

MATERIAL DATA SHEET

Recombinant Human Ubiquitin S65A

Cat. # UM-S65A

Serine/Threonine kinase PINK1 (PTEN-induced putative kinase protein 1) plays a critical role in preventing mitochondrial dysfunction during cellular stress. PINK is translated in the cytosol, then translocated to the outer mitochondrial membrane where it is rapidly cleaved and degraded as a part of normal mitochondrial function. In damaged (depolarized) mitochondria PINK becomes stabilized and accumulates, resulting in the subsequent phosphorylation of numerous proteins on the mitochondrial surface including Mfn2. Ultimately PARK2 (E3 Ubiquitin Ligase Parkin) is recruited to the damaged mitochondria where it is activated by PINK-mediated phosphorylation of PARK2 at serine 65, and PARK2 interaction with phosphorylated Ubiquitin (also phosphorylated by PINK on serine 65). This signaling cascade is critical for clearing the damaged mitochondria via selective autophagy (mitophagy) by mediating activation and translocation of PARK2. Recombinant Ubiquitin Mutant S65A may be used as a negative control in experiments examining the *in vitro* phosphorylation of Ubiquitin using PINK1 kinase from Red Flour Beetle (*Tribolium castaneum*) or other sources. Ubiquitin mutant S65A is not phosphorylated by PINK1 *in vitro*, even in extended reactions.

Product Information

Quantity:	100 µg
MW:	8.6 kDa
Source:	<i>E. coli</i> -derived Accession # P0CG47 Contains a Ser-to-Ala substitution at position 65.
Stock:	Lyophilized from a solution in deionized water.
Solubility:	Reconstitute at 2 mg/ml in an aqueous solution
Purity:	>98%, by SDS-PAGE under reducing conditions and visualized by Colloidal Coomassie® Blue stain.

Use & Storage

Use: Recombinant Ubiquitin Mutant S65A may be used as a negative control in experiments examining PINK1 kinase activity. Ubiquitin S65A is not phosphorylated by PINK1 *in vitro*. Reaction conditions will need to be optimized for each specific application.

Storage: Use a manual defrost freezer and avoid repeated freeze-thaw cycles.

- 12 months from date of receipt, -20 to -70 °C as supplied.
- 3 months, -20 to -70 °C under sterile conditions after reconstitution.

Literature

References:

1. Fiesel F.C., *et al.* (2015) EMBO Reps. DOI 10.15252/embr.201540514
2. Kane L.A., *et al.* (2014) J. Cell Biol. **205**: 143
3. Matsuda N., *et al.* (2010) J. Cell Biol. **189**: 211
4. Ordureau A., *et al.* (2014) Mol Cell. **56**: 360
5. Vives-Bauza C., *et al.* (2010) Proc. Natl. Acad. Sci. **107**: 378
6. Wauer T., *et al.* (2015) EMBO J. **34**: 307

For research use only. Not for use in humans.