

UbiQ

targeting the ubiquitin system

5-carboxyRh110-Nedd8-Dha (human sequence, synthetic)

UbiQ code : UbiQ-122

Batch # : B01072015-001

Amount : 50 ug, lyophilized powder

Purity : ≥95%

Mol. Weight : 8.95 kDa

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

Productsheet

Background. UbiQ-122 is an activity-based probe for Nedd8 E1, E2 and (HECT/RBR type) E3 ligases. It is based on Nedd8 in which Gly76 has been replaced by a dehydroalanine (Dha) residue. The N-terminus is labeled with the green fluorescent 5-carboxyrhodamine110 dye (cRh110, $\lambda_{ex} = 480 \text{ nm}$; $\lambda_{em} = 520 \text{ nm}$). It is processed in a native manner by Nedd8 E1, E2 and (HECT/RBR) E3 enzymes and during this process it forms an electrophilic intermediate that can react with the active site Cys residue of the E1, E2 and (HECT/RBR) E3 enzyme, thereby creating a covalent bond (Figure 1C).

A

cRh110-MLIKVKTLTGKEIEIDIEPTDKVERIKERVEEKEGIPPQQRLIYSGKQMNDEKTAADYKILGGSVLHLVLALRG-Dha

B

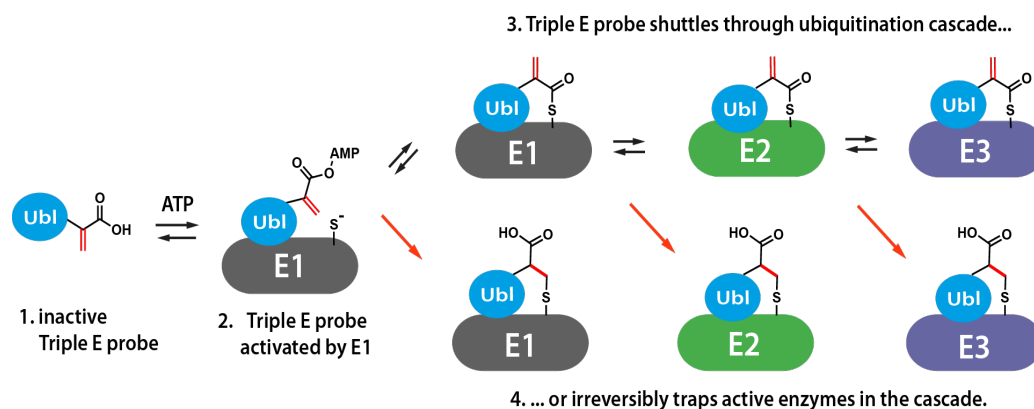


Figure 1. A: sequence. B: D: Mode of action Nedd8-Dha activity-based probes.

important: sample preparation

- dissolve the powder in as little DMSO as possible (20 - 40 mg/mL)
- add the DMSO stock to 300 mM NaCl (please note the order of addition) and mix - *at this step we have included a high salt aq. solution because Nedd8 is more stable at high salt concentration.*
- buffer the aq. solution as desired
- For full details please see open-access reference 1: <https://www.ncbi.nlm.nih.gov/pmc/articles/PMC5108872/>

Literature. (1) Mulder et al. *Nat Chem Biol* 2016, 12, 523.