

## Di-Ubiquitin VME explorer panel (human sequence, synthetic)

UbiQ code : UbiQ-L04 Batch # : B01022015-001

Amount :  $7 \times 10$  ug lyophilized powder

Purity : ±90%\* Mol. Weight : 17.11 kDa

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

## **Productsheet**

**Background.** UbiQ-L04 contains a panel of seven activity-based probes for deubiquitinating enzymes (DUBs) based on the diUb structure. Here, a Lys residue has been replaced by a diaminobutyric acid residue equipped with an electrophile (Figure 1A). The Dab(VME) electrophile traps the active site cysteine of a DUB and is a mimic of the native isopeptide Lys(Gly) linkage (Figure 1A). The activity-based probes can be used for activity profiling experiments and structural studies. Please note the native distance between the proximal and distal Ub is preserved as much as possible in our probe design (Figure 1A).

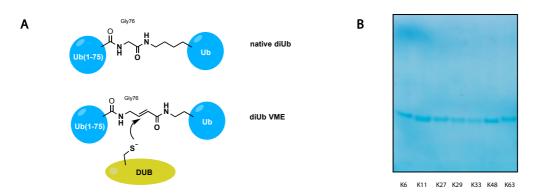


Figure 1. A: Design and mode of action diUb VME probes. B: SDS-PAGE analysis UbiQ-L04. 12% Bolt Bis-Tris Plus gel (Lifetechnologies), MES buffer. Staining with Coomassie Brilliant Blue G-250.

\*: 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.

\*\*: higher molecular weight artefacts are observed sometimes during SDS-PAGE analysis of monoUb reagents (especially with reactive DUB activity-based probes). There is no proof for these higher mol. weight bands being present in the material as judged by LC-MS analysis. 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 loading buffer.

## important: sample preparation

- dissolve the powder in as little DMSO as possible (e.g., 20 mg/mL= 10 ug in 0.5 uL DMSO)
- add this DMSO stock to milliQ (please note the order of addition) and mix by vortexing
- next buffer as desired (with e.g., 1M HEPES to 50 mM HEPES)
- for experimental details please see ref. 1: http://www.ncbi.nlm.nih.gov/pmc/articles/PMC4159580/

Literature. (1) Mulder & El Oualid et al. *ChemBioChem* **2014**, *15*, 946. (2) 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.