

MATERIAL DATA SHEET

Recombinant Human HA Ubiquitin Aldehyde C-Terminal Derivative Cat. # U-211

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% aa sequence identity shared between human and mouse Ubiquitin (1). 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 (E1) enzyme, 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) (2,3). In addition to protein degradation, ubiquitination has been shown to mediate a variety of biological processes such as signal transduction, endocytosis, and postendocytic sorting (4-7).

Ubiquitin-aldehyde is a potent and specific inhibitor of most deubiquitinating enzymes (DUBs) such as Ubiquitin C-terminal hydrolases (UCHs) and Ubiquitin-specific proteases (USPs). It prevents the hydrolysis of poly-Ubiquitin chains on substrate proteins *in vitro* and thus enhances poly-Ubiquitin chain accumulation. This tagged version contains an N-terminal HA peptide sequence (YPYDVPDYA) derived from the influenza Hemagglutinin protein. This epitope allows for the sensitive identification or purification of DUBs since it is specifically recognized by Anti-HA antibodies and/or Anti-HA-agarose.





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Product Information

Quantity: 50 μg

MW: 9.8 kDa

Source: *E. coli*-derived

Contains an N-terminal HA (YPYDVPDYA) tag and a mixture of derivatized and

underivatized C-terminal Aldehyde, quantity is by derivatized content.

Accession # P0CG47

Stock: Lyophilized from a solution in MES.

Solubility: Reconstitute at 2 mg/mL in an aqueous solution.

Purity: >95%, by HPLC.

Use & Storage

Use: Add Recombinant Human HA-Ubiquitin Aldehyde to in vitro assays to inhibit

deubiquitinating enzymes. The HA-tag allows for detection and purification of deubiquitinating enzyme activity. Reaction conditions will need to be optimized for each specific application. We recommend an initial Recombinant Human HA-

Ubiquitin Aldehyde concentration of 2-5 μM.

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.



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Literature

References:

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