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MATERIAL DATA SHEET

Recombinant Human phospho-Tetra-Ubiquitin/Ub4 WT Chains (M1-linked, pS65)

Cat. # UC-750

Linkage specific phosphorylated Poly-Ubiquitin chains may be used as a substrate for *in vitro* reactions with deubiquitinating enzymes ("DUB's") that cleave the peptide or isopeptide linkage between adjacent Ubiquitin molecules. Phosphorylated Poly-Ubiquitin chains can also be used to investigate mechanisms of binding and recognition between the chains and other proteins that contain Ubiquitin-Associated domains (UBAs), Ubiquitin-interacting motifs (UIMs), ZnF's and/or other Ubiquitin-sensing elements.

Linear (Met1-linked) Tetra-Ubiquitin chains phosphorylated at Serine 65 are manufactured using recombinant wild-type human recombinant Ubiquitin and PINK1 kinase. The use of purely enzymatic techniques avoids the potential for contaminating synthetic intermediates. The purity of each production lot is assessed using the Absolute Quantitation of Ubiquitin method (Ub-AQUA), an LCMS-based technique that provides extremely accurate information on the composition of Poly-Ubiquitin samples.

Product Information	
Quantity:	25 µg
MW:	34 kDa
Source:	*E. coli*-derived Accession # P0CG47
Stock:	1 mg/ml (29 µM) in sterile, deionized water
Purity:	>95%, by SDS-PAGE under reducing conditions and visualized by Colloidal Coomassie® Blue stain. **Ub-AQUA analysis**: p-S65: 93.5%

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Use & Storage

Use: Ubiquitin chains vary in length, linkage, and function. Linear ("Met1" or "M1"), pS65 Tetra-Ubiquitin chains are ideal for investigating Ubiquitin-binding proteins and as substrates for Ubiquitin-specific isopeptidases. Reaction conditions will need to be optimized for each specific application. IMPORTANT: Heating this product in SDS-PAGE buffer or terminating reactions containing this product with heated SDS--PAGE buffer could lead to unexpected, high apparent molecular weight banding or smearing on gels that is not representative of product purity. For optimal results, we recommend incubation in SDSPAGE buffer + DTT at ≤40 °C for 20 minutes prior to gel electrophoresis

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

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

Literature

References:

1. Heo, J.M., Ordureau A., *et al.* (2015) Mol. Cell **60(1)**: 7-20
2. Kirkpatrick D.S., *et al.* (2006) Nat Cell Biol. **8(7)**: 700-10
3. Ordureau, A., *et al.* (2014) Mol. Cell **56(3)**: 360–375
4. Ordureau, A., *et al.* (2015) Pro. Nat. Acad. of Sci. USA **112(21)**: 6637–6642
5. Phu L., *et al.* (2011) Mol Cell Proteomics **10(5)**: M110.003756

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