

**DESCRIPTION**

**Source** Mouse myeloma cell line, NS0-derived human Aminopeptidase LRAP/ERAP2 protein  
Ala56-Thr960, with an N-terminal 6-His tag  
Accession # Q6P179

**N-terminal Sequence Analysis** His

**Predicted Molecular Mass** 105 kDa

**SPECIFICATIONS**

**SDS-PAGE** 122 kDa, reducing conditions

**Activity** Measured by its ability to cleave the fluorogenic peptide substrate, Arg-7-amido-4-methylcoumarin (Arg-AMC).  
The specific activity is >50 pmol/min/μg, as measured under the described conditions.

**Endotoxin Level** <1.0 EU per 1 μg of the protein by the LAL method.

**Purity** >90%, by SDS-PAGE visualized with Silver Staining and quantitative densitometry by Coomassie® Blue Staining.

**Formulation** Supplied as a 0.2 μm filtered solution in Tris, NaCl and Glycerol. See Certificate of Analysis for details.

**Activity Assay Protocol**

- Materials**
- Assay Buffer: 100 mM MES, pH 6.5
  - Recombinant Human Aminopeptidase LRAP/ERAP2 (Catalog # 3830-ZN)
  - Substrate: H-Arg-AMC (Bachem, Catalog # I-1050)
  - F16 Black Maxisorp Plate (Nunc, Catalog # 475515)
  - Fluorescent Plate Reader (Model: SpectraMax Gemini EM by Molecular Devices) or equivalent

- Assay**
1. Dilute rhLRAP to 5 μg/mL in Assay Buffer.
  2. Dilute Substrate to 100 μM in Assay Buffer.
  3. Load 50 μL of 5 μg/mL rhLRAP into a plate, and start the reaction by adding 50 μL of 100 μM Substrate. Include a Substrate Blank containing 50 μL of Assay Buffer and 50 μL of Substrate.
  4. Read at excitation and emission wavelengths of 380 nm and 460 nm (top read), respectively, in kinetic mode for 5 minutes.
  5. Calculate specific activity:

$$\text{Specific Activity (pmol/min/}\mu\text{g)} = \frac{\text{Adjusted } V_{\text{max}}^* \text{ (RFU/min)} \times \text{Conversion Factor}^{**} \text{ (pmol/RFU)}}{\text{amount of enzyme (}\mu\text{g)}}$$

\*Adjusted for Substrate Blank

\*\*Derived using calibration standard 7-Amino-4-Methyl Coumarin (Sigma-Aldrich, Catalog # A9891).

- Final Assay Conditions**
- Per Well:
- rhLRAP: 0.25 μg
  - Substrate: 50 μM

**PREPARATION AND STORAGE**

**Shipping** The product is shipped with polar packs. Upon receipt, store it immediately at the temperature recommended below.

- Stability & Storage** Use a manual defrost freezer and avoid repeated freeze-thaw cycles.
- 6 months from date of receipt, -20 to -70 °C as supplied.
  - 3 months, -20 to -70 °C under sterile conditions after opening.

**BACKGROUND**

Leukocyte-derived arginine aminopeptidase (LRAP) and endoplasmic reticulum-aminopeptidase 2 (ERAP2) are the two names given to the protein encoded by the LRAP gene (1, 2). Induced by interferon-γ, LRAP is able to trim various MHC class I antigenic peptide precursors. It belongs to the oxytocinase subfamily of M1 aminopeptidases, which also includes aminopeptidases PILS/ARTS1/ERAP1 and LNPEP/PLAP (1, 3). In addition to antigen presentation, the members of this subfamily are also important in maintenance of pregnancy, memory retention, and blood pressure regulation (3).

**References:**

1. Tanioka, T. *et al.* (2003) J. Biol. Chem. **278**:32275.
2. Tanioka, T. *et al.* (2005) FEBS J. **272**:916.
3. Tsujimoto, M. and A. Hattori (2005) Biochim. Biophys. Acta **1751**:9.