

# Biotin-Ub-PA

Cat. # M1020

**Quantity:** 25 µg  
**Species:** Human  
**Source:** Synthetic  
**MW:** 8885 Da  
**Form:** Lyophilized powder  
**Quality Assurance:** ≥95% by RP-HPLC

**Description:** Biotin-Ub-PA is a newly developed potent and specific inhibitor of deubiquitinating enzymes (DUBs), which has a N-terminal biotin tag. An aminohexanoic acid (Ahx) linker is used to create extra space between the biotin and Ub protein for efficient access of the biotin binding entity. This activity probe can be used for activity profiling experiments and determining DUB inhibitor specificity. It has two unique capabilities: it forms a covalent linkage with the active site Cys residue of a DUB that can be cleaved by acid treatment (5% aq. TFA), allowing for proteomic analyses. Biotin-Ub-PA targets all major cysteine DUB families.

**Images:**



**SDS-PAGE analysis.**  
Coommassie blue staining, 12% SDS-PAGE gel.

**Storage:** Powder at  $-20^{\circ}\text{C}$ ; solution at  $-80^{\circ}\text{C}$ . Protect from light and avoid multiple freeze/thaw cycles.

**Sample preparation (important!):**

- 1) Centrifuge the tube at 10,000 xg for 2 min to pellet the powder.
- 2) Dissolve the powder in a small amount of DMSO (e.g. 25 µg powder in 1 µL DMSO). Vortex the tube to completely dissolve the powder. Keep under room temperature for 5 min, and then centrifuge under room temperature at 10,000 xg for 2 min to collect solution to the tube bottom.
- 3) Add 49 µL cold buffer (such as 20 mM Tris, pH 7.2, 150 mM NaCl and 10% glycerol) directly into the tube bottom in once, and pipette up and down to mix (avoid generating bubbles and note the order of addition).
- 4) The stock solution is 0.5 µg/µL (56 µM). Working concentrations vary from 100 nM–2 µM.



**Literature:**

1. Ekkebus *et al.* , (2013) J. Am. Chem. Soc. 135, 2867.
2. Sommer *et al.* , (2013) Bioorg. Med. Chem. 21, 2511.
3. Galardy *et al.* , (2005) Methods in Enzymology 399, 120.
4. de Jong *et al.* . (2012) ChemBioChem 13, 2251.

