

Anti-Mast Cell Tryptase Antibody [SC68-07]

ET1610-64



Product Type:	Recombinant Rabbit monoclonal IgG, primary antibodies
Species reactivity:	Human, Mouse
Applications:	WB, IHC-P, IP, mIHC
Molecular Wt:	Predicted band size: 30 kDa
Clone number:	SC68-07

Description:	Mast cells are connective tissue cells derived from blood-forming tissues that line arterial walls and secrete substances, which mediate inflammatory and immune responses. Mast cell chymase, known as CMA1, is a major secreted serine protease that is involved in vasoactive peptide generation, extracellular matrix degradation and regulation of gland secretion. The human chymase gene, which maps to human chromosome 14q11.2, encodes a preproenzyme with a 19-amino acid signal peptide, an acidic 2-amino acid propeptide and a 226-amino acid catalytic domain. Tryptases comprise a family of trypsin-like serine proteases that are enzymatically active as heparin-stabilized tetramers. There are four functional genes for tryptase: α I, β I, β II and γ I, which map to human chromosome 16p13.3, with β tryptases representing the main isoenzymes expressed in mast cells. Mast cell proteases are a family of rodent protein homologs to human tryptases that are specifically expressed in mast cells and may serve as highly specific markers in the analysis of mast cell heterogeneity, differentiation and function.
Immunogen:	Recombinant protein within Human Mast Cell Tryptase aa 10-275 / 275.
Positive control:	Human lung tissue lysates, mouse skin tissue lysates, human lung tissue, human tonsil tissue, human small intestine tissue, human prostate carcinoma tissue.
Subcellular location:	Secreted.
Database links:	SwissProt: Q15661 Human Q02844 Mouse
Recommended Dilutions:	
WB	1:1,000
IHC-P	1:5,000-1:10,000
IP	Use at an assay dependent concentration.
mIHC	1:3,000-1:5,000
Storage Buffer:	1*TBS (pH7.4), 0.05% BSA, 40% Glycerol. Preservative: 0.05% Sodium Azide.
Storage Instruction:	Shipped at 4°C. Store at +4°C short term (1-2 weeks). It is recommended to aliquot into single-use upon delivery. Store at -20°C long term.
Purity:	Protein A affinity purified.

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Images

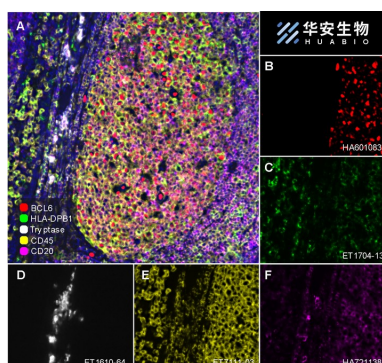


Fig1: Fluorescence multiplex immunohistochemical analysis of Human tonsil (Formalin/PFA-fixed paraffin-embedded sections). Panel A: the merged image of anti-BCL6 (HA601083, Red), anti-HLA-DPB1 (ET1704-13, Green), anti-Tryptase (ET1610-64, White), anti-CD20 (HA721138, Magenta) and anti-CD45 (ET7111-03, Yellow) on tonsil. HRP Conjugated UltraPolymer Goat Polyclonal Antibody HA1119/HA1120 was used as a secondary antibody. The immunostaining was performed with the Sequential Immuno-staining Kit (IRISKit™MH010101, www.luminiris.cn). The section was incubated in five rounds of staining: in the order of HA601083 (1/200 dilution), ET1704-13 (1/2,000 dilution), ET1610-64 (1/5,000 dilution), HA721138 (1/2,000 dilution) and ET7111-03 (1/500 dilution) for 20 mins at room temperature. Each round was followed by a separate fluorescent tyramide signal amplification system. Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 30 mins at 95°C. DAPI (blue) was used as a nuclear counter stain. Image acquisition was performed with Olympus VS200 Slide Scanner.

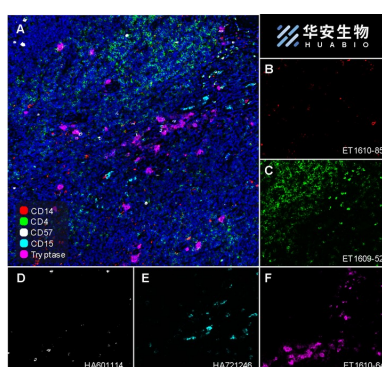


Fig2: Fluorescence multiplex immunohistochemical analysis of Human tonsil (Formalin/PFA-fixed paraffin-embedded sections). Panel A: the merged image of anti-CD14 (ET1610-85, Red), anti-CD4 (ET1609-52, Green), anti-CD57 (HA601114, White), anti-CD15 (HA721246, Cyan) and anti-Tryptase (ET1610-64, Magenta) on tonsil. Panel B: anti-CD14 stained on monocytes. Panel C: anti-CD4 stained on helper T cells and Treg cells. Panel D: anti-CD57 stained on NK cells and T cells. Panel E: CD15 stained on granulocytes and monocytes. Panel F: anti-Tryptase stained on Mast cells. HRP Conjugated UltraPolymer Goat Polyclonal Antibody HA1119/HA1120 was used as a secondary antibody. The immunostaining was performed with the Sequential Immuno-staining Kit (IRISKit™MH010101, www.luminiris.cn). The section was incubated in five rounds of staining: in the order of ET1610-85 (1/800 dilution), ET1609-52 (1/800 dilution), HA601114 (1/1,000 dilution), HA721246 (1/500 dilution), and ET1610-64 (1/3,000 dilution) for 20 mins at room temperature. Each round was followed by a separate fluorescent tyramide signal amplification system. Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 30 mins at 95°C. DAPI (blue) was used as a nuclear counter stain. Image acquisition was performed with Olympus VS200 Slide Scanner.

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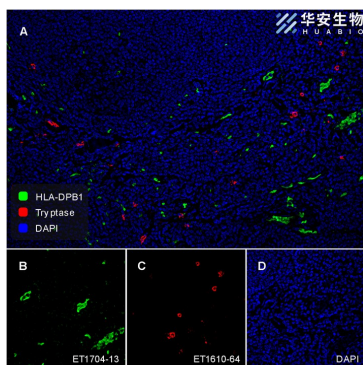
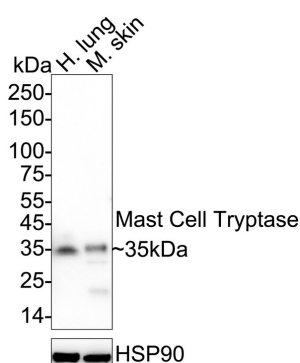


Fig3: Fluorescence multiplex immunohistochemical analysis of human tonsil (Formalin/PFA-fixed paraffin-embedded sections). Panel A: the merged image of anti-HLA-DBP1 (ET1704-13, Green) and anti-Tryptase (ET1610-64, Red) on tonsil. HRP Conjugated UltraPolymer Goat Polyclonal Antibody HA1119/HA1120 was used as a secondary antibody. The immunostaining was performed with the Sequential Immunostaining Kit (IRISKit™MH010101, www.luminiris.cn). The section was incubated in two rounds of staining: in the order of ET1704-13 (1/2,000 dilution) and ET1610-64 (1/5,000 dilution) for 20 mins at room temperature. Each round was followed by a separate fluorescent tyramide signal amplification system. Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 30 mins at 95°C. DAPI (blue) was used as a nuclear counter stain. Image acquisition was performed with Zeiss Observer 7 Inverted Fluorescence Microscope.

Fig4: Western blot analysis of Mast Cell Tryptase on different lysates with Rabbit anti-Mast Cell Tryptase antibody (ET1610-64) at 1/1,000 dilution.



Lane 1: Human lung tissue lysate (40 µg/Lane)

Lane 2: Mouse skin tissue lysate (40 µg/Lane)

Predicted band size: 30 kDa

Observed band size: 35 kDa

Exposure time: 59 seconds; ECL: K1801;

4-20% SDS-PAGE gel.

Proteins were transferred to a PVDF membrane and blocked with 5% NFDM/TBST for 1 hour at room temperature. The primary antibody (ET1610-64) at 1/1,000 dilution was used in 5% NFDM/TBST at 4°C overnight. Goat Anti-Rabbit IgG - HRP Secondary Antibody (HA1001) at 1/50,000 dilution was used for 1 hour at room temperature.

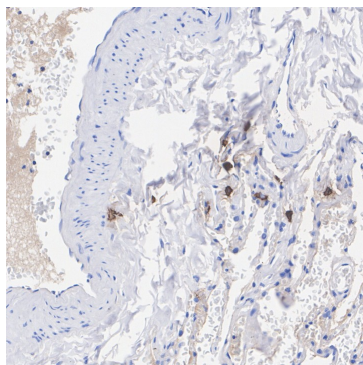


Fig5: Immunohistochemical analysis of paraffin-embedded human lung tissue with Rabbit anti-Mast Cell Tryptase antibody (ET1610-64) at 1/10,000 dilution.

The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 20 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH₂O and PBS, and then probed with the primary antibody (ET1610-64) at 1/10,000 dilution for 1 hour at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.

Note: All products are "FOR RESEARCH USE ONLY AND ARE NOT INTENDED FOR DIAGNOSTIC OR THERAPEUTIC USE".

Background References

1. De Martin S et al. Expression and distribution of the adrenomedullin system in newborn human thymus. PLoS One 9:e97592 (2014).
2. Edmunds MC et al. Paradoxical effects of heme arginate on survival of myocutaneous flaps. Am J Physiol Regul Integr Comp Physiol 306:R10-22 (2014).

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