UbiQ
targeting the ubiquitin system

K11 Di-Ubiquitin VME (human, synthetic)

UbiQ code	:	UbiQ-082
Batch #	:	B01112014-001
Amount	:	25 ug, lyophilized powder
Purity	:	±90% by RP-HPLC and SDS-PAGE analysis*
Mol. Weight	:	17.11 kDa
Storage	:	upon arrival store powder at –20°C; solution at –80°C. Please avoid multiple freeze/thaw cycles.

Productsheet

Background. UbiQ-082 is a potent, irreversible and specific inhibitor of deubiquitinating enzymes (DUBs) based on K11 linked diUb.[1] Here Lys11 has been replaced by a diaminobutyric acid residue equipped with a VME type warhead[3] - the Dab(VME) type of structure is a DUB reactive mimic of the native isopeptidic linked Lys(Gly) residue (Figure 1). DUB activity based probes can be used for activity profiling experiments and structural studies.[1-8] Please note that the native distance between the proximal and distal Ub is preserved as much as possible in UbiQ-081.

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) and mix by vortexing**
- **final stocks of e.g. 0.5 mg/mL will contain 2.5 vol% DMSO.**
- **buffer the aq. stock as desired (with e.g. 1M HEPES or Tris, pH 7.5 - 8)**
- **in general, a DMSO concentration up to 5 vol% is well tolerated by DUBs.**
- **For more details see (open-access) reference: http://www.ncbi.nlm.nih.gov/pubmed/24623714**

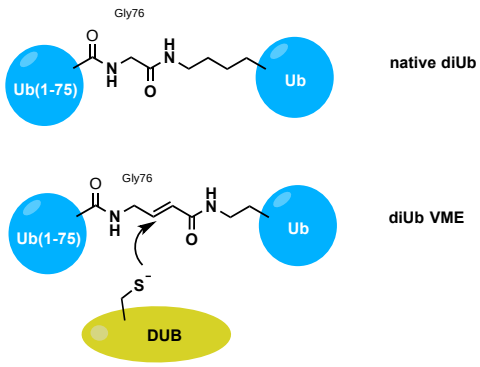

Figure 1

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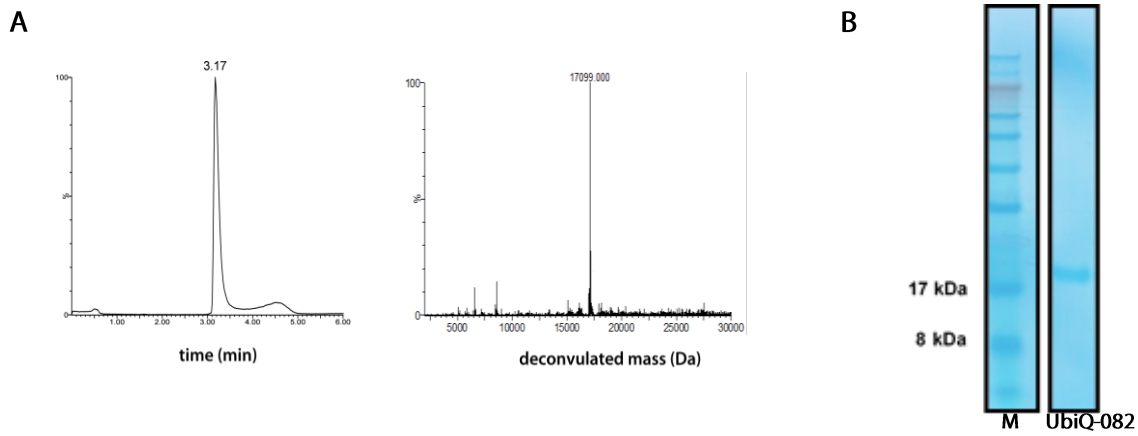

Figure 1. A: LC-MS analysis UbiQ-082 B01112014-001. Mobile phase A= 1% CH_3CN, 0.1% formic acid in milliQ and B= 1% milliQ and 0.1% formic acid in CH_3CN. XBridge BEH300 C18 5µm 4.6x100mm; column T= 40°C, flow= 0.8 mL/min. Gradient: 30–95% over 3.5 min. **B: SDS-PAGE analysis.** 12% Bolt Bis-Tris Plus gel (Life technologies) and MES running buffer. Marker= SeeBlue Plus2 Pre-stained Standard (Invitrogen). CBB staining was performed with Coommassie G-250.

* Based on SDS-PAGE analysis there is some Ub(1-75) present in the sample but this does not interfere with labeling experiments with DUBs.

** In some cases we and others have observed the appearance of higher mol. weight bands ("smearing") during SDS-PAGE analysis of (di)Ub conjugates. We do not have (analytical) evidence these are actual contaminants present in the diUb sample but that they are aggregates formed during SDS-PAGE. We have also not witnessed any effect of this phenomenon on experiments performed with our diUb material.

Literature. (1) Mulder & El Oualid et al. *ChemBioChem* **2014**, *15*, 946. (2) Misaghi et al. *J. Biol. Chem.* **2005**, *280*, 1512. (3) de Jong et al. *ChemBioChem* **2012**, *13*, 2251. (4) Altun et al. *Chem. Biol.* **2011**, *18*, 1401. (5) Haj-Yahya et al. *Org. Lett.*, **2014**, *16*, 540. (6) Li et al. *Chem. Commun.* **2014**, *50*, 216. (7) Iphöfer et al. *ChemBioChem* **2012**, *13*, 1416. (8) McGouran et al. *Chem. Biol.* **2013**, *20*, 1447.

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