

Ubiquitin-AMC

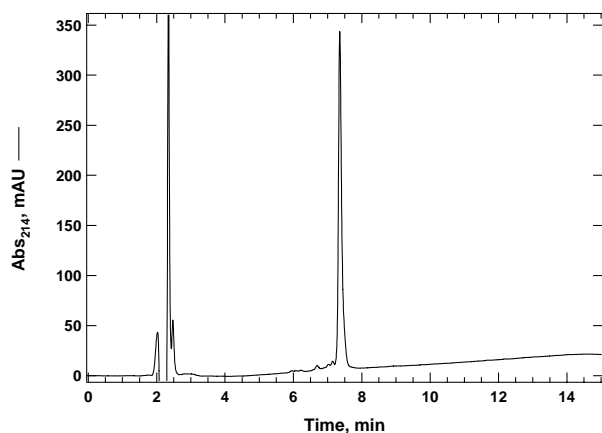
Cat. # SI220

Background: Ubiquitin-AMC is prepared via the conjugation of 7-amido-4-methylcoumarin (AMC) to the C-terminal Gly of mature Ubiquitin. This conjugation quenches the intrinsic fluorescence of AMC. Upon incubation with a protease recognizing Ubiquitin, such as USP2 or UCHL3, AMC is released and the increase in fluorescence at 460 nm (Exc. At 380 nm) can be measured. The enhanced purity of Ubiquitin-AMC compared to that of other commercial vendors leads to a greater signal to noise ratio. This protein contains no extraneous tags.

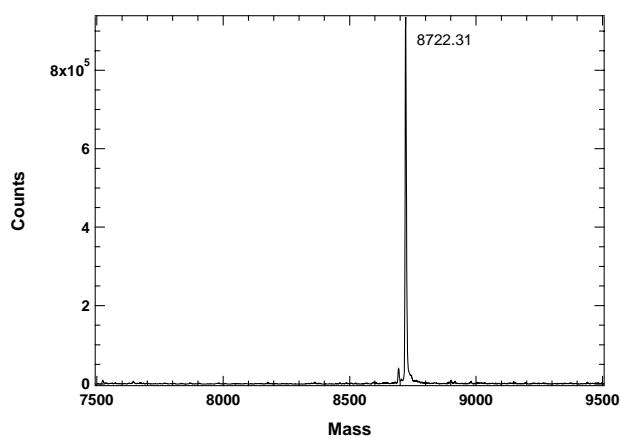
Application: Used for determining the activity and specificity of ubiquitin and ubiquitin-like hydrolases. High throughput screening for modulators of deubiquitylase activity.

Product Information

Purity:	≥ 95% by HPLC
Molecular Weight:	8722 Da by MS
Physical State:	Lyophilized powder
Quantity:	50 µg
Solubility:	≥ 1 mg/mL in DMSO or 0.05% HOAc
Storage:	Dry, -20° C. Solution, -80° C. Avoid repeated freeze/thaw cycles



RP-HPLC Trace, C18 column



Deconvoluted mass spectrum

References

- 1) Shanmugham A and Ova H. *DUBs and disease: activity assays for inhibitor development*. Curr Opin Drug Discov Devel. 2008;11(5):688-96.
- 2) Mason, D.E., Ek, J., Peters, E.C. and Harris J.L. *Substrate profiling of deubiquitin hydrolases with a positional scanning library and mass spectrometry*. Biochemistry 2004, 43: p6535-44
- 3) Dang, L. C., F. D. Melandri, and R. L. Stein. *Kinetic and mechanistic studies on the hydrolysis of ubiquitin C-terminal 7-amido-4-methylcoumarin by deubiquitinating enzymes*. Biochemistry 1998 37: p1868-1879.
- 4) Stein RL, Chen Z, Melandri F. *Kinetic studies of isopeptidase T: modulation of peptidase activity by ubiquitin*. Biochemistry. 1995 34(39):p12616-23.

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