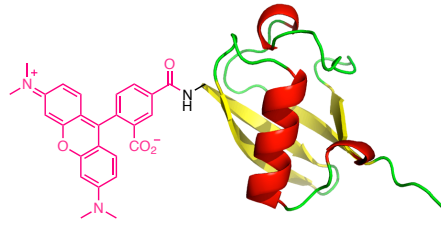


UbiQ

targeting the ubiquitin system



TAMRA-Ub (human sequence, synthetic)

UbiQ code : UbiQ-003

Batch # : B01072014-001

Amount : bulk, lyophilized powder

Purity : $\geq 95\%$ HPLC

MW : 8.98 kDa

Storage : upon arrival, powder at -20°C ; solution at -80°C . Please store dark and avoid multiple freeze/thaw cycles.

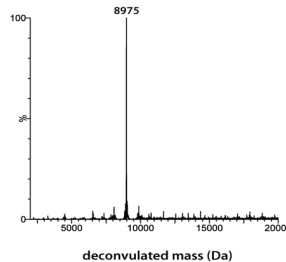
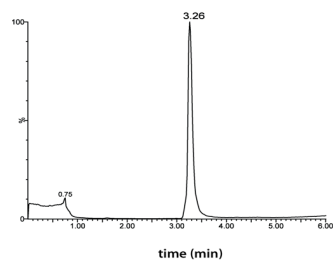
Productsheet

Background. UbiQ-003 is a Ub protein labeled on the N-terminus with the fluorescent dye TAMRA (5-tetramethylrhodamine, exc 550 nm, emi 590 nm). It has been prepared by total chemical synthesis¹ and allows detection of ubiquitylation by in-gel fluorescence.^{2,3} This direct and more sensitive read-out gives more distinct labeling patterns than immunoblotting and does not suffer from background labeling due to cross-reactivity (as seen sometimes with antibody based detection).

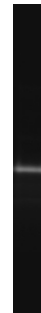
sequence

TAMRA-MQIFVKTLTGKTTITLEVEPSDTIENVKAKIQDKEGIPDPQRLIFAGKQLEDGRTLSDYNIQKESTLHLVLRGG

A



B



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 . XBridge BEH300 C18 $5\mu\text{m}$ $4.6 \times 100\text{mm}$; column T = 40°C , flow = 0.8 mL/min. Gradient: 30–95% over 3.5 min. **B: Fluorescent scan (550/590 nM), SDS-PAGE analysis.** 12% Bis-Tris, MES buffer. Please note that during fluorescence scanning of SDS-PAGE gels with (fluorescent) Ub proteins, the appearance of higher mol. weight bands ("smearing") can be observed. This can be caused by (heat-induced) aggregation (Morimoto et al. *Sci Rep* **2018**, *8*, article 2711). If possible, avoid heating the samples in Laemmli sample buffer for SDS-PAGE analysis.

important: sample preparation

- dissolve the powder in as little DMSO as possible (e.g. 20 mg/mL) and add this DMSO stock slowly to milliQ water (please note the order of addition).
- to ensure proper folding (and avoid precipitation), we advise to first buffer the aq. stock to 50 mM NaOAc pH 4.5
- next, buffer as desired.
- for examples of UbiQ-003 applications, please see ref. 4: <https://www.nature.com/articles/s41598-018-19538-0>

Literature. (1) El Oualid et al. *Angew Chem Int Ed* **2010**, *49*, 10149. (2) de Jong et al. *ChemBioChem* **2012**, *13*, 2251. (3) Smit et al. *J Biol Chem* **2013**, *288*, 31728. (4) Juenemann et al. *Sci Rep* **2018**, *8*, article number 1405.