

# UbiQ

targeting the ubiquitin system

## TAMRA-Ub (human sequence, synthetic)

UbiQ code : UbiQ-003

Batch # : B01072014-001

Amount : 100 ug, lyophilized powder

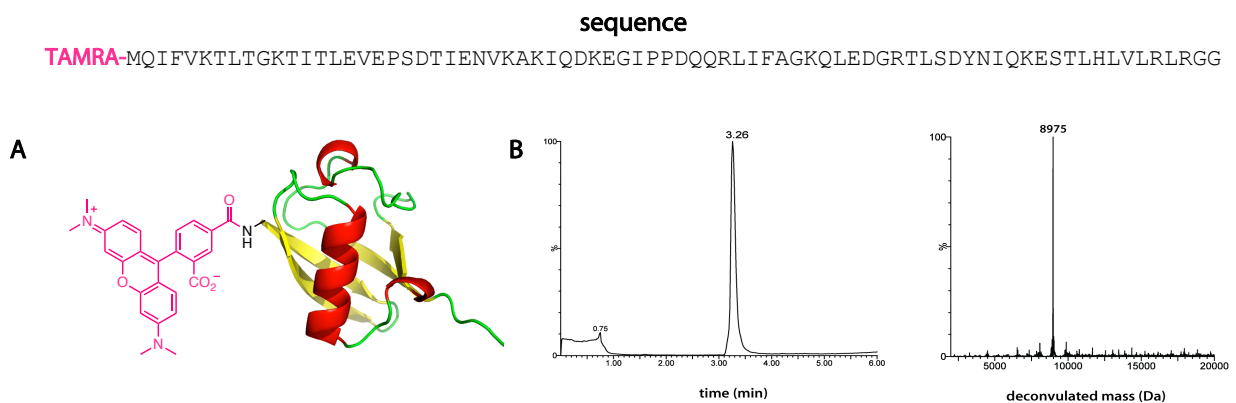
Purity :  $\geq 95\%$  HPLC

MW : 8.98 kDa

Storage : upon arrival, powder at  $-20^{\circ}\text{C}$ ; solution at  $-80^{\circ}\text{C}$ . Please store dark and avoid multiple freeze/thaw cycles.

## Productsheet

**Background.** UbiQ-003 (TAMRA-Ub) is based on ubiquitin (Ub) that is functionalised on the N-terminus with the fluorescent dye 5-tetramethylrhodamine (TAMRA, Figure 1A, exc 550 nm, emi 590 nm). UbiQ-003 can be used for the detection of ubiquitination by in-gel fluorescence analysis.



**Figure 1.** A: TAMRA-Ub. B: LC-MS analysis. Mobile phase A = 1%  $\text{CH}_3\text{CN}$ , 0.1% formic acid in water (milliQ) and B = 1% water (milliQ) and 0.1% formic acid in  $\text{CH}_3\text{CN}$ . XBridge BEH300 C18  $5\mu\text{m}$   $4.6 \times 100\text{mm}$ ; column T =  $40^{\circ}\text{C}$ , flow = 0.8 mL/min. Gradient: 30–95%B over 3.5 min.

### important: sample preparation

- dissolve the powder in as little DMSO as possible (e.g., 20 mg/mL)
- 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>

Please note that during SDS-PAGE analysis of 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 and/or add 4M urea to the SDS-PAGE samples.

**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.