

Ub-Rh110Gly (human sequence, synthetic)

UbiQ code : UbiQ-002

Batch # : B01022015-001

Amount : 100 ug, lyophilized powder

Purity : $\geq 95\%$ by RP-HPLC and SDS-PAGE

MW : 8.93 kDa

Storage : upon arrival powder at -20°C ; solution at -80°C . Please protect from light and avoid multiple freeze/thaw cycles.

Productsheet

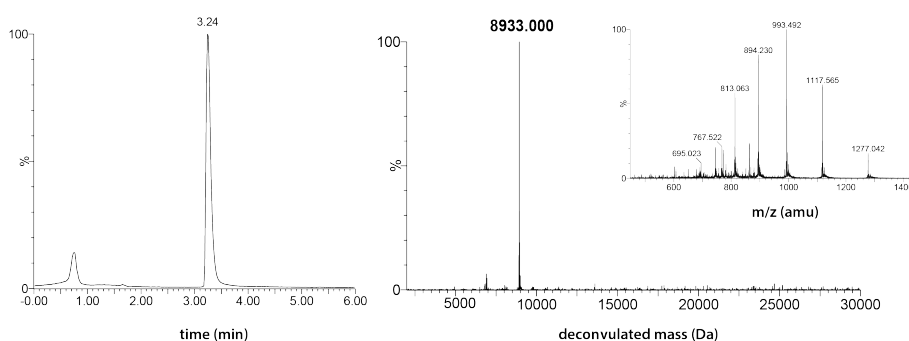
Background. Ub-Rh110Gly is a quenched, fluorescent substrate for deubiquitylases (DUBs), especially ubiquitin C-terminal hydrolases,^{1,2} prepared by chemical synthesis.³ Cleavage of the amide bond between Gly76 and the rhodamine110 moiety releases Rh110Gly which results in an increase in fluorescence at 535 nm (Exc. 485 nm).

Sequence

MQIFVKTLTGKTITLEVEPSDTIENVKAKIQDKEGIPPDQQLRIFAGKQLEDGRTLSDYNIQKESTLHLVLRIRGG-Rh110G

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.
- In general, DMSO conc up to 5 vol% are well tolerated by most DUBs



LC-MS analysis. Mobile phase A= 1% CH_3CN , 0.1% formic acid in milliQ and B= 1% 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.

Literature. (1) Tirat et al., *Analytical Biochem.* **2005**, *343*, 244-255. (2) Hassiepin et al. *Analytical Biochem.* **2007**, *371*, 201-207. (3) El Oualid et al. *Angew. Chem. Int. Ed.* **2010**, *49*, 10149.