

MATERIAL DATA SHEET

Ubch7/UBE2L3-Ubiquitin Charged, *human recombinant* Cat. # E2-803

Ubiquitin-conjugating Enzyme H7 (Ubch7), also known as Ubiquitin-conjugating Enzyme E2L3 (UBE2L3), is a member of the Ubiquitin-conjugating (E2) enzyme family. It has a predicted molecular weight of approximately 18 kDa. The human Ubch7 protein shares 100% amino acid (aa) sequence identity with the mouse and rat orthologs. Ubch7 has an E2 catalytic core domain that contains an active site cysteine residue and comprises 152 of its 154 aa residues. Ubch7 is catalytically active with HECT and RBR domain-containing families of Ubiquitin ligases (E3s). Ubch7 localizes to both the nucleus and cytoplasm in human cells. In mice, its ortholog is expressed in many tissues including brain, muscle, heart, lung, lymph node, spleen, thymus, and testis. Ubch7 depletion results in an extended S phase and a reduced rate of proliferation, suggesting that it may play a role in the cell cycle. In humans, single nucleotide polymorphisms in Ubch7 are associated with systemic lupus erythematosus and Crohn's disease, suggesting that Ubch7 is important for proper immune system function.

This product is an enzymatically generated Ubch7/UBE2L3-Ubiquitin thioester complex that has been highly purified to remove E1 Ubiquitin Activating enzyme, uncharged Ubch7/UBE2L3, free Ubiquitin, and Mg^{2+} -ATP. The product provides a convenient starting material for use in single-turnover "Ubiquitin Discharge Assays," eliminating the need to either inhibit the E1 Ubiquitin Activating enzyme with potentially confounding chemical treatments or remove ATP via enzyme additions.

Product Information

Quantity:	100 µg
Stock:	X mg/ml (X µM) in 50 mM HEPES pH 7.5, 50 mM NaCl, 1 mM TCEP
MW:	26 kDa (18 kDa UBE2L3, 8.6 kDa Ubiquitin)
Purity:	> 95 % by SDS-PAGE under non-reducing conditions and visualized by Colloidal Coomassie Blue Stain.

Use & Storage

Use:	Pre-charged UBE2L3 may be added directly to <i>in vitro</i> reactions containing E3 Ubiquitin ligases and ligase substrates (if applicable)—no ATP, E1 enzyme, or extra Ubiquitin are required. Reaction conditions will need to be optimized for each specific application. Note: Reducing agents including dithiothreitol (DTT) or mercaptoethanol (βME) may cause unintended thiolytic release of Ubiquitin from the complex—care must be taken if these compounds are present in buffers. We suggest using a thioester friendly reductant such as TCEP if possible.
Storage:	Store at -80°C. Avoid multiple freeze-thaw cycles. Avoid DTT, βME, and other thiol-containing compounds in buffers.

840 Memorial Drive, Cambridge, MA 02139 Phone: 617-576-2210 FAX: 617-492-3565

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Literature

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