Human Neprilysin/CD10 Antibody

Monoclonal Mouse IgG_{2B} Clone # 715820

Catalog Number: MAB11821

DESCRIPTION

Species Reactivity	Human
Specificity	Detects human Neprilysin/CD10 in direct ELISAs and Western blots. In direct ELISAs, less than 5% cross-reactivity with recombinant mouse (rm) Neprilysin is observed, and no cross-reactivity with rmKell, recombinant human (rh) ECE-1, rhECE-2, or rhNeprilysin-2 is observed.
Source	Monoclonal Mouse IgG_{2B} Clone # 715820
Purification	Protein A or G purified from hybridoma culture supernatant
Immunogen	Chinese hamster ovary cell line CHO-derived recombinant human Neprilysin/CD10 Tyr52-Trp750 Accession # P08473
Formulation	Lyophilized from a 0.2 μm filtered solution in PBS with Trehalose. See Certificate of Analysis for details. *Small pack size (-SP) is supplied either lyophilized or as a 0.2 μm filtered solution in PBS.

APPLICATIONS

Please Note: *Optimal dilutions should be determined by each laboratory for each application. General Protocols are available in the Technical Information section on our website.*

	Recommended Concentration	Sample
Western Blot	0.1 µg/mL	See Below
Multiplex Immunofluorescence	0.25 µg/mL	Immersion fixed paraffin-embedded sections of human Kidney
Immunohistochemistry	0.5-25 µg/mL	See Below

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China | info.cn@bio-techne.com TEL: 400.821.3475

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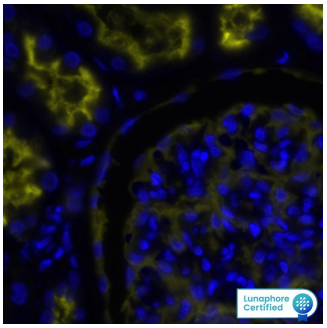
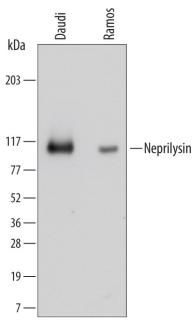
DATA

Multiplex Immunofluorescence



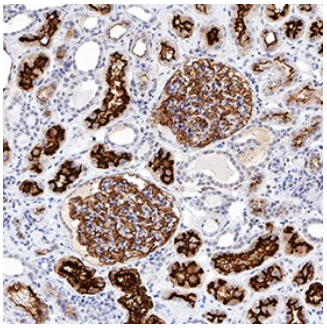
Detection of Neprilysin/CD10 in Human Kidney via seqIF™ staining on COMET™ Neprilysin/CD10 Antibody was detected in immersion fixed paraffin-embedded sections of human Kidney using Mouse Anti-Human Neprilysin/CD10, Monoclonal Antibody (Catalog # MAB11821) at 0.25ug/mL at 37 ° Celsius for 2 minutes. Before incubation with the primary antibody, tissue underwent an all-in-one dewaxing and antigen retrieval preprocessing using PreTreatment Module (PT Module) and Dewax and HIER Buffer H (pH 9; Epredia Catalog # TA-999-DHBH). Tissue was stained using the Alexa Fluor™ 555 Goat anti-Mouse IgG Secondary Antibody at 1:100 at 37 ° Celsius for 2 minutes. (Yellow; Lunaphore Catalog # DR555MS) and counterstained with DAPI (blue; Lunaphore Catalog # DR100). Specific staining was localized to the membrane. Protocol available in COMET™ Panel Builder.

Western Blot



Detection of Human Neprilysin/CD10 by Western Blot. Western blot shows lysates of Daudi human Burkitt's lymphoma cell line and Ramos human Burkitt's lymphoma cell line. PVDF membrane was probed with 0.1 µg/mL of Mouse Anti-Human Neprilysin/CD10 Monoclonal Antibody (Catalog # MAB11821) followed by HRP-conjugated Anti-Mouse IgG Secondary Antibody (Catalog # Catalog # HAF007). A specific band was detected for Neprilysin/CD10 at approximately 100 kDa (as indicated). This experiment was conducted under reducing conditions and using Immunoblot Buffer Group 1.

Immunohistochemistry



Neprilysin/CD10 in Human Kidney. Neprilysin/CD10 was detected in immersion fixed paraffin-embedded sections of human kidney using Mouse Anti-Human Neprilysin/CD10 Monoclonal Antibody (Catalog # MAB11821) at 0.5 µg/mL for 1 hour at room temperature followed by incubation with the Anti-Mouse IgG VisUCyte™ HRP Polymer Antibody (Catalog # Catalog # VC001). Tissue was stained using DAB (brown) and counterstained with hematoxylin (blue). Specific staining was localized to convoluted tubules and glomeruli. View our protocol for IHC Staining with VisUCyte HRP Polymer Detection Reagents.

PREPARATION AND STORAGE

Reconstitution	Sterile PBS to a final concentration of 0.5 mg/mL. For liquid material, refer to CoA for concentration.
Shipping	Lyophilized product is shipped at ambient temperature. Liquid small pack size (-SP) is shipped with polar packs. Upon receipt, store immediately at the temperature recommended below.
Stability & Storage	**Use a manual defrost freezer and avoid repeated freeze-thaw cycles.** • 12 months from date of receipt, -20 to -70 °C as supplied. • 1 month, 2 to 8 °C under sterile conditions after reconstitution. • 6 months, -20 to -70 °C under sterile conditions after reconstitution.

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BACKGROUND

Neprilysin/CD10, also known as NEP and neutral endopeptidase 24.11, is a zinc metallopeptidase expressed at the cell surface of a variety of cells. The enzyme functions both as an endopeptidase with a thermolysin-like specificity and as a dipeptidylcarboxypeptidase. NEP has been shown to be involved in the degradation of enkephalins in the mammalian brain and the inactivation of circulating atrial natriuretic peptide (1, 2). NEP has also been identified as the common acute lymphocytic leukemia antigen (CALLA), and is expressed on the surface of lymphocytes in some disease states (3, 4). These and other observations have resulted in considerable interest in NEP as a target for analgesics and antihypertensive drugs. NEP is also a major degrading enzyme of amyloid β peptide (Aβ) in the brain, indicating that down-regulation of NEP activity, which could be caused by aging, can contribute to the development of Alzheimer's disease by promoting Aβ accumulation (5).

References:

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2. Kenny, A.J. and Stephenson, S.L. (1988) FEBS Lett. **232**:1.
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4. Shipp, M.A. *et al*. (1988) Proc. Natl. Acad. Sci. USA **85**:4819.
5. Itwata, N. *et al*. (2001) Science **292**:1550.

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