Human Neutrophil Elastase/ELA2 Antibody

Monoclonal Mouse IgG_{2A} Clone # 950302

Catalog Number: MAB91672

DESCRIPTION

Species Reactivity	Human
Specificity	Detects human Neutrophil Elastase/ELA2 in direct ELISAs.
Source	Monoclonal Mouse IgG_{2A} Clone # 950302
Purification	Protein A or G purified from hybridoma culture supernatant
Immunogen	Chinese hamster ovary cell line CHO-derived recombinant human Neutrophil Elastase/ELA2 Met1-Asn252 Accession # P08246
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.

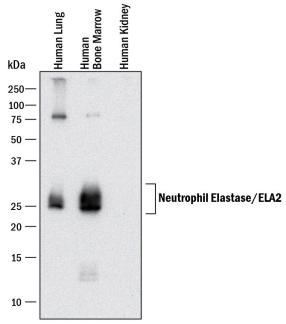
APPLICATIONS

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	1 µg/mL	See Below
Immunocytochemistry	8-25 µg/mL	See Below
Immunohistochemistry	5-25 µg/mL	See Below
ELISA	This antibody functions as an ELISA detection antibody when paired with Mouse Anti-Human Neutrophil Elastase/ELA2 Monoclonal Antibody (Catalog # MAB91673). *This product is intended for assay development on various assay platforms requiring antibody pairs. We recommend the Human Neutrophil Elastase/ELA2 DuoSet ELISA Kit (Catalog # DY9167-05) for convenient development of a sandwich ELISA.*	

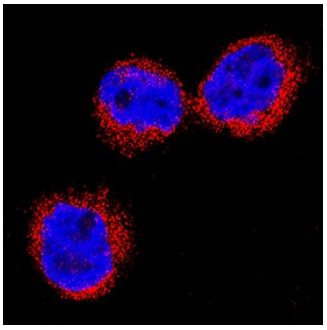
DATA

Western Blot



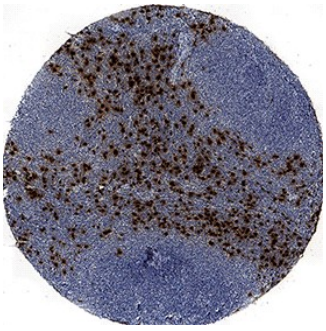
Detection of Human Neutrophil Elastase/ELA2 by Western Blot. Western blot shows lysates of human lung tissue, human bone marrow, and human kidney tissue (negative control). PVDF membrane was probed with 1 µg/mL of Mouse Anti-Human Neutrophil Elastase/ELA2 Monoclonal Antibody (Catalog # MAB91672) followed by HRP-conjugated Anti-Mouse IgG Secondary Antibody (Catalog # HAF018). Specific bands were detected for Neutrophil Elastase/ELA2 at approximately 25-30 kDa (as indicated). This experiment was conducted under reducing conditions and using Immunoblot Buffer Group 1.

Immunocytochemistry



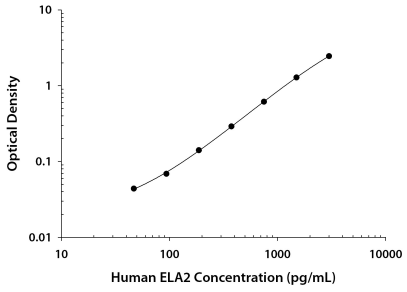
Neutrophil Elastase/ELA2 in THP-1 Human Cell Line. Neutrophil Elastase/ELA2 was detected in immersion fixed THP-1 human acute monocytic leukemia cell line using Mouse Anti-Human Neutrophil Elastase/ELA2 Monoclonal Antibody (Catalog # MAB91672) at 8 µg/mL for 3 hours at room temperature. Cells were stained using the NorthernLights™ 557-conjugated Anti-Mouse IgG Secondary Antibody (red; Catalog # NL007) and counterstained with DAPI (blue). Specific staining was localized to cytoplasm. View our protocol for Fluorescent ICC Staining of Non-adherent Cells.

Immunohistochemistry



Neutrophil Elastase/ELA2 in Human Lymphoma. Neutrophil Elastase/ELA2 was detected in immersion fixed paraffin-embedded sections of human lymphoma using Mouse Anti-Human Neutrophil Elastase/ELA2 Monoclonal Antibody (Catalog # MAB91672) at 5 µg/mL overnight at 4 °C. Tissue was stained using the Anti-Mouse IgG VisUCyte™ HRP Polymer Antibody (brown; Catalog # VC001) and counterstained with hematoxylin (blue). Specific staining was localized to cytoplasm of cancer cells. View our protocol for IHC Staining with VisUCyte HRP Polymer Detection Reagents.

ELISA



Human Neutrophil Elastase/ELA2 ELISA Standard Curve. Recombinant Human Neutrophil Elastase/ELA2 protein was serially diluted 2-fold and captured by Mouse Anti-Human Neutrophil Elastase/ELA2 Monoclonal Antibody (Catalog # MAB91673) coated on a Clear Polystyrene Microplate (Catalog # DY990). Mouse Anti-Human Neutrophil Elastase/ELA2 Monoclonal Antibody (Catalog # MAB91672) was biotinylated and incubated with the protein captured on the plate. Detection of the standard curve was achieved by incubating Streptavidin-HRP (Catalog # DY998) followed by Substrate Solution (Catalog # DY999) and stopping the enzymatic reaction with Stop Solution (Catalog # DY994).

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Human Neutrophil Elastase/ELA2 Antibody

Monoclonal Mouse IgG_{2A} Clone # 950302

Catalog Number: MAB91672

PREPARATION AND STORAGE

Reconstitution	Reconstitute at 0.5 mg/mL in sterile PBS.
Shipping	The product is shipped at ambient temperature. 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.** ● 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.

BACKGROUND

Neutrophil Elastase (ELA2, ELANE), also known as HNE, is a chymotrypsin family serine protease that plays a key role in pathogen clearance (1-3). It is expressed by promyelocytes and stored in the intracellular azurophilic granules of polymorphonuclear leukocytes (PMN) (4). These granules fuse with phagosomes, enabling Neutrophil Elastase to participate in the digestion and killing of endocytosed microbes. The enzyme is released by activated neutrophils at sites of inflammation, and it can remain associated with the cell surface or function as a component of neutrophil extracellular nets (NETs) which trap and kill microbial pathogens (5, 6). It also can degrade multiple extracellular matrix proteins including Elastin and Fibronectin (5). In the lung, this activity contributes to pathology in emphysema, cystic fibrosis, and adult respiratory distress syndrome (ARDS) (1). Neutrophil Elastase can be inhibited by Serpin A1/alpha 1-Antitrypsin, SLPI, Serpin B1, and Trappin-2/Elafin (7-11). Its activity in the lung is increased by exposure to tobacco smoke which inactivates Serpin A1 through methionine oxidation (12). Mature human Neutrophil Elastase shares 73% amino acid sequence identity with mouse and rat Neutrophil Elastase (13, 14). Multiple mutations in the human ELANE gene are causative of severe congenital and cyclic neutropenias (15).

References:

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