

Lot # XXXXX

## MATERIAL DATA SHEET

### **Biotinylated Ubiquitin, *human recombinant***

**Cat. # UB-570**

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. 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.

Ubiquitin modified with biotin via primary amine coupling results in multiple biotinylated ubiquitin species modified at the N-terminus, as well as lysine residues. Although having a fully functional C-terminus, lysine modification may partially limit the ability of this reagent to propagate polyubiquitin chains. Biotinylated ubiquitin can be detected using avidin-linked reagents.

#### Product Information

<b>Quantity:</b>	100 µg
<b>Stock:</b>	X mg/ml in 10 mM HEPES, pH 7.5
<b>MW:</b>	8.6 kDa (unlabeled), extent of biotinylation varies by lot
<b>Purity:</b>	> 95% by SDS-PAGE under reducing conditions and visualized by Colloidal Coomassie Blue stain

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

[www.bostonbiochem.com](http://www.bostonbiochem.com)

The contents of this datasheet (unless otherwise noted) are Copyright © 2008 Boston Biochem, Inc. All rights reserved. Duplication in whole or in part is strictly prohibited without the express written consent of Boston Biochem, Inc. "Boston Biochem" is a Trademark of Boston Biochem, Inc., 840 Memorial Drive, Cambridge, MA 02139.

### Use & Storage

<b>Use:</b>	Biotinylated Recombinant Human ubiquitin can be conjugated to substrate proteins via the subsequent actions of a Ubiquitin-Activating Enzyme (E1), a Ubiquitin-Conjugating Enzyme (E2), and a Ubiquitin Ligase (E3). Reaction conditions will need to be optimized for each specific application. We recommend using Biotinylated Recombinant Human Ubiquitin in conjunction with native ubiquitin at a combined concentration of 10-50 µM with a 1:1 to 1:5 ratio of biotinylated ubiquitin:native ubiquitin. The resulting poly-Ubiquitin chains can be visualized or captured with avidin-linked reagents.
<b>Storage:</b>	Store solution at -20°C or -80°C.

### Literature

<b>References:</b>	Ciechanover, A. <i>et al.</i> (1980) <u>Proc. Natl. Acad. Sci. USA</u> <b>77</b> : 1365
	Greene, W. <i>et al.</i> (2012) <u>PLoS Pathog.</u> <b>8</b> :e1002703
	Hershko, A. <i>et al.</i> (1980) <u>Proc. Natl. Acad. Sci. USA</u> <b>77</b> : 1783
	Sharp, P.M. & W.-H. Li. (1987) <u>Trends Ecol. Evol.</u> <b>2</b> : 328
	Tong, X. <i>et al.</i> (2012) <u>J. Biol. Chem.</u> <b>287</b> : 25280
	Wei, W. <i>et al.</i> (2004) <u>Nature</u> <b>428</b> : 194
	Wertz, I.E. <i>et al.</i> (2004) <u>Nature</u> <b>430</b> : 694

***For Laboratory Research Use Only, Not For Use in Humans***

Rev: 8/22/2018

840 Memorial Drive, Cambridge, MA 02139 Phone: 617-576-2210 FAX: 617-492-3565  
[www.bostonbiochem.com](http://www.bostonbiochem.com)

The contents of this datasheet (unless otherwise noted) are Copyright © 2008 Boston Biochem, Inc. All rights reserved. Duplication in whole or in part is strictly prohibited without the express written consent of Boston Biochem, Inc. "Boston Biochem" is a Trademark of Boston Biochem, Inc., 840 Memorial Drive, Cambridge, MA 02139.