or secondary formol sublimate after primary formalin fixation. Testing material fixed in formalin has revealed a progressive loss of reactivity when the fixation time exceeds 24 hours.
Recommendations on Use	Immunohistochemistry: Typical working dilution 1:200–1:300. Trypsin digestion of paraffin sections may enhance staining in some cases. 60 minutes primary antibody incubation at 25 °C. Standard ABC technique. Western Blotting: Typical working dilution 1:50–1:100.
Positive Controls	Immunohistochemistry: Tonsil. Western Blotting: Tonsil.
Staining Pattern	Generally, in paraffin-embedded tissue cytoplasmic immunoglobulin will stain, for example, in myeloma or plasmacytoma. Membrane bound immunoglobulin is generally only demonstrated using frozen sections.
Storage and Stability	Store unopened lyophilized antibody at 4 °C. Under these conditions, there is no significant loss in product performance up to the expiry date indicated on the vial label. The reconstituted antibody is stable for at least two months when stored at 4 °C. For long term storage, it is recommended that aliquots of the antibody are frozen at -20 °C (frost-free freezers are not recommended). Repeated freezing and thawing must be avoided. Prepare working dilutions on the day of use.
General Overview	Immunoglobulins are polypeptides and comprise five major classes; immunoglobulin G (IgG), IgA, IgM, IgD and IgE. Each immunoglobulin consists of two identical heavy (H) chains and two identical light (L) chains. These are also subdivided into sub classes eg. IgG1. There are two classes of light chain; kappa and lambda. The ratio of kappa chains and light chains varies between Ig classes and subclasses, but is also species specific. In humans, approximately 60% of light chains are kappa. However, in any particular immunoglobulin molecule the light chain will be either kappa or lambda - never a mixture. B cells contain either kappa or lambda mRNA.
General References	Reimer C B, Philips D J, Aloisio C H, et al.. Hybridoma. 3: 263–275 (1984).

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BIOSYSTEMS

Instructions for Use

Trypsin Digestion for Immunohistochemical Demonstration on Paraffin Sections

1. Preheat the following to 37 °C using a water bath:
   (i) 200 mL of TBS
   (ii) 200 mL of distilled water.
2. Dissolve 0.2 g Trypsin 250 and 0.2 g Calcium chloride in the 200 mL of TBS.
3. Once the Trypsin solution is at 37 °C, pH to 7.8 with 1 M sodium hydroxide.
4. Place rehydrated paraffin sections in the distilled water to preheat the sections to 37 °C for a minimum of 5 minutes.
5. Incubate sections in Trypsin solution at 37 °C. The time required will depend on the antibody and tissue, however, 30 minutes is usually sufficient.
6. Rinse sections in running tap water.
7. Proceed with immunohistochemistry protocol.

Reagents Required but not Supplied

50 mM Tris-buffered saline

Trypsin 250: Difco order code 0152–13 (available from Becton Dickinson).

Calcium chloride

1 M Sodium Hydroxide

** Trypsin containing chymotrypsin should always be used. The enzyme activities can vary from a supplier and between batches. Such variations may affect the incubation time required.*