UbiQ
targeting the ubiquitin system

Di-ubiquitin explorer panel *(human sequence, synthetic and recombinant)*

UbiQ code	:	UbiQ-L01
Batch #	:	B01012018-001
Amount	:	8 x 10 µg lyophilized powder, M1 (i.e. linear), K6, K11, K27, K29, K33, K48 and K63 linked diUb
Purity	:	≥95%
Mol. Weight	:	17.11 kDa
Storage	:	upon arrival, powder at −20°C; solution at −80°C. Please avoid multiple freeze/thaw cycles.

Productsheet

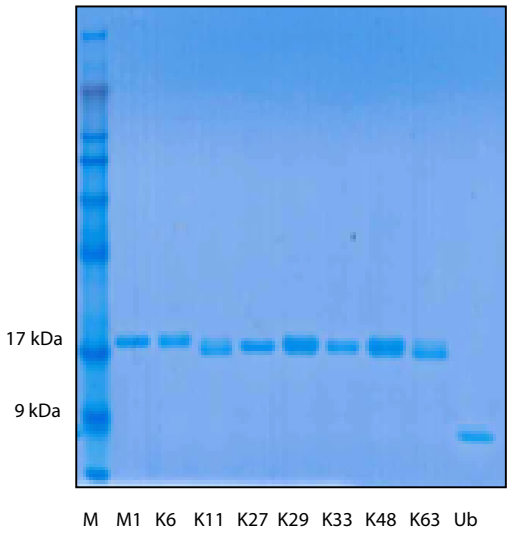
Background. UbiQ-L01 is a panel of all eight native linked di-ubiquitin (diUb) conjugates: M1 (i.e. linear), K6, K11, K27, K29, K33, K48 and K63 linked. The seven isopeptide linked diUb conjugates have been prepared by chemical ligation, the linear (M1) diUb conjugate by recombinant expression. UbiQ-L01 can be used to: [1-4]

- investigate linkage specificity of proteases that cleave the (iso)peptide linkage between two ubiquitin proteins.
- investigate mechanism of binding and recognition of proteins that contain ubiquitin-associated domains or ubiquitin-interacting motifs (UIMs).

Important - Sample preparation.

- add 0.5 µL DMSO to the 10 µg diUb sample and dissolve by a quick spin in the (ultra)centrifuge.
- add the DMSO stock (20 mg/mL= 1169 µM) to the required buffer (please note order of addition).
- for convenience one can use the lid of the eppendorf tube to hold the buffer while adding the 0.5 µL DMSO.
- dilution of 0.5 µL DMSO stock into 20 µL buffer affords a diUb solution of 30 µM with 2.5 vol% DMSO.
- buffer exchange using 3 kDa spin filters can be used to remove the DMSO if desired.
- Please note that a DMSO conc of <5 vol% is well tolerated in most experiments (e.g. with DUBs)

SDS-PAGE analysis. 12% Bolt Bis-Tris Plus gel (Lifetechnologies) in MES buffer at 190V, Staining with Coomassie Brilliant Blue G-250. Marker= SeeBlue® Plus2 (Invitrogen). We have observed the appearance of higher mol. weight bands ("smearing") during SDS-PAGE analysis of (di)Ub conjugates, believed to be caused by (heat-induced) aggregation.[5] If possible avoid heating the samples in Laemmli sample buffer for SDS-PAGE analysis.



Literature. (1) El Oualid et al. *Angew. Chem. Int. Ed.* **2010**, *49*, 10149. (2) Faesen et al. *Chemistry & Biology* **2011**, 18, 1550. (3) Dikic et al. *Nature Rev. Mol. Cell Biol.* **2010**, *10*, 659. (4) Licchesi et al. *Nature Struct. & Mol. Biol.* **2012**, *19*, 62. (5) Morimoto et al. *Sci Rep* **2018**, *8*, article number: 2711.

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