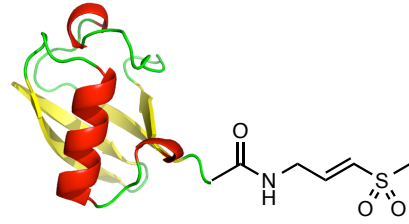


UbiQ

targeting the ubiquitin system



Ub-VS (human sequence, synthetic)

UbiQ code : UbiQ-108
Batch # : B20022015-001
Amount : 50 ug, lyophilized powder
Purity : $\geq 95\%$ by RP-HPLC and SDS-PAGE
Mol. Weight : 8.62 kDa
Storage : upon arrival powder at -20°C ; buffered solution at -80°C . Please avoid multiple freeze/thaw cycles.

Productsheet

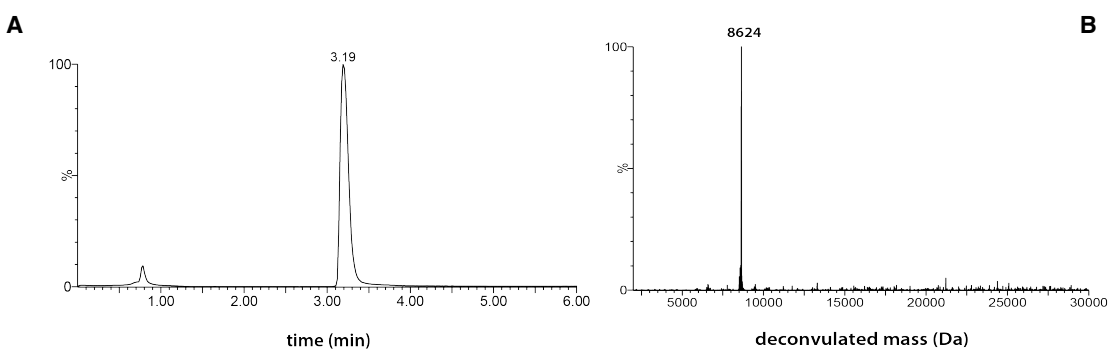
Background. Ub-VS (**UbiQ-108**) is an activity-based probe for deubiquitylating enzymes (DUBs) which can be used for activity profiling experiments, determining DUB inhibitor specificity and structural biology studies of DUB-Ub complexes.^{1,2}

Sequence

MQIFVKLTGKTITLEVEPSDTIENVKAKIQDKEGIPPDQQLIFAGKQLEDGRTLSDYNIQKESTLHLVLRRLRG-**VS**

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 (please note this is our preferred order of addition).
- next buffer with e.g. 1M HEPES to 50 mM HEPES. In general HEPES and Tris buffers are standard for DUB assays. Please note that certain DUBs react different to low or high NaCl concentrations.
- 2 - 5 mM TCEP or DTT can be used as reducing agent for the DUB.³
- a final buffered stock of for example 0.5 mg/mL contains 2.5 vol% DMSO; in general DMSO concentrations of up to 5 vol% are well tolerated by DUBs.
- if required, total removal of DMSO is accomplished by dialysis or spin-filtration (3 kDa cut-off membrane).



A: 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. **B: SDS-PAGE analysis.** 12% Bolt Bis-Tris Plus gel (Life technologies) and MES running buffer. CBB staining was performed with Coomassie G-250.

Literature. (1) Galardy et al. *Methods in Enzymology* **2005**, *399*, 120. (2) de Jong et al. *ChemBioChem* **2012**, *13*, 2251. (3) Wrigley et al. *Cell Biochem. Biophys.* **2011**, *60*, 99.