UbiQ
targeting the ubiquitin system

K382 Ub-p53(357-389)-FP *(human sequence, synthetic)*

UbiQ code	:	UbiQ-042
Batch #	:	B09082012-001
Amount	:	25 ug, lyophilized powder
Purity	:	≥95% by RP-HPLC
Mol. Weight	:	12.6 kDa
Storage	:	upon arrival, powder at −20°C; solution at −80°C. Store dark and avoid multiple freeze/thaw cycles.

Productsheet

Background. UbiQ-042 is al fluorescence polarization reagent based on the C-terminal peptide sequence 357 – 389 of p53, which contains various lysine residues that are (mono)ubiquitinated. In the case of UbiQ-041, the peptide is modified on the N-terminus with a 5-carboxytetramethylrhodamine and conjugated at Lys382 to ubiquitin (Ub) via a native isopeptide bond. UbiQ-042 contains a PGGS motif (aa 359-362) which was found to be important for USP7 binding.

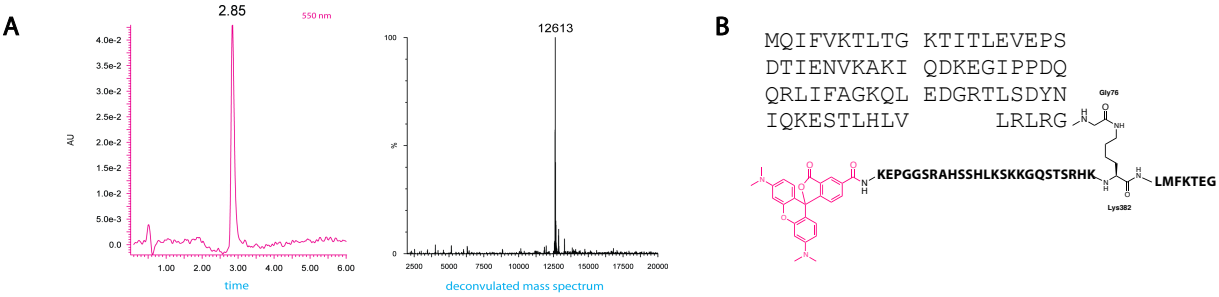

Figure 1. A: LC-MS analysis. Mobile phase A= 1% CH_3CN, 0.1% formic acid in water (milliQ) and B= 1% water (milliQ) and 0.1% formic acid in CH_3CN. Phenomenex Kinetex C18, (2.1×50 mm, 2.6 µM); flow rate= 0.5 mL/min, column T= 40°C. Gradient: 5-95%B over 3.5 min. B: Sequence UbiQ-042.

Important: sample preparation

- prepare (for example) a 2 mM DMSO stock (23 mg/mL)
- add the DMSO stock to milliQ, e.g. a 2 mM DMSO stock diluted 20× in milliQ affords a 100 µM stock which can be aliquoted and stored.
- for assays this 100 µM stock can be diluted for example 1000× in buffer affording a final assay solution of 100 nM. The DMSO concentration during the assay is now 0.01 vol%.
- for full experimental details please see reference 5.

Literature. (1) Tirat et al. *Anal. Biochem.* **2005**, *343*, 244. (2) Huang et al. *Methods in Mol Biol* **2009**, *565*, 127. (3) Levine et al. *Anal. Biochem.* **1997**, *247*, 83. (4) Faesen et al. *Chem. Biol.* **2011**, *18*, 1550. (5) Geurink et al. *ChemBiochem* **2012**, *13*, 293. (6) Sheng et al. *Nat. Struct. Mol. Biol.* **2006**, *13*, 285.

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