

Lot # XXXXX

MATERIAL DATA SHEET

ISG15 AMC, human recombinant **Cat. # UL-553**

Interferon-stimulated Gene 15 (ISG15), also known as Ubiquitin Cross-reacting Protein (UCRP), is a Ubiquitin-like protein that is covalently coupled to target proteins in a process termed ISGylation. It is a 165 amino acid (aa) polypeptide with a predicted molecular weight of 18 kDa. Structurally, ISG15 consists of two tandem Ubiquitin-like domains that share a similar structure with Ubiquitin and other Ubiquitin-like modifiers. Modification of targets by ISG15 occurs in a stepwise enzymatic process similar to that of Ubiquitin. Enzymes regulating ISGylation include the activating (E1) enzyme UBE1L, the conjugating (E2) enzyme UbcH8, and ligases (E3) such as EFP/TRIM25 and HERC5. Removal of ISG15 is catalyzed by the deconjugating enzyme UBP43/USP18. Functionally, ISG15 has putative roles in the immune response and tumorigenesis. This is reflected by intracellular ISG15 targets that include Cyclin D1, tumor suppressor p63, IRF3, and a range of viral proteins. It is induced by type 1 interferons and microbial infection, and knockout mice exhibit an increased sensitivity to infection by some viruses. ISG15 can also be secreted by cells of the immune system and may act in a cytokine-like manner. For instance, it is produced by human granulocytes in response to mycobacterium exposure, and natural killer cells and T cells respond to extracellular ISG15 with IFN-gamma production. Further supporting a role in immune function, ISG15 mutations are associated with MSMD, an inherited disorder characterized by increased susceptibility to mycobacterial infection.

This fluorogenic substrate for ISG15 hydrolases is based on the carboxy-terminus derivatization of ISG15 with 7-amido-4-methylcoumarin (AMC). ISG15 AMC is useful for studying enzymes (such as UBP43 and Papain-Like Protease from SARS coronavirus) when detection sensitivity or continuous monitoring of activity is essential.

Product Information

Quantity:	50 µg
Stock:	X mg/ml (X µM) in 50 mM HEPES pH 7.5, 100 mM NaCl, 2mM DTT, 20% Glycerol
MW:	19 kDa

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Use & Storage

Use:	Substrate concentrations for assay range from 0.05-0.5 μ M depending on conditions. AMC fluorescence can be monitored using excitation and emission wavelengths of 345 nm and 445 nm respectively.
Storage:	Store at -80°C. Avoid multiple freeze/thaw cycles. Thaw tubes by rolling back-and-forth between hands. Mix gently and often. We do not recommend placing vials in 37°C waterbath to thaw.

Literature

References:	<p>Bogunovic, D. <i>et al.</i> (2012) <u>Science</u> 337:1684. Harty, R.N. <i>et al.</i> (2009) <u>J. Innate. Immun.</u> 1:397. Jeon, Y.J. <i>et al.</i> (2012) <u>J. Clin. Invest.</u> 122:2622. Malakhov, M.P. <i>et al.</i> (2002) <u>J. Biol. Chem.</u> 277:9976. Owashi, M. <i>et al.</i> (2003) <u>Biochem. Biophys. Res. Comm.</u> 309:533. Wong, J.J. <i>et al.</i> (2006) <u>Proc. Natl. Acad. Sci. USA</u> 103:10735. Yuan, W. & R.M. Krug (2001) <u>EMBO J.</u> 20:362. Zhang, D. & D.-E. Zhang (2011) <u>J. Interferon Cyto. Res.</u> 31:119. Zhao, C. <i>et al.</i> (2004) <u>Proc. Natl. Acad. Sci. USA</u> 101:7578. Zou, W. & D.E. Zhang (2006) <u>J. Biol. Chem.</u> 281:3989.</p>
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