bs-12578R **[Primary Antibody]**

phospho-Bcl2 (Thr69) Rabbit pAb

www.bioss.com.cn
sales@bioss.com.cn
techsupport@bioss.com.cn
400-901-9800

DATASHEET

Host: Rabbit **Isotype:** IgG

Clonality: Polyclonal

GeneID: 596 **SWISS:** P10415

Target: Bcl2 (Thr69)

Immunogen: KLH conjugated Synthesised phosphopeptide derived from human Bcl-2 around the phosphorylation site of Thr69: AR(p-T)SP.

Purification: affinity purified by Protein A

Concentration: 1mg/ml

Storage: 0.01M TBS (pH7.4) with 1% BSA, 0.02% Proclin300 and 50% Glycerol.
Shipped at 4°C. Store at -20°C for one year. Avoid repeated freeze/thaw cycles.

Background: The Bcl-2 gene was isolated at the chromosomal breakpoint of t(14;18)-bearing follicular B cell lymphomas(1,2).Bcl-2 blocks cell death following a variety of stimuli and confers a death-sparing effect to certain hematopoietic cell lines following growth factor withdrawal (3,5).Bcl-2 appears to function in several subcellular locations yet lacks any known motifs that would confer insight into its mechanism of action (6,7).A more recently identified protein,designated Bax p21(i.e., Bcl-associated X protein),has extensive amino acid homology with Bcl-2 and both homodimerizes and forms heterodimers with Bcl-2(8). Overexpression of Bax accelerates apoptotic death induced by cytokine deprivation in an IL-3 dependent cell line and Bax also counters the death repressor activty of Bcl-2(8).

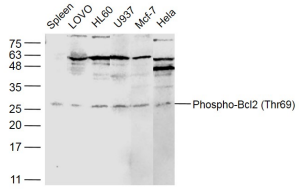
Applications: **WB** (1:500-2000)
IHC-P (1:100-500)
IHC-F (1:100-500)
IF (1:100-500)
Flow-Cyt (1μg/Test)

Reactivity: Human, Mouse, Rat

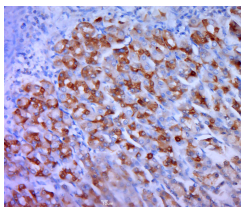
Predicted MW.: 26 kDa

Subcellular Location: Cell membrane ,Cytoplasm ,Nucleus

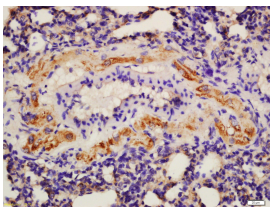
VALIDATION IMAGES



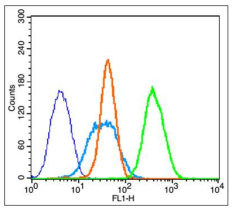
Sample: Spleen (Mouse) Lysate at 40 ug LOVO Cell (Human) Lysate at 40 ug HL60 Cell (Human) Lysate at 40 ug U937 Cell (Human) Lysate at 40 ug MCF-7 Cell (Human) Lysate at 40 ug Hela Cell (Human) Lysate at 40 ug Primary: Anti- Phospho-Bcl2 (Thr69) (bs-12578R) at 1/300 dilution Secondary: IRDye800CW Goat Anti-Rabbit IgG at 1/20000 dilution Predicted band size: 26 kD Observed band size: 26 kD



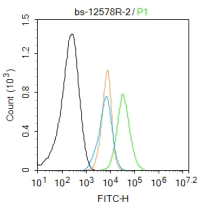
Paraformaldehyde-fixed, paraffin embedded (rat stomach tissue); Antigen retrieval by boiling in sodium citrate buffer (pH6.0) for 15min; Block endogenous peroxidase by 3% hydrogen peroxide for 20 minutes; Blocking buffer (normal goat serum) at 37°C for 30min; Antibody incubation with (P-Bcl2 (Thr69)) Polyclonal Antibody, Unconjugated (bs-12578R) at 1:400 overnight at 4°C, followed by a conjugated secondary (sp-0023) for 20 minutes and DAB staining.



Tissue/cell: rat lung tissue; 4% Paraformaldehyde-fixed and paraffin-embedded; Antigen retrieval: citrate buffer (0.01M, pH 6.0), Boiling bathing for 15min; Block endogenous peroxidase by 3% Hydrogen peroxide for 30min; Blocking buffer (normal goat serum,C-0005) at 37°C for 20 min; Incubation: Anti-Phospho-Bcl2 (Thr69) Polyclonal Antibody, Unconjugated(bs-12578R) 1:200, overnight at 4°C, followed by conjugation to the secondary antibody(SP-0023) and DAB(C-0010) staining



Blank control(blue):K562 (fixed with 2%



Blank control:HL-60. Primary Antibody (green

paraformaldehyde for 10 min at 37°C). Primary Antibody:Rabbit Anti-Phospho-Bcl2 (Thr69) antibody (bs-12578R,Green); Dilution: 1µg in 100 µL 1X PBS containing 0.5% BSA; Isotype Control Antibody: Rabbit IgG(orange) ,used under the same conditions; Secondary Antibody: Goat anti-rabbit IgG-FITC(white blue), Dilution: 1:200 in 1 X PBS containing 0.5% BSA.

line): Rabbit Anti-Phospho-Bcl2 (Thr69) antibody (bs-12578R) Dilution: 1µg /10^6 cells; Isotype Control Antibody (orange line): Rabbit IgG . Secondary Antibody : Goat anti-rabbit IgG-AF488 Dilution: 1µg /test. Protocol The cells were fixed with 4% PFA (10min at room temperature)and then permeabilized with 0.1%PBST for 20 min at room temperature. The cells were then incubated in 5%BSA to block non-specific protein-protein interactions for 30 min at room temperature .Cells stained with Primary Antibody for 30 min at room temperature. The secondary antibody used for 40 min at room temperature. Acquisition of 20,000 events was performed.