#12624 Store at -20° Btk (D3H5) Rabbit mAb (Biotinylated)

100 μl (10 western blots)

New 05/13

For Research Use Only. Not For Use In Diagnostic Procedures.

Applications W Endogenous	Species Cross-Reactivity* H, M, (R, Hm, Mk, B, Dg, Pg, Hr)	Molecular Wt. 77 kDa	lsotype Rabbit IgG	
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Description: This Cell Signaling Technology antibody is conjugated to biotin under optimal conditions. The biotinylated antibody is expected to exhibit the same species cross-reactivity as the unconjugated Btk (D3H5) Rabbit mAb #8547.

Background: Bruton's tyrosine kinase (Btk) is a member of the Btk/Tec family of cytoplasmic tyrosine kinases. Like other Btk family members, it contains a pleckstrin homology (PH) domain and Src homology SH3 and SH2 domains. Btk plays an important role in B cell development (1.2). Activation of B cells by various ligands is accompanied by Btk membrane translocation mediated by its PH domain binding to phosphatidylinositol-3,4,5-trisphosphate (3-5). The membrane-localized Btk is active and associated with transient phosphorylation of two tyrosine residues, Tyr551 and Tyr223. Tyr551 in the activation loop is transphosphorylated by the Src family tyrosine kinases, leading to autophosphorylation at Tyr223 within the SH3 domain, which is necessary for full activation (6,7). The activation of Btk is negatively regulated by PKC_B through phosphorylation of Btk at Ser180, which results in reduced membrane recruitment, transphosphorylation, and subsequent activation (8). The PKC inhibitory signal is likely to be a key determinant of the B cell receptor signaling threshold to maintain optimal Btk activity (8).

Specificity/Sensitivity: Btk (D3H5) Rabbit mAb (Biotinylated) recognizes endogenous levels of total Btk protein.

Source/Purification: Monoclonal antibody is produced by immunizing animals with a synthetic peptide corresponding to residues surrounding Asp195 of human Btk protein.



Western blot analysis of Jurkat and Ramos cell extracts using Btk (D3H5) Rabbit mAb (Biotinylated).

Entrez-Gene ID #695 Swiss-Prot Acc. #Q06187

Storage: Supplied in 10 mM sodium HEPES (pH 7.5), 150 mM NaCl, 100 µg/ml BSA, 50% glycerol and less than 0.02% sodium azide. Store at -20°C. Do not aliquot the antibody.

🖗 Cell Signaling

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*Biotinylated antibodies are designed to be detected using streptavidin or anti-biotin antibody conjugates.

Recommended Antibody Dilutions: Western blotting

1:1000

For product specific protocols please see the web page for this product at www.cellsignal.com.

Please visit www.cellsignal.com for a complete listing of recommended complementary products.

Background References:

(1) Khan, W.N. (2001) Immunol. Res. 23, 147-156.

(2) Lewis, C.M. et al. (2001) Curr. Opin. Immunol. 13, 317-325.

(3) Salim, K. et al. (1996) EMBO J. 15, 6241-6250.

(4) Rameh, L.E. et al. (1997) J. Biol. Chem. 272, 22059-22066.

(5) Varnai, P. et al. (1999) J. Biol. Chem. 274, 10983-10989.

(6) Rawlings, D.J. et al. (1996) Science 271, 822-825.

(7) Park, H. et al. (1996) Immunity 4, 515-525.

(8) Kang, S.W. et al. (2001) EMBO J. 20, 5692-5702.

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IMPORTANT: For western blots, incubate membrane with diluted antibody in 5% w/v BSA, 1X TBS, 0.1% Tween-20 at 4°C with gentle shaking, overnight.

ChIP—Chromatin Immunoprecipitation IF-Immunofluorescence Applications Key: W—Western IP—Immunoprecipitation IHC—Immunohistochemistry F-Flow cytometry E-P-ELISA-Peptide Species Cross-Reactivity Key: H—human M—mouse R—rat Hm—hamster Mk—monkey Mi—mink C—chicken Dm—D. melanooaster X—Xenopus Z—zebrafish B—bovine Dg—dog Pg—pig Sc—S. cerevisiae Ce—C. elegans Hr—horse All-all species expected Species enclosed in parentheses are predicted to react based on 100% homology.