UbiQ
targeting the ubiquitin system

Ub-AMC *(human sequence, synthetic)*

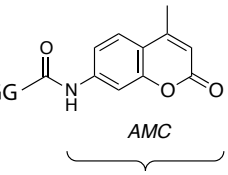
UbiQ code	:	UbiQ-001
Batch #	:	B01012013-001
Amount	:	100 ug, lyophilized powder
Purity	:	≥95% by RP-HPLC
Mol. Weight	:	8716 Da by MS (calc Mw 8722 Da)
Storage	:	powder at −20°C; buffered solution at −80°C. Please avoid multiple freeze/thaw cycles.

Productsheet

Background. Ub-AMC is a quenched fluorogenic substrate for deubiquitinating enzymes.[1,2] It is based on the C-terminal derivatization of ubiquitin with 7-amido-4-methylcoumarin (AMC).[3] Release of AMC fluorescence by deubiquitinating enzymes is monitored using Ex380 nm and Em460 nm wavelengths, respectively.

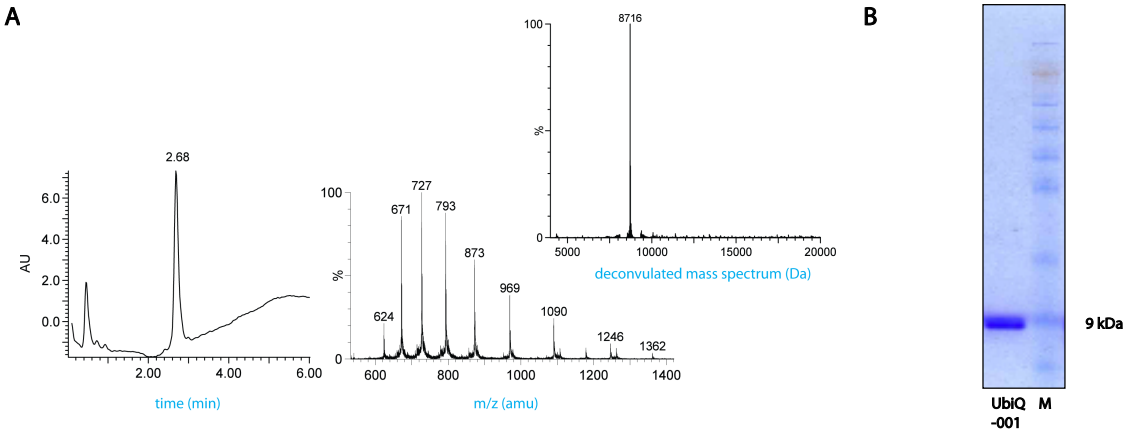
Sequence

MQIFVKTLTGKTITLEVEPSDTIENVKAKIQDKEGIPPDQQRLIFAGKQLEDGRTLSDYNIQKESTLHLVLRLRGG



Important: sample preparation

- **dissolve the powder in as little DMSO as possible (e.g. 20 mg/mL)**
- **add this DMSO stock slowly to milliQ (please note the order of addition)**
- **buffer the aq. solution as desired**
- **final stocks of e.g. 0.5 mg/mL will contain 2.5 vol% DMSO.**
- **buffer exchange using 3 kDa spin filters or dialysis membrane allows total removal of DMSO if desired.**



A: LC-MS analysis. Mobile phase A = 1% CH_3CN, 0.1% formic acid in water (milliQ) and B = 1% water (milliQ) and 0.1% formic acid in CH_3CN. Phenomenex Kinetex C18, (2.1×50 mm, 2.6 µM); flow rate = 0.8 mL/min, runtime = 6 min, column T = 40°C. Gradient: 5% ⇨ 95% over 3.5 min.
B: SDS-PAGE analysis. 12% SDS-PAGE gel. M= SeeBlue® Plus2 (Invitrogen). Coomassie Brilliant Blue

Literature. (1) Dang et al. *Biochemistry* **1998**, *37*, 1868. (2) Mason et al. *Biochemistry* **2004**, *43*, 6535. (3) El Oualid et al. *Angew Chem Int Ed* **2010**, *49*, 10149.

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