Product Datasheet

TUBA1B Antibody (orb345511)

Catalog Number	orb345511
Category	Antibodies
Description	Alpha-Tubulin antibody
Target	TUBA1B
Clonality	Polyclonal
Species/Host	Rabbit
Isotype	IgG
Conjugation	Unconjugated
Reactivity	Human, Mouse, Rat
Form/Appearance	Liquid (sterile filtered)
Concentration	1.1 mg/mL
Buffer/Preservatives	Preservative: 0.01% (w/v) Sodium Azide. Stabilizer: None; Buffer: 0.02 M Potassium Phosphate, 0.15 M Sodium Chloride, pH 7.2
Purity	Anti-Tubulin Loading Control Antibody is directed against human alpha Tubulin protein. The Loading Control Antibody was affinity purified from monospecific antiserum by immunoaffinity purification. A BLAST analysis was used to suggest that this antibody would react with alpha Tubulin from a wide range of organisms, including avian, mammalian aquatic, parasitic and alga sources based on 100% homology for the immunogen sequence. Cross reactivity will occur with all isoforms of alpha tubulin. Such broad reactivity makes this antibody useful as an excellent loading control.

Biorbyt Ltd.
7 Signet Court, Swann Road
Cambridge
CB5 8LA
United Kingdom
Email: info@biorbyt.com, support@biorbyt.com
Phone: +44 (0)1223 859353 | Fax: +1 (415) 651-8558

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

Immunogen	Anti-Tubulin Loading Control Antibody was prepared from whole rabbit serum produced by repeated immunizations with a synthetic peptide corresponding to amino acids 427-441 of Human alpha Tubulin.
UniProt ID	**P68363**
Tested applications	ELISA, IF, IHC, WB
Dilution range	ELISA: 1:5,000, IHC: 1:500 - 1:2,000, IF: 1:500 - 1:2,000, WB: 1:500 - 1:3,000
Application notes	Anti-Tubulin Antibody has been tested for use in ELISA, immunofluorescence, and western blot. Specific conditions for reactivity should be optimized by the end user. Expect a band at ~50 kDa in size corresponding to alpha tubulin by western blotting in most cell lysates or extracts.
Antibody Type	Primary Antibody
Storage	Store vial at -20° C or below prior to opening. This vial contains a relatively low volume of reagent (25 µL). To minimize loss of volume dilute 1:10 by adding 225 µL of the buffer stated above directly to the vial. Recap, mix thoroughly and briefly centrifuge to collect the volume at the bottom of the vial. Use this intermediate dilution when calculating final dilutions as recommended below. Store the vial at -20°C or below after dilution. Avoid cycles of freezing and thawing.
Dry Ice Shipping	**Please note: This product requires shipment on dry ice. A dry ice surcharge will apply.**
Note	For research use only
NCBI	**17986283**
Expiration Date	12 months from date of receipt.

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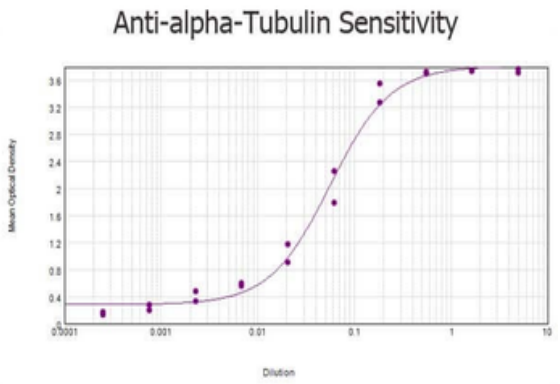
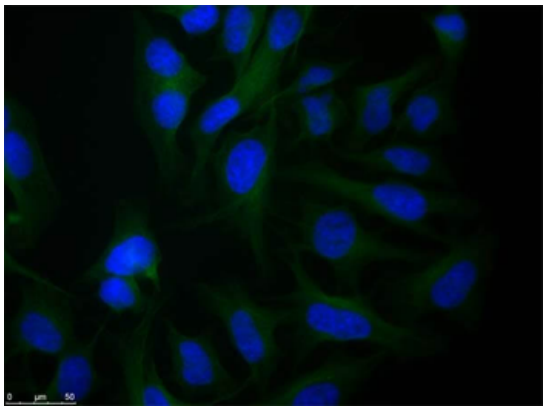


ELISA results of purified Rabbit anti-alpha-Tubulin Antibody tested against BSA-conjugated peptide of immunizing peptide. Each well was coated in duplicate with 0.1 µg of conjugate. The starting dilution of antibody was 5 µg/ml and the X-axis represents the Log10 of a 3-fold dilution. This titration is a 4-parameter curve fit where the IC50 is defined as the titer of the antibody. Assay performed using 3% fish gel, Goat anti-Rabbit IgG Antibody Peroxidase Conjugated (Min X Bv Ch Gt GP Ham Hs Hu Ms Rt & Sh Serum Proteins) (p/n orb347654) and TMB ELISA Peroxidase Substrate (p/n orb348651).



Immunofluorescence microscopy of Rabbit Anti-alpha-Tubulin antibody using HeLa cells fixed with PFA. Anti-alpha-Tubulin Antibody was used at 1 µg/ml, O/N at 4^{o}C. Secondary antibody: Anti-RABBIT IgG DyLight™ 488 Conjugated Preadsorbed at 2 ug/ml for 1 h at RT. Localization: TUBA1B is the major constituent of microtubules in the cytoplasm. Staining: Tubulin as green fluorescent signal with DAPI (blue) nuclear counterstain.

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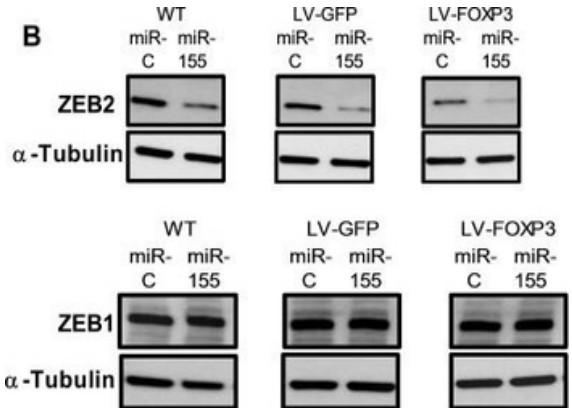

miR-155 and FOXP3 down regulate endogenous ZEB2 in human breast cancer cells resulting in altered levels of EMT markers Vimentin and E-cadherin(A) Relative abundance of ZEB2 and ZEB1 protein in WT, GFP or FOXP3 overexpressing BT549 cells transfected with miR-155 or miR-control. Relative abundance of protein was determined by quantitation of the abundance of ZEB2 or ZEB1 proteins normalised to reference protein α-Tubulin by western blot analysis. Quantitation of bands was carried out using Image J software. Mean + SD plotted. Student's t test ***P 0.001. ZEB1 protein expression as above. n = 3 experiments. (B) ZEB2 and ZEB1 protein in WT, GFP or FOXP3 overexpressing BT549 cells transfected with miR-155 or miR-control by western blot. Representative western blot shown. (C) Relative abundance of Vimentin and E-cadherin protein in WT, GFP or FOXP3 overexpressing BT549 cells transfected with miR-155 or miR-control. Relative abundance of protein was determined by quantitating the abundance of E-cadherin or Vimentin proteins and normalising to reference protein β-Actin by western blot analysis. Quantitation of bands was carried out using Image J software. Mean + SD plotted. Student's t test ***P 0.001, **P 0.01. n = 3 experiments. (D) Vimentin and E-cadherin protein in WT, GFP or FOXP3 overexpressing BT549 cells transfected with miR-155 or miR-control analysed by western blot. Representative western blot shown.

Western Blot of Rabbit Anti-Alpha Tubulin Antibody. Lane 1: whole cell lysates from mouse brain (p/n orb348715). Lane 2: rat brain (p/n orb348727). Lane 3: A431 cells (p/n orb348665). Lane 4: Jurkat cells (p/n orb348674). Lane 5: HeLa cells (p/n orb348668). Load: 35 µg per lane. Primary antibody: Alpha Tubulin antibody at 1:1200 for overnight at 4°C. Secondary antibody: IRDye800™ rabbit secondary antibody at 1:10000 for 45 min at RT. Block: 5% BLOTTO (p/n orb348624) overnight at 4°C. Predicted/Observed size: ~50 kDa corresponding to alpha tubulin (arrowhead). Other band(s): none.

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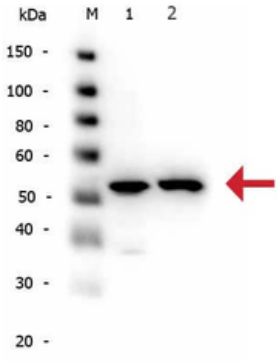
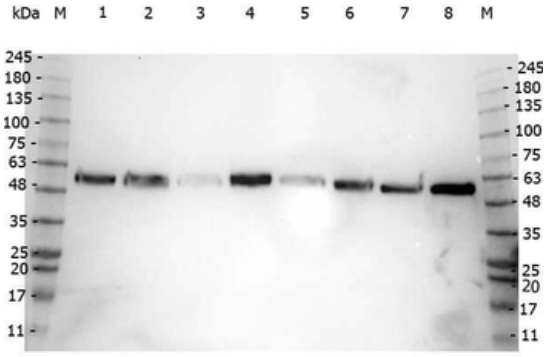


Western Blot of Rabbit anti-alpha-Tubulin antibody. Lane 1: HeLa WCL (p/n orb348668). Lane 2: NIH/3T3 WCL (p/n orb348714). Load: 10 µg per lane. Primary antibody: alpha-Tubulin antibody at 1:1000 for overnight at 4°C. Secondary antibody: Peroxidase rabbit secondary antibody (p/n orb347654) at 1:40000 for 30 min at RT. Block: Blocking Buffer for Fluorescent Western Blotting (p/n orb348637) for 30 min at RT. Predicted/Observed size: 50 kDa, 50 kDa for alpha-Tubulin. Other band(s): N/A.



Western Blot of Rabbit anti-Alpha-Tubulin antibody. Marker: Opal Pre-stained ladder. Lane 1: HEK293 lysate (p/n orb348669). Lane 2: HeLa Lysate (p/n orb348668). Lane 3: MCF-7 Lysate (p/n orb348664). Lane 4: Jurkat Lysate. Lane 5: A431 Lysate (p/n orb348665). Lane 6: LNCaP Lysate (p/n orb348694). Lane 7: A-172 Lysate (p/n orb348708). Lane 8: NIH/3T3 Lysate (p/n orb348714). Load: 35 µg per lane. Primary antibody: Alpha-Tubulin antibody at 1:2000 for overnight at 4°C. Secondary antibody: Peroxidase rabbit secondary antibody (p/n orb347654) at 1:30000 for 60 min at RT. Blocking Buffer: 1% Casein-TTBS for 30 min at RT. Predicted/Observed size: 50 kDa for Alpha-tubulin.

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