

## DESCRIPTION

<b>Species Reactivity</b>	Human
<b>Specificity</b>	Detects both the pro and mature forms of human Legumain/Asparaginyl Endopeptidase in ELISAs and Western blots.
<b>Source</b>	Monoclonal Mouse IgG <sub>1</sub> Clone # 312109
<b>Purification</b>	Protein A or G purified from hybridoma culture supernatant
<b>Immunogen</b>	NS0-derived recombinant human Legumain Val18-Tyr433 Accession # Q99538
<b>Formulation</b>	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
<b>Western Blot</b>	1 µg/mL	Recombinant Human Legumain (Catalog # 2199-CY)
<b>Human Legumain Sandwich Immunoassay</b>		<b>Reagent</b>
<b>ELISA Capture</b>	2-8 µg/mL	Human Legumain/Asparaginyl Endopeptidase Antibody (Catalog # MAB21992)
<b>ELISA Detection</b>	0.1-0.4 µg/mL	Human Legumain/Asparaginyl Endopeptidase Biotinylated Antibody (Catalog # BAF2199)
<b>Standard</b>		Recombinant Human Legumain/Asparaginyl Endopeptidase (Catalog # 2199-CY)

## PREPARATION AND STORAGE

<b>Reconstitution</b>	Reconstitute at 0.5 mg/mL in sterile PBS.
<b>Shipping</b>	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
<b>Stability &amp; Storage</b>	<b>Use a manual defrost freezer and avoid repeated freeze-thaw cycles.</b> <ul style="list-style-type: none"> <li>• 12 months from date of receipt, -20 to -70 °C as supplied.</li> <li>• 1 month, 2 to 8 °C under sterile conditions after reconstitution.</li> <li>• 6 months, -20 to -70 °C under sterile conditions after reconstitution.</li> </ul>

## BACKGROUND

Legumain is a lysosomal cysteine protease whose activity is found in several tissues tested (1, 2). Legumain plays a pivotal role in the endosomal/lysosomal degradation system because the Legumain deficiency causes the accumulation of pro cathepsins B, H and L, another group of lysosomal cysteine proteases (3). Over-expression of Legumain in tumors is significant for invasion/metastasis (4). Also known as Asparaginyl Endopeptidase, it specifically cleaves peptide bonds with Asn at the P1 position. Nevertheless, it also cleaves peptide bonds with Asp at the P1 position. Auto-activation of pro Legumain involves both types of the cleavage, which result in the removal of the pro peptides in both C- and N-termini (5). In addition, Legumain activates pro MMP-2 and processes bacterial antigens for MHC class II presentation and pro thymosin α to thymosin α<sub>1</sub> and thymosin α<sub>11</sub>, two acidic peptides with immunoregulatory properties (6-8). Human Legumain is synthesized as a 433 amino acid precursor with a signal peptide (residues 1-17). The pro enzyme (residues 18-433) was expressed with an N-terminal His tag. This activity of Legumain can be inhibited by rhCystatins C and E/M and recombinant mouse Cystatin C (R&D Systems, Catalog # 1196-PI, 1286-PI and 1238-PI, respectively).

## References:

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3. Shirahama-Noda, K. *et al.* (2003) *J. Biol. Chem.* **278**:33194.
4. Liu, C. *et al.* (2003) *Cancer Res.* **63**: 2957.
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6. Chen, J.M. *et al.* (2001) *Biol. Chem.* **382**:777.
7. Schwarz, G. *et al.* (2002) *Biol. Chem.* **383**:1813.
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