



Catalog #	Aliquot Size
L585-31HP-20	20 µg
L585-31HP-500	0.5 mg
L585-31HP-1000	1 mg

Lys-c (lysyl-endopeptidase), Active

Recombinant *A. lyticus* protein expressed in *E. coli* cells

Catalog # L585-31HP

Lot # U4783-11

Product Description

Recombinant *Achromobacter lyticus* Lys-c (lysyl-endopeptidase) (206-473aa) was expressed in *E. coli* cells using a C-terminal His tag. The enzyme commission number is EC 3.4.21.50.

Alternative Name(s)

API, Protease I, Lysyl endopeptidase

Formulation

Recombinant protein lyophilized in the presence of 50mM Tris-HCl, pH 8.0, and 3% mannitol.

Reconstitution Protocol

Add distilled H₂O to a final concentration of 1 mg/ml.

Storage and Stability

Store product at -20°C. For most favorable performance, avoid repeated handling and multiple freeze/thaw cycles once the protein has been resolubilized in sterile water.

Digestion Conditions

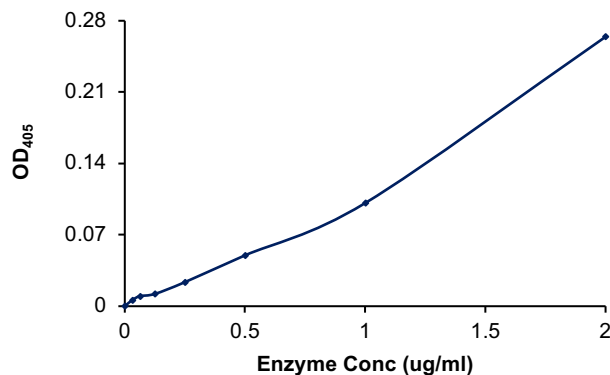
Catalytic pH range: 9.0 ~ 9.5

Catalytic temperature range: 30 ~ 37°C

Scientific Background

Lysyl-endopeptidase (Lys-c) was isolated from the Gram-negative soil bacterium *Achromobacter lyticus* by Masaki et al. The protein hydrolyzes amide and peptide ester bonds at the carboxylic side of lysine and S-aminoethylcysteine residues making it an important tool for enzymatic protein sequencing and Lys-X compound synthesis. The enzyme functions optimally between 30 – 37°C and suffers from degradation when subjected to temperatures above 50°C. Lysyl-endopeptidase retains complete activity after incubation in 4M urea or in 0.1% SDS solution for up to 6 hours at 30°C. Conversely, the enzyme is inhibited by DFP, PMSF, TLCK. Lysyl-endopeptidase is involved during the production of recombinant insulin and in proteomics research.

Specific Activity



The activity of Lys-c is **1.11 U/mg** as per the activity assay protocol.

Lys-c (lysyl-endopeptidase), Active

Recombinant *A. lyticus* protein expressed in *E. coli* cells

Catalog #	L585-31HP
Specific Activity	1.11 U/mg
Lot #	U4783-11
Stability	1yr at -20°C from date of shipment
Storage & Shipping	Store product at -20°C. For optimal storage, aliquot reconstituted product into smaller quantities and store at recommended temperature. For most favorable performance, avoid repeated handling and multiple freeze/thaw cycles. Product shipped at ambient temperature.

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Activity Assay Protocol

Reaction Components

Active Lys-C (Catalog #: L585-31HP)

Active Lys-C diluted with Lys-C Reaction Buffer and assayed as outlined in sample activity plot. (Note: these are suggested working dilutions and it is recommended that the researcher perform a serial dilution of the enzyme for optimal results).

Lys-C Reaction Buffer

Buffer components: 180 mM Tris-HCl pH 9.0

Substrate

Ac-Lys-pNA dissolved in Lys-C Reaction Buffer at 30 mM stock solution.

Assay Protocol

The Lys-C endopeptidase assay is performed by monitoring the absorbance of pNA, the product from hydrolysis of the chromogenic substrate Ac-Lys-pNA in the presence of the enzyme.

Step 1. Thaw the Active Lys-C on ice, equilibrate buffer and substrate to room temperature.

Step 2. Prepare the following working solutions with Lys-C Reaction Buffer:

- o 2X desired concentrations of Active Lys-C
- o 0.5 mM Ac-Lys-pNA

Step 3. In a black full-area 96-well plate with clear bottom, add the following reaction components to bring the reaction volume to 100 μ L:

Component 1. 50 μ L of Lys-C working solutions

Component 2. 50 μ L of 0.5 mM Ac-Lys-pNA

Note: A blank control can be set up as outlined in step 3 by replacing the enzyme working solution with an equal volume of the reaction buffer.

Step 4. Mix the plate for 1 minute on a tabletop orbital shaker and incubate at 24°C for 30 minutes.

Step 5. At the end of the incubation period, briefly mix the plate and read absorbance at 405 nm.

Step 6. Subtract the background absorbance (of the blank control) from all sample readings and determine the specific activity using the formula below:

Enzyme Specific Activity (SA) (μ mol/min/mg)

$$= \frac{320 (\mu\text{L cm}^{-1}) \times A_{\text{corr}}}{\epsilon (\text{L mol}^{-1}\text{cm}^{-1}) \times \text{Reaction Time (min)} \times \text{Enzyme Amount (mg)}}$$

A_{corr} - background-corrected absorbance

ϵ - molar extinction coefficient of pNA (9500 L mol⁻¹ cm⁻¹)

Note: the above formula is only valid for standard 96-well plate.

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SAFETY DATA SHEET

Article 1 - Product Identification

Product Name: Lys-c (Lysyl-Endopeptidase), Active

Catalog # L585-31HP

This product is sold only for research use by qualified laboratory personnel, and is not to be used as a drug, medical device, food additive, cosmetic, nor household chemical. It is not to be used in diagnostic, therapeutic, consumer, agricultural, nor pesticidal applications.

Manufacturer's Name: SignalChem Biotech Inc.
 Street Address: 110-13120 Vanier Place
 City, Prov. Postal Code: Richmond, BC, V6V 2J2
 Fax: 604-232-4601
 EMERGENCY PHONE: 604-232-4600

Article 2 - Hazards Identification

Emergency Overview: The product contains no substances which at their given concentration, are considered to be hazardous to health.
 WHMIS Classification: Not WHMIS controlled
 GHS Classification: Not a dangerous substance according to GHS.

Potential health effects:

Inhalation: H333 – May be harmful if swallowed
 Skin: H313 – May be harmful in contact with skin
 Eyes: May cause eye irritation
 Ingestion: H303 – May be harmful if swallowed

Precautionary Statements:

Inhalation: P304+P312 – If inhaled, remove to fresh air. Consult a physician if necessary.
 Skin: P302+P312 – If on skin, wash with soap and plenty of water. Consult a physician if necessary
 Eyes: Wash with plenty of water as a precaution.
 Ingestion: P301+P312 – Never give anything by mouth to an unconscious person. Rinse mouth with water. Consult a physician if necessary.

Article 3 - Composition/Information on Ingredients

Common Name	Chemical Name/Formula	CAS-No.	Concentration
Protein	-	NA	≤100%

Article 4 - First-aid Measures

- **General information:** Consult a physician by providing the SDS.
- **After inhalation:** Breath in fresh air. If cannot breath, give artificial respiration and consult a physician.
- **After skin contact:** Immediately wash with soap and plenty of water and rinse thoroughly. Consult a physician.
- **After eye contact:** Rinse opened eyes with plenty of water for at least 15 minutes. Consult a physician.
- **After swallowing:** Rinse the mouth with plenty of water and consult a physician.

Article 5 - Fire-fighting Measures

- **Suitable extinguishing media:** Use water spray, extinguishing powder, carbon dioxide, or other appropriate measure that is suitable to the environment.
- **Specific hazards arising from the substance or mixture:** None known.
- **Special protective equipment and precautions for fire-fighters:** Self-contained breathing apparatus if necessary.

Article 6 – Accidental Release Measures

- **Personal precautions, protective equipment and emergency procedures:** Apply standard laboratory practices and personal protective equipment. Avoid breathing vapors, mist, or gas. Ensure adequate ventilation.
- **Environmental precautions:** Do not allow to enter drains.
- **Methods and materials for containment and cleaning up:** Prevent further leakage or spillage if safe to do so. Use personal protective equipment as required. Cover powder spill with plastic sheet or tarp to minimize spreading and keep powder dry. Avoid creating dust. Clean contaminated surface thoroughly.

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Article 7 - Handling and Storage

- **Precautions for safe handling:** Wear chemical safety goggles and compatible chemical-resistant gloves. Avoid inhalation, contact with eyes, skin or clothing.
- **Conditions for safe storage:** Keep container tightly closed in a dry and well-ventilated place. Recommended storage temperature: -20 °C.

Article 8 - Exposure Controls/Personal Protection

- **Appropriate engineering controls:**
Apply adequate ventilation including mechanical exhaust or laboratory fume hood. Follow standard laboratory practices.
- **Individual protection measures:**
 - Respiratory protection:**
Ensure adequate ventilation, especially in confined areas.
 - Skin/body protection:**
Wear protective gloves and clothing. Use proper glove removal technique to avoid skin contact. Discard gloves after use by following the applicable laboratory regulations. Wash and dry hands.
 - Eye/face protection:**
Safety glasses with side-shields (or goggles) approved under appropriate government standards.
 - General hygiene considerations:**
Handle in accordance with good industrial hygiene and safety practice.

Article 9 – Physical and Chemical Properties

Appearance: Solid.	Danger of explosion: No data available.
Odour/Odour Threshold: Not determined.	Explosion limits: No data available.
pH: No data available.	Decomposition temperature: No data available.
Melting point/freezing point: Not determined.	Vapor pressure at 20 °C: No data available.
Boiling point/Boiling range: No data available .	Density: No data available.
Flash point: No data available .	Relative density: No data available.
Flammability (solid, gaseous): Not determined.	Vapor density: No data available.
Ignition temperature: No data available.	Evaporation rate: No data available.
Auto-igniting: No data available .	Solubility in / Miscibility with Water: Fully miscible.

Article 10 - Stability and Reactivity

- **Reactivity:** Stable under recommended transport and storage conditions.
- **Chemical stability:** Stable under recommended transport and storage conditions.
- **Possible hazardous reactions:** No dangerous reactions known.
- **Conditions to avoid:** No data available.
- **Incompatible materials:** No data available.
- **Hazardous decomposition products:** No data available.

Article 11 - Toxicological Information

- **Acute toxicity:** Product does not present an acute toxicity hazard based on known or supplied information.
- **LD/LC50:** Not available.
- **Skin corrosion/irritation:** Not available.
- **Serious eye damage/eye irritation:** Not available.
- **Respiratory or skin sensitization:** Not available.
- **Germ cell mutagenicity:** Not available.
- **Carcinogenicity:** No components are listed in IARC, or NTP, or OSHA, or ACGIH.
- **Reproductive toxicity:** Not available.
- **Teratogenicity:** Not available.
- **Specific target organ toxicity - single exposure/ - repeated exposure (GHS):** Not available.
- **Aspiration hazard:** Not available.
- **Potential health effects:**
 - Inhalation:** May be harmful if inhaled. May cause respiratory tract irritation.
 - Ingestion:** May be harmful if swallowed.
 - Skin:** May be harmful if absorbed through skin. May cause skin irritation.
 - Eyes:** May cause eye irritation.
- **Signs and Symptoms of Exposure:** Not available.
- **Synergistic effects:** Not available.

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Article 12 - Ecological Information

- **Eco-toxicity:** Not applicable.
- **Biodegradability:** Not applicable.
- **Bio-accumulative potential:** Not applicable.
- **Mobility in soil:** Not applicable.
- **PBT and vPvB assessment:** Not applicable.
- **Other adverse effects:** Not applicable.

Article 13 - Disposal Considerations

- **Disposal methods:** In accordance to applicable national, regional, or local laws and regulations. For additional handling information and protection of employees please refer to Article 7 and 8.
- **Contaminated packaging:** Disposal should be made in accordance to official regulations. Use water or cleansing agents to clean the area.

Article 14 - Transport Information

- **DOT:** Not dangerous goods.
- **IMDG:** Not dangerous goods.
- **IATA:** Not dangerous goods.

Article 15 - Regulatory Information

Safety, health, and environmental regulations/legislation specific for the substance or mixture.

Canadian substance listings:

- **WHMIS Classification:** Non-hazardous.
- **GHS label elements:** Not applicable.
- **Signal word:** Not applicable.
- **Hazard statements:** Not applicable.

Article 16 - Other Information

The above information is believed to be correct but does not purport to be all-inclusive and shall be used only as a guide. SignalChem shall not be held liable for any damage resulting from handling or from contact with the above product. See the Technical Specification, Packing Slip, Invoice, and Product Catalog for additional terms and conditions of sale.

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