Novocastra™ Liquid Mouse Monoclonal Antibody Carbonic Anhydrase IX

Product Code: NCL-L-CAIX

Intended Use FOR RESEARCH USE ONLY.
Specificity Human Carbonic Anhydrase IX
Clone TH22
Ig Class IgG2a

Antigen Used for Immunizations Prokaryotic recombinant protein corresponding to 118 amino acids of the human Carbonic Anhydrase IX molecule.

Hybridoma Partner Mouse myeloma (p3-NS1-Ag4.1)

Preparation Liquid tissue culture supernatant containing 15 mM sodium azide. Volume as indicated on vial label.

Effective on Frozen Tissue Not evaluated.
Effective on Paraffin Wax Embedded Tissue Yes (using heat induced epitope retrieval with Citrate-based buffer, pH 6.0: see overleaf).

Recommendations on Use Immunohistochemistry: Typical working dilution 1:100. Heat induced epitope retrieval technique using Citrate-based buffer, pH 6.0. 30 minutes primary antibody incubation at 25°C. Polymer detection recommended.

Positive Controls Immunohistochemistry: Stomach

Staining Pattern Membrane and Cytoplasmic

Storage and Stability Store liquid 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. Prepare working dilutions on the day of use.

General Overview Carbonic anhydrase (CA) is an enzyme that assists rapid interconversion of carbon dioxide and water into carbonic acid, protons, and bicarbonate ions. Originally named MN/G250, carbonic anhydrase IX (CAIX) is a cell surface transmembrane protein, that is predominantly found in the gastrointestinal tract and gall bladder. The glandular regions of normal colon are reported to be negative, but in the case of adenocarcinoma, the glands are positive. CAIX is also reported to be expressed in common epithelial tumors such as carcinomas of the esophagus, lung, colon, kidney, cervix and non-small cell lung carcinoma. In breast carcinomas, CAIX expression has been reported to be associated with malignant tissue. Expression of CAIX is reported to be absent in normal kidney, chromophobe carcinomas or oncocytomas, but expressed in clear cell renal carcinomas.

Instructions for Use
Heat Induced Epitope Retrieval
Combined With Polymer Detection For
Immunohistochemical Demonstration
On Paraffin Sections

1. Cut and mount sections on slides coated with a suitable tissue adhesive.
2. Deyparaffinize sections and rehydrate to distilled water.
3. Place sections in 0.5% hydrogen peroxide/methanol for 10 minutes (or use other appropriate endogenous peroxidase blocking procedure). Wash sections in tap water.
4. Heat 1500 mL of the recommended epitope retrieval solution (Citrate based pH 6.0 - Epitope Retrieval Solution unless otherwise indicated overleaf) in a stainless steel pressure cooker until boiling. Cover but do not lock lid.
5. Position slides into metal staining racks (do not place slides close together as uneven staining may occur) and lower into pressure cooker ensuring slides are completely immersed in epitope retrieval solution. Lock lid.
6. When the pressure cooker reaches operating temperature and pressure (after about 5 minutes) start a timer for 1 minute (unless otherwise indicated on the data sheet).
7. When the timer rings, remove pressure cooker from heat source and run under cold water with lid on. DO NOT OPEN LID UNTIL THE INDICATORS SHOW THAT PRESSURE HAS BEEN RELEASED. Open lid, remove slides and place immediately into a bath of tap water.
8. Wash sections once using fresh Tris-Buffered Saline (TBS, pH 7.6) buffer for 5 minutes.
9. Place sections in diluted normal serum (eg NCL-G-SERUM) for 10 minutes.
10. Incubate sections with primary antibody.
11. Wash twice, each time using fresh TBS buffer for 5 minutes.
12. For visualization of the bound primary antibody, follow instructions supplied with the Polymer Detection System.
13. Counterstain with hematoxylin (if required), dehydrate and mount.
* (In most applications, Phosphate Buffered Saline, pH 7.6, can be used instead of TBS, pH 7.6).

Safety Note
To ensure the correct and safe use of your pressure cooker, PLEASE READ THE MANUFACTURER’S INSTRUCTIONS.