PRODUCT DATASHEET

ASH1L-[BRD] (His) (ASH-1 like protein; huASH1; KMT2H)

CATALOG NO.: RD-11-202 **LOT NO.:**

DESCRIPTION: Human recombinant ASH1L bromodomain (residues 2428-2559; Genbank Accession # NM_018489; MW = 17.7 kDa) expressed in *E. coli* with an N-terminal His-tag. ASH1L is a large (333 kDa), multi-domain protein associated with actively transcribed regions of chromatin. Its bromodomain lies C-terminal to its SET domain, which confers histone H3K36 methyltransferase activity[1-3]. ASH1L is the human homolog of *Drosophila* Ash1, a Trithorax group protein. Like its counterpart in *Drosophila*[4], ASH1L has been found to play a role in the regulation of Hox gene expression[5,6]. The ASH1L bromodomain displays strong binding to various Lys(Ac) residues in singly acetylated histone peptide microarrays (histones H1.4K74Ac, H2AK36Ac, H2BK85Ac, H3K56Ac, H4K59Ac/K79Ac)[7].

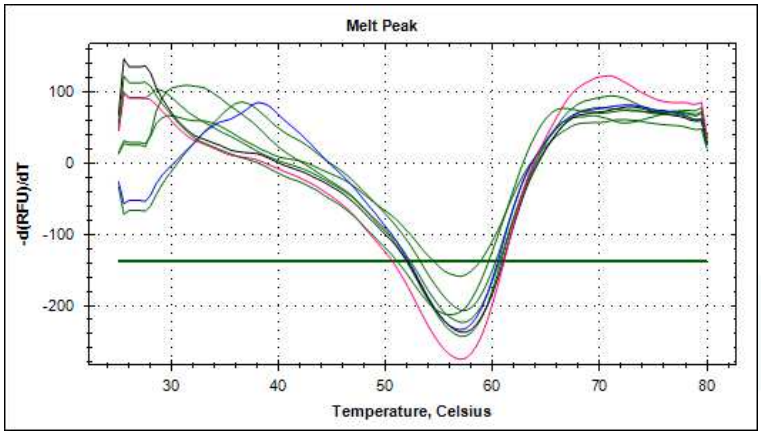
PURITY: >95% by SDS-PAGE

SUPPLIED AS: _ µg/µL in 50mM Tris HCl, pH 7.5, 500mM NaCl, 1mM TCEP, 10% glycerol (v/v) as determined by OD_{280}

STORAGE: -70°C. Thaw quickly and store on ice before use. The remaining, unused, undiluted protein should be snap frozen, for example in a dry/ice ethanol bath or liquid nitrogen. Minimize freeze/thaws if possible, but very low volume aliquots (<5 µl) or storage of diluted protein is not recommended.

REFERENCES: 1) Y Tanaka *et al. Gene* 2007 **397** 161; 2) S. An *et al. J. Biol. Chem.* 2011 **286** 8369; 3) D.S. Cabianca *et al. Cell* 2012 **149** 819; 4) C. Beisel *et al. Nature* 2002 **419** 857; 5) G. Gregory & *et al. Mol. Cell. Biol.* 2007 **27** 8466; 6) Y Tanaka *et al. PLOS One* 2011 **6** e28171; 7) P. Filippakopoulos *et al. Cell* 2012 **149** 214

Coomassie blue-stained SDS-PAGE (4-12% acrylamide) of 2 µg and 4 µg of RBC ASH1L-[BRD] (His). MW markers (left) are, from top, 220, 160, 120, 100, 90, 80, 70, 60, **50**, 40, 30, 25, **20**, 15, 10 kDa.



Differential Scanning Fluorimetry of RBC ASH1L-[BRD] (His) in the Presence or Absence of Common Bromodomain Ligands. Thermal denaturation of ASH1L-[BRD] (His) (0.2 mg/mL) is detected (CFX384 TMTouch thermal cycler, 'FRET' channel; Bio- Rad) by increased binding and fluorescence of the dye SYPRO®Orange (Life Technologies). Shown are the first derivatives of fluorescence vs. temperature plots in which the "peaks" indicate the Tm's (inflection points) of the original plots. None of the added ligands (JQ1, PFI1, CBP112, Bromosporine, SGC-CBP30, BET151 or RVX-208; all 25 µM) stabilized the protein folding, as indicated by the absence of any substantial shift in the Tm from that of the solvent control (57°C, 0.25% DMSO).

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