

MATERIAL DATA SHEET

Ubiquitin Mutant with K6 only, human recombinant Cat. # UM-K60

Ubiquitin is a 76 amino acid (aa) protein that is ubiquitously expressed in all eukaryotic organisms. Ubiquitin is highly conserved with 96% aa sequence identity shared between human and yeast ubiquitin, and 100% as sequence identity shared between human and mouse ubiquitin. In mammals, four ubiquitin genes encode for two ubiquitin-ribosomal fusion proteins and two poly-ubiquitin proteins. Cleavage of the ubiquitin precursors by deubiquitinating enzymes gives rise to identical ubiquitin monomers each with a predicted molecular weight of 8.6 kDa. Conjugation of ubiquitin to target proteins involves the formation of an isopeptide bond between the C-terminal glycine residue of ubiquitin and a lysine residue in the target protein. This process of conjugation, referred to as ubiquitination or ubiquitylation, is a multi-step process that requires three enzymes: a Ubiquitin-Activating Enzyme (E1), a Ubiquitin-Conjugating (E2) enzyme, and a Ubiquitin ligase (E3). Ubiquitination is classically recognized as a mechanism to target proteins for degradation and as a result, ubiquitin was originally named ATP-dependent Proteolysis Factor 1 (APF-1). In addition to protein degradation, ubiquitination has been shown to mediate a variety of biological processes such as signal transduction, endocytosis, and post-endocytic sorting. Highly purified ubiquitin processed for the quantitative removal of glycine and buffer salts which can interfere with chemical and in vitro reactions. This ubiquitin mutant contains only a single lysine, K6, with all other lysines mutated to arginine. This ubiquitin is able to form polyubiquitin chains with other ubiquitin molecules only via the K6 lysine or amino terminus.

Product Information

Quantity: 1 mg, lyophilized powder

MW: 8.7 kDa

Solubility: Reconstitute in aqueous buffers at 5-10 mg/ml.

Purity: > 95% by SDS-PAGE under reducing conditions and visualized by Colloidal

Coomassie Blue stain

Use & Storage

The lysine residue utilized for Ubiquitin chain formation is functionally important. Single lysine Ubiquitin mutants are ideal for investigating biological processes involving specific Ubiquitin chain linkage. Recombinant Human

Use: Ubiquitin K6-only allows for the formation of only K6-linked or linear ("Met1")

polyubiquitin chains. Reaction conditions will need to be optimized for each specific application. We recommend an initial Recombinant Human Ubiquitin

K6-only concentration of 0.2-1 mM.

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

reconstituted product at -20° to -80°C

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Literature

References: Baboshina D.V., et al. (1996) <u>J.Biol.Chem.</u> **271**:2823-2831

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