

# **Human BMI-1 Antibody**

Monoclonal Mouse IgG<sub>2A</sub> Clone # 384515 Catalog Number: MAB33341

DESCRIPTION			
Species Reactivity	Human		
Specificity	Detects human BMI-1 in direct ELISAs and Western blots.		
Source	Monoclonal Mouse IgG <sub>2A</sub> Clone # 384515		
Purification	Protein A or G purified from hybridoma culture supernatant		
Immunogen	E. coli-derived recombinant human BMI-1 Asp96-Gly326 Accession # P35226		
Formulation	Lyophilized from a 0.2 μm filtered solution in PBS with Trehalose. See Certificate of Analysis for details. *Small pack size (-SP) is supplied either lyophilized or as a 0.2 μm filtered solution in PBS.		

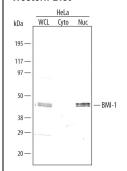
#### **APPLICATIONS**

Please Note: Optimal dilutions should be determined by each laboratory for each application. General Protocols are available in the Technical Information section on our website.

	Recommended Concentration	Sample
Western Blot	0.1 μg/mL	See Below
Chromatin Immunoprecipitation (ChIP)	5 μg/5 x 10 <sup>6</sup> cells	See Below
Intracellular Staining by Flow Cytometry	2.5 µg/10 <sup>6</sup> cells	See Below
CyTOF-ready	Ready to be labeled using established conjugation methods. No BSA or other carrier proteins that could interfere with conjugation.	

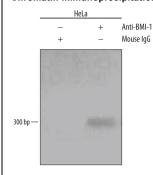
#### DATA

#### Western Blot

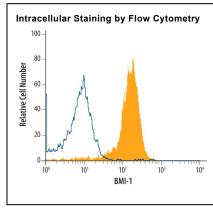


Detection of Human BMI-1 by Western Blot. Western blot shows lysates of HeLa human cervical epithelial carcinoma cell line. Gels were loaded with 30 μg of whole cell lysate (WCL), 20 μg of cytoplasmic (Cyto), and 10 μg of nuclear extracts (Nuc). PVDF membrane was probed with 0.1 μg/mL Mouse Anti-Human BMI-1 Monoclonal Antibody (Catalog # MAB33341) followed by HRP-conjugated Anti-Mouse IgG Secondary Antibody (Catalog # HAF007). A specific band for BMI-1 was detected at approximately 45 kDa (as indicated). This experiment was conducted under reducing conditions and using Immunoblot Buffer Group 4.

#### Chromatin Immunoprecipitation (ChIP)



Detection of BMI-1-regulated Genes by Chromatin Immunoprecipitation. HeLa human cervical epithelial carcinoma cell line was fixed using formaldehyde, resuspended in lysis buffer, and sonicated to shear chromatin, BMI-1/DNA complexes were immunoprecipitated using 5 µg Mouse Anti-Human BMI-1 Monoclonal Antibody (Catalog # MAB33341) or control antibody (Catalog # MAB003) for 15 minutes in an ultrasonic bath, followed by Biotinylated Anti-Mouse IgG Secondary Antibody (Catalog # BAF007). Immunocomplexes were captured using 50 µL of MagCellect Streptavidin Ferrofluid (Catalog # MAG999) and DNA was purified using chelating resin solution. The hoxc13 promoter was detected by standard PCR.



Detection of BMI-1 in HeLa **Human Cell Line by Flow** Cytometry. HeLa human cervical epithelial carcinoma cell line was stained with Mouse Anti-Human BMI-1 Monoclonal Antibody (Catalog # MAB33341, filled histogram) or isotype control antibody (Catalog # MAB003, open histogram), followed by Phycoerythrin-conjugated Anti-Mouse IgG F(ab')<sub>2</sub> Secondary Antibody (Catalog # F0102B). To facilitate intracellular staining, cells were fixed with paraformaldehyde and permeabilized with saponin.

### PREPARATION AND STORAGE

**Reconstitution** Reconstitute at 0.5 mg/mL in sterile PBS

The product is shipped at ambient temperature. Upon receipt, store it immediately at the temperature recommended below.

\*Small pack size (-SP) is shipped with polar packs. Upon receipt, store it immediately at -20 to -70 °C

### Stability & Storage

Shipping

#### Use a manual defrost freezer and avoid repeated freeze-thaw cycles.

- 12 months from date of receipt, -20 to -70 °C as supplied.
- 1 month, 2 to 8 °C under sterile conditions after reconstitution.
- 6 months, -20 to -70 °C under sterile conditions after reconstitution.

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### BACKGROUND

BMI-1 (B cell-specific Moloney-MLV integration site #1) is a 45 kDa protooncogene that is a class II member of the Polycomb group of genes. It participates in the formation of a large multimeric complex termed PRC1 that inhibits target gene transcription. Loss of BMI-1 function precludes stem cells from self-replicating. Human BMI-1 contains an N-terminal RING-finger domain (aa 17-56), an NLS (aa 81-95) and a C-terminal Pro/Ser-rich region (aa 251-326). Human BMI-1 shares 99%, 97%, 99% and 99% aa sequence identity with bovine, mouse, feline and canine BMI-1, respectively.

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