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**MATERIAL DATA SHEET**

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**Recombinant Human HA GATE-16 AMC****Cat. # UL-445**

Golgi-associated ATPase Enhancer of 16 kDa (GATE-16), also known as GABARAPL2, is a 117 amino acid (aa) polypeptide and a member of the Autophagy-related 8 (ATG8) family of proteins (1). GATE-16 has 100% aa sequence identity with its mouse and rat orthologs, and is orthologous to the yeast ATG8. ATG8 family members show structural similarity with Ubiquitin, but lack aa sequence similarity. GATE-16 is best known for its role in autophagy (2,3). GATE-16 covalently attaches to phosphatidylethanolamine (PE) the phagophore (autophagosome precursor) membrane using a Ubiquitin-like conjugation system that includes Ubiquitin-activating (E1)-, Ubiquitin-conjugating (E2)-, and Ubiquitin Ligase (E3)-like enzymes. Here it is involved in the later stages of autophagosome formation (4,5). It may also be involved in cargo recruitment to autophagosomes (1).

This is a fluorogenic substrate for ATG8-specific C-terminal hydrolases such as ATG4B (Catalog # E-400) based on the C-terminal derivatization of ATG8 with 7-amido-4-methylcoumarin (AMC). ATG8-AMC is useful for studying such enzyme activities when detection sensitivity or continuous monitoring of activity is essential. NOTE: this protein has an N-terminal HA tag.

**Product Information**

<b>Quantity:</b>	25 µg
<b>MW:</b>	14 kDa
<b>Source:</b>	<i>E. coli</i> -derived Contains an N-terminal HA (YPYDVPDYA) tag Accession # P60520
<b>Stock:</b>	X mg/ml (X µM) in 50 mM MOPS pH 6.5, 100 mM NaCl
<b>Purity:</b>	>90%, by SDS-PAGE under reducing conditions and visualized by silver stain.

## Use & Storage

**Use:** Recombinant Human HA-GATE-16 AMC is a fluorogenic substrate for some ATG-specific isopeptidases. Release of AMC fluorescence can be monitored with an excitation wavelength of 345 nM and an emission wavelength of 445 nM. Reaction conditions will need to be optimized for each specific application. We recommend an initial Recombinant Human HA-GATE-16 AMC concentration of 0.1-1  $\mu$ M.

**Storage:** **Protect from light. Use a manual defrost freezer and avoid repeated freeze-thaw cycles.**

- 12 months from date of receipt, -70 °C as supplied.
- 3 months, -70 °C under sterile conditions after opening.

## Literature

### References:

1. Shpilka, T. *et al.* (2011) *Genome Biol.* **12**:226.
2. Wang, H. *et al.* (1999) *Nature* **397**:69.
3. Leil, T.A. *et al.* (2004) *J. Neurosci.* **24**:11429.
4. Weidberg, H. *et al.* (2010) *EMBO J.* **29**:1792.
5. Weidberg, H. *et al.* (2011) *Ann. Rev. Biochem.* **80**:125.

***For research use only. Not for use in humans.***