

DESCRIPTION	
Species Reactivity	Human
Specificity	Detects human Ezrin in direct ELISAs and Western blots.
Source	Recombinant Monoclonal Rabbit IgG Clone # 2086A
Purification	Protein A or G purified from cell culture supernatant
Immunogen	<i>E. coli</i> -derived recombinant human Ezrin Lys438-Arg562 Accession # P15311
Formulation	Supplied as a solution in PBS containing BSA, Glycerol and Sodium Azide. 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	0.2 µg/mL	See Below
Immunocytochemistry	1-15 µg/mL	See Below
Immunohistochemistry	1-25 µg/mL	See Below
Intracellular Staining by Flow Cytometry	0.25 µg/10 ⁶ cells	See Below

DATA

Western Blot

Detection of Human Ezrin by Western Blot. Western blot shows lysates of HeLa human cervical epithelial carcinoma cell line, OVCAR-3 human ovarian carcinoma cell line, and human lung tissue. PVDF membrane was probed with 0.2 µg/mL of Rabbit Anti-Human Ezrin Monoclonal Antibody (Catalog # MAB7239) followed by HRP-conjugated Anti-Rabbit IgG Secondary Antibody (Catalog # HAF008). A specific band was detected for Ezrin at approximately 80 kDa (as indicated). This experiment was conducted under reducing conditions and using Immunoblot Buffer Group 1.

Intracellular Staining by Flow Cytometry

Detection of Ezrin in HeLa Human Cell Line by Flow Cytometry. HeLa human cervical epithelial carcinoma cell line was stained with Rabbit Anti-Human Ezrin Monoclonal Antibody (Catalog # MAB7239, filled histogram) or isotype control antibody (Catalog # AB-105-C, open histogram), followed by Allophycocyanin-conjugated Anti-Rabbit IgG Secondary Antibody (Catalog # F0111). To facilitate intracellular staining, cells were fixed with Flow Cytometry Fixation Buffer (Catalog # FC004) and permeabilized with Flow Cytometry Permeabilization/Wash Buffer I (Catalog # FC005). View our protocol for Staining Intracellular Molecules.

Immunocytochemistry

Ezrin in HeLa Human Cell Line. Ezrin was detected in immersion fixed HeLa human cervical epithelial carcinoma cell line using Rabbit Anti-Human Ezrin Monoclonal Antibody (Catalog # MAB7239) at 1 µg/mL for 3 hours at room temperature. Cells were stained using the NorthernLights™ 557-conjugated Anti-Rabbit IgG Secondary Antibody (red; Catalog # NL004) and counterstained with DAPI (blue). Specific staining was localized to cytoplasm. View our protocol for Fluorescent ICC Staining of Cells on Coverslips.

Immunohistochemistry

Ezrin in Human Pancreas. Ezrin was detected in immersion fixed paraffin-embedded sections of human pancreas using Rabbit Anti-Human Ezrin Monoclonal Antibody (Catalog # MAB7239) at 1 µg/mL for 1 hour at room temperature followed by incubation with the Anti-Rabbit IgG VisUCyte™ HRP Polymer Antibody (Catalog # VC003). Tissue was stained using DAB (brown) and counterstained with hematoxylin (blue). Specific staining was localized to cytoplasm in exocrine cells. View our protocol for IHC Staining with VisUCyte HRP Polymer Detection Reagents.

PREPARATION AND STORAGE

Shipping	The product is shipped with polar packs. Upon receipt, store it immediately at the temperature recommended below. *Small pack size (-SP) is shipped with polar packs. Upon receipt, store it immediately at -20 to -70 °C
Stability & Storage	Use a manual defrost freezer and avoid repeated freeze-thaw cycles. <ul style="list-style-type: none"> • 12 months from date of receipt, -20 to -70 °C, as supplied. • 1 month, 2 to 8 °C under sterile conditions after opening. • 6 months, -20 to -70 °C under sterile conditions after opening.

BACKGROUND

EZRIN (also Cytovillin, Villin2 and p81) is a founding member of the ERM family, Band 4.1 Superfamily of proteins. Although its predicted MW is 69 kDa, it runs anomalously at 77-82 kDa in SDS-PAGE. ERZIN is expressed by epithelial cells where it serves as a linker between the cell membrane and the actin cytoskeleton. Its presence is particularly strong in microvilli where it helps organize this structure. In addition, ERZIN also organizes microtubules in lymphocytes at or near the immunological synapse by interacting with Glg1. Human EZRIN is 585 amino acids (aa) in length. It contains a band 4.1 homology/FERM domain that binds CD44, ICAM-1, EBP50 and ERM family members (aa 1-295), a central α -helical region (aa 296-352), and a C-terminal ERM and actin-binding/FERM C domain (aa 353-586). EZRIN exists as either a monomer, or a homo/heterodimer. EZRIN is not constitutively active, but must be phosphorylated and unfolded to bind to cytoplasmic proteins. Over aa 438-562, human EZRIN shares 96% aa identity with mouse EZRIN.

PRODUCT SPECIFIC NOTICES

* Contains <0.1% Sodium Azide, which is not hazardous at this concentration according to GHS classifications. Refer to SDS for additional information and handling instructions.