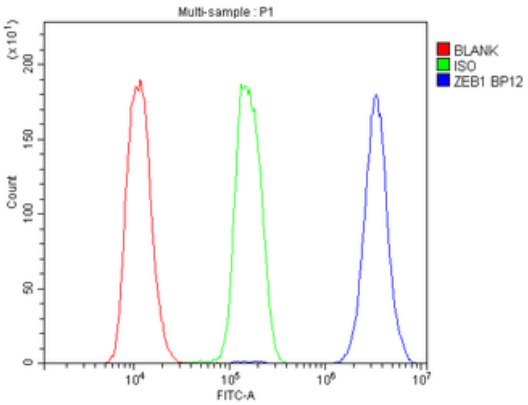
Product Datasheet

Anti-AREB6/ZEB1 Antibody (orb570306)

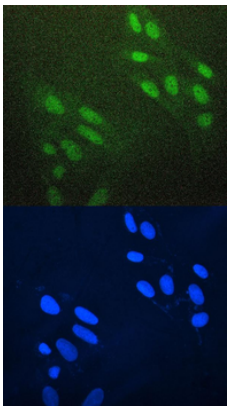
Description	Anti-AREB6/ZEB1 Antibody
Species/Host	Rabbit
Reactivity	Human
Conjugation	Unconjugated
Tested Applications	FC, ICC, IF, IHC, WB
Immunogen	A synthetic peptide corresponding to a sequence in the middle region of human ZEB1, which shares 94.9% and 100% amino acid (aa) sequence identity with rat ZEB1.
Form/Appearance	Lyophilized
Concentration	Adding 0.2 ml of distilled water will yield a concentration of 500 µg/ml.
Storage	Maintain refrigerated at 2-8°C for up to 2 weeks. For long term storage store at -20°C in small aliquots to prevent freeze-thaw cycles.
Note	For research use only
Application notes	Western blot, 0.25-0.5µg/ml, Human Immunohistochemistry (Paraffin-embedded Section), 0.5-1µg/ml, Human Immunocytochemistry/Immunofluorescence, 2µg/ml, Human Flow Cytometry (Fixed), 1-3µg/1x106 cells, Human. Add 0.2ml of distilled water will yield a concentration of 500ug/ml
Isotype	Rabbit IgG
Clonality	Polyclonal
MW	200 kDa
Uniprot ID	P37275
Expiration Date	12 months from date of receipt.

Biorbyt Ltd.
7 Signet Court, Swann's Road,
Cambridge, CB5 8LA, United Kingdom
Email: info@biorbyt.com, support@biorbyt.com
Phone: +44 (0) 1223 859-353 | Fax: +1 (415) 651-8558

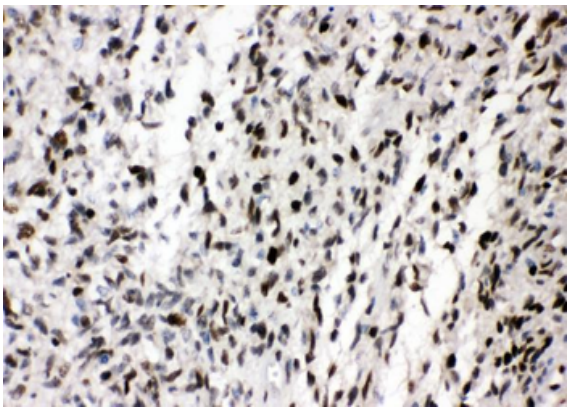
Biorbyt LLC.
68 TW Alexander Drive,
Durham, NC, 27713, United States
Email: info@biorbyt.com, support@biorbyt.com
Phone: +1 (415) 906-5211 | Fax: +1 (415) 651-8558



Flow Cytometry analysis of K562 cells using anti-ZEB1 antibody. Overlay histogram showing K562 cells (Blue line). To facilitate intracellular staining, cells were fixed with 4% paraformaldehyde and permeabilized with permeabilization buffer. The cells were blocked with 10% normal goat serum. And then incubated with rabbit anti-ZEB1 Antibody (1 µg/1x10^6 cells) for 30 min at 20°C. DyLight®488 conjugated goat anti-rabbit IgG (5-10 µg/1x10^6 cells) was used as secondary antibody for 30 minutes at 20°C. Isotype control antibody (Green line) was rabbit IgG (1 µg/1x10^6) used under the same conditions. Unlabelled sample (Red line) was also used as a control.



IF analysis of ZEB1 using anti-ZEB1 antibody ZEB1 was detected in immunocytochemical section of U20S cell. Enzyme antigen retrieval was performed using IHC enzyme antigen retrieval reagent for 15 mins. The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2 µg/mL rabbit anti-ZEB1 Antibody overnight at 4°C. DyLight488 Conjugated Goat Anti-Rabbit IgG was used as secondary antibody at 1:100 dilution and incubated for 30 minutes at 37°C. The section was counterstained with DAPI. Visualize using a fluorescence microscope and filter sets appropriate for the label used.



IHC analysis of ZEB1 using anti-ZEB1 antibody. ZEB1 was detected in paraffin-embedded section of human glioma tissues. Heat mediated antigen retrieval was performed in citrate buffer (pH6, epitope retrieval solution) for 20 mins. The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1 µg/ml rabbit anti-ZEB1 Antibody overnight at 4°C. Biotinylated goat anti-rabbit IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using Strepavidin-Biotin-Complex (SABC) with DAB as the chromogen.

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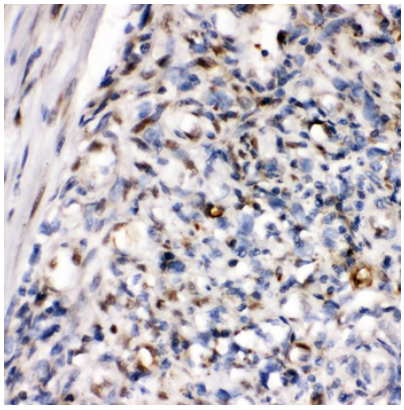
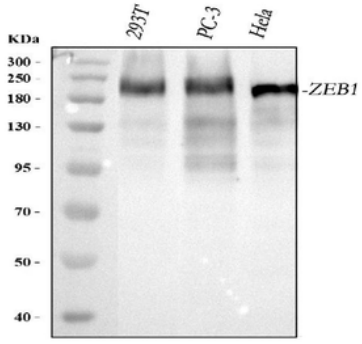


IHC analysis of ZEB1 using anti-ZEB1 antibody. ZEB1 was detected in paraffin-embedded section of human melanoma tissues. Heat mediated antigen retrieval was performed in citrate buffer (pH6, epitope retrieval solution) for 20 mins. The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1 µg/ml rabbit anti-ZEB1 Antibody overnight at 4°C. Biotinylated goat anti-rabbit IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using Strepavidin-Biotin-Complex (SABC) with DAB as the chromogen.



Western blot analysis of ZEB1 using anti-ZEB1 antibody. Electrophoresis was performed on a 5-20% SDS-PAGE gel at 70V (Stacking gel) / 90V (Resolving gel) for 2-3 hours. The sample well of each lane was loaded with 30 ug of sample under reducing conditions. Lane 1: human 293T whole cell lysates, Lane 2: human PC-3 whole cell lysates, Lane 3: human HeLa whole cell lysates. After electrophoresis, proteins were transferred to a nitrocellulose membrane at 150 mA for 50-90 minutes. Blocked the membrane with 5% non-fat milk/TBS for 1.5 hour at RT. The membrane was incubated with rabbit anti-ZEB1 antigen affinity purified polyclonal antibody at 0.5 µg/mL overnight at 4°C, then washed with TBS-0.1% Tween 3 times with 5 minutes each and probed with a goat anti-rabbit IgG-HRP secondary antibody at a dilution of 1:5000 for 1.5 hour at RT. The signal is developed using an Enhanced Chemiluminescent detection (ECL) kit with Tanon 5200 system. A specific band was detected for ZEB1 at approximately 200 kDa. The expected band size for ZEB1 is at 124 kDa.

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