REACTION BIOLOGY

PRODUCT DATASHEET

BRD3-2 (GST) (Bromodomain containing protein 3 (RING3L), bromodomain 2)

CATALOG NO.: RD-11-164 **LOT NO.:**

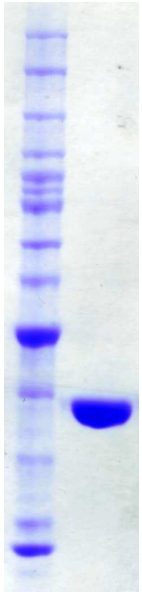
DESCRIPTION: Human recombinant BRD3, bromodomain-2 (residues 306-416; Genbank Accession # NM_007371; MW = 40.1 kDa) expressed as an N-terminal GST fusion protein in *E. coli*. BRD3, like other human members of the BET family of chromatin-binding proteins (BRD2, BRD4, BRDT), comprises two bromodomains (see reviews[1,2]), protein modules that bind ε-*N*-acetyllysine residues[3,4]. Recent results suggest an important role for BRD3 in linking acetylation of both histones and non-histone proteins to gene transcription. When overexpressed in 293 cells, BRD3, along with BRD2, binds the hyperacetylated chromatin of transcribed genes, regions enriched in acetylated histone H4 lysine-5 (H4K5Ac), H4K12Ac, H3K14Ac, but deficient in H4K16Ac and H3K9me[5]. In an *in vitro* RNA polymerase II transcription system, binding of either BRD3 or BRD2 to a chromatin template assembled with hyperacetylated histones enabled transcription through the nucleosomes[5]. Although BRD3-2 has been shown to bind tetracetylated histone H4 tail sequence (H4K5AcK8AcK12AcK16Ac), the interaction is weaker than that of BRD3-1[6]. The BET family inhibitor, I-BET151, has shown efficacy in mouse models of MLL-fusion leukemias, displacing BRD3 and BRD4 from chromatin and inhibiting transcription of genes, e.g. *BCL2*, upregulated by the MLL-fusions[7].

PURITY: >95% by SDS-PAGE

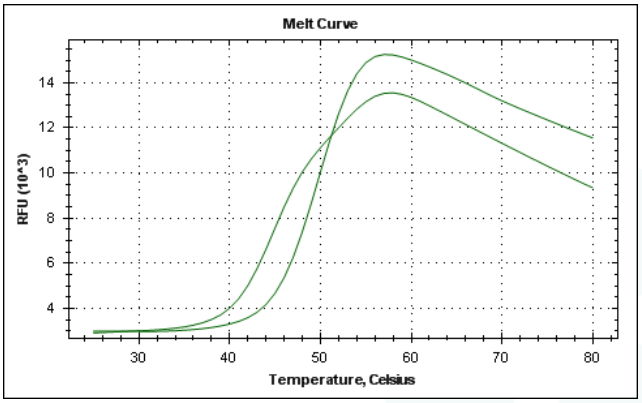
SUPPLIED AS: _ µg/µL in 20 mM Tris/HCl, pH 7.5, 150 mM NaCl, 1.0 mM TCEP, 10% glycerol (w/v) as determined by OD_{280}

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 enzyme is not recommended.

REFERENCES: 1) B. Florence & D.V. Faller *Front. Biosci.* 2001 **6** D1008; 2) S.-Y. Wu & C.-M. Chiang *J. Biol. Chem.* 2007 **282** 13141; 3) D.J. Owen *et al. EMBO J.* 2000 **19** 6141; 4)) L. Zeng & M.-M. Zhou *FEBS Lett.* 2002 **513** 124; 5) G. LeRoy *et al. Mol. Cell* 2008 **30** 51; 6) J.M. Lamonica *et al. Proc. Natl. Acad. Sci. USA* 2011 **108** E159; 7) M.A. Dawson *et al. Nature* 2011 **478** 529



Coomassie blue stained SDS-PAGE (4-12% acrylamide) of 3 µg of RBC BRD3-2 (GST). MW markers (left lane) are, from top, 220, 160, 120, 100, 90, 80, 70, 60, 50, 40, 30, 25, 20, 15 & 10 kDa.



Differential Scanning Fluorimetry of RBC BRD3-2 (GST) in Presence or Absence of (+)-JQ1. Thermal denaturation of BRD3-2 (GST) is detected (CFX384™ Touch thermal cycler, 'FRET' channel; Bio-Rad) by increased binding and fluorescence of the dye SYPRO® Orange (Life Technologies). Addition of the BET bromodomain inhibitor/ligand (+)-JQ1 (10 µM) stabilizes the protein folding and shifts the T_m (inflection point) from 45°C to 50°C.

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