

Biotin-Ahx-Ub pTyr59 (*human sequence, synthetic*)

UbiQ code : UbiQ-095

Batch # : B01102016-001

Amount : 50 ug, lyophilized powder

Purity : $\geq 95\%$ by RP-HPLC and SDS-PAGE

Mol. Weight : 8.98 kDa

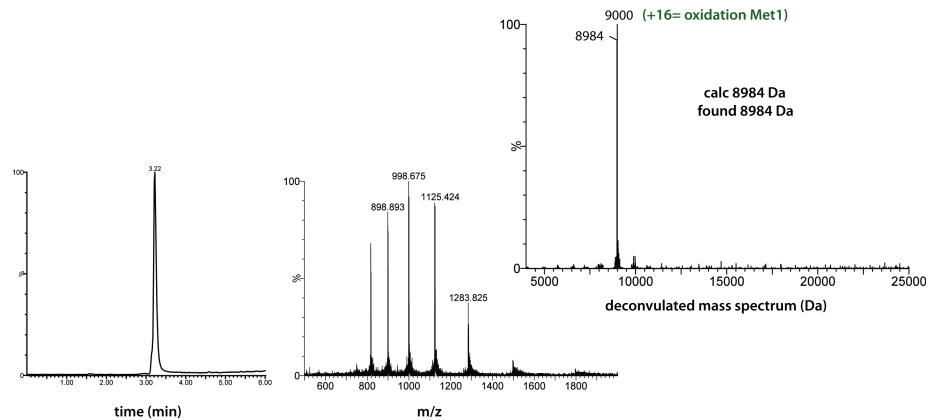
Storage : upon arrival powder at -20°C ; buffered solution at -80°C . Please avoid multiple freeze/thaw cycles.

Productsheet

Background. Biotin-Ahx-Ub pTyr59 (UbiQ-095) is a ubiquitin protein that is phosphorylated on Tyr59. Phosphoproteomic studies have identified several phosphorylated sites in ubiquitin, among them Tyr59.¹⁻⁵ It is labeled on the *N*-terminus with biotin; an aminohexanoic acid (Ahx) linker is used to create extra space between the biotin and Ub protein for efficient access of biotin binding entities. It has been made by total chemical synthesis⁶ and is therefore well-defined in terms of biotin and pTyr site and incorporation efficiency (100%).

Sequence

biotin-Ahx-MQIFVKLTGKITLEVEPSDTIENVKAKIQDKEGIPPDQQLRIFAGKQLEDGRTLSDY^PNIQKESTLHLVLRRGG



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 . XBridge BEH300 C18 5 μm 4.6x100mm; column T = 40°C , flow = 0.8 mL/min. Gradient: 30–95% over 3.5 min.

Important: sample preparation

- dissolve the powder in as little DMSO as possible (e.g. 40 mg/mL)
- **add this DMSO stock slowly to milliQ (please note the order of addition)**
- buffer the aq. solution as desired (final stocks of e.g. 0.5 mg/mL will contain 1.25 vol% DMSO)
- buffer exchange using 3 kDa spin filters or dialysis membrane allows total removal of DMSO if desired.



Literature.

- 1) Bennetzen et al. *Mol Cell Proteomics* **2010**, *9*, 1314.
- 2) Bian et al. *J Proteomics* **2014**, *96*, 253.
- 3) Kettenbach et al. *Sci Signal*, **2011**, *4*, rs5.
- 4) Sharma et al. *Cell Rep* **2014**, *8*, 1583.
- 5) Zhou et al. *J Proteome Res* **2013**, *12*, 260.
- 6) El Oualid et al. *Angew Chem Int Ed* **2010**, *49*, 10149.