

Product Datasheet

ACTB Antibody (orb420310)

Catalog Number	orb420310
Category	Antibodies
Description	beta Actin antibody
Target	ACTB
Clonality	Polyclonal
Species/Host	Rabbit
Isotype	IgG
Conjugation	Unconjugated
Reactivity	Frog, Human, Mouse
Form/Appearance	Liquid (sterile filtered)
Concentration	1.0 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-beta-Actin Loading Control Antibody reacts with Human, Rat, Monkey and Mouse. beta-Actin Loading Control Antibody is expected to cross-react with a wide range of species due to sequence homology. Anti-Beta Actin is affinity-purified antibody is directed against human beta Actin protein. A BLAST analysis was used to suggest that this antibody would react with beta Actin from a wide range of organisms, including most vertebrates and some yeast. Broad reactivity makes this antibody an excellent loading control.

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Immunogen	beta-Actin Loading Control Antibody was prepared from whole rabbit serum produced by repeated immunizations with a synthetic peptide corresponding to a C-Terminal region near amino acids 350-375 of Human beta Actin.
UniProt ID	P60709
Tested applications	ELISA, IF, WB
Dilution range	ELISA: 1:10,000 - 1:40,000, IF: 1:500 - 1:2,000, WB: 1:1,000 - 1:4,000
Application notes	Anti-beta Actin Antibody has been tested for use in ELISA, immunofluorescence, and western blot. Specific conditions for reactivity should be optimized by the end user. Beta actin present in fibroblast connective tissue stains very brightly. Beta actin present in neuromuscular junctions also stains. Paraformaldehyde fixation yields brighter staining than formalin or methanol fixation. Expect a band at ~42 kDa in size corresponding to beta actin by western blotting in the appropriate cell lysate or extract.
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	NP_001092.1
Expiration Date	12 months from date of receipt.

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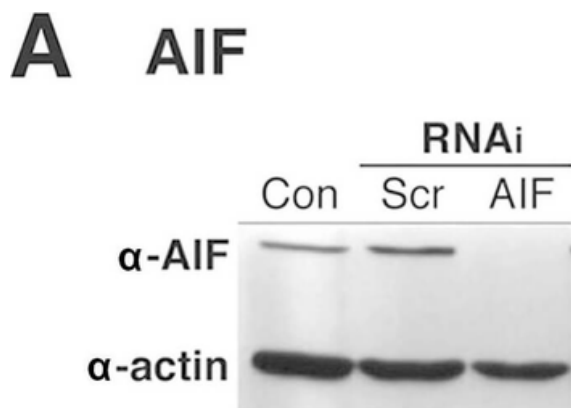
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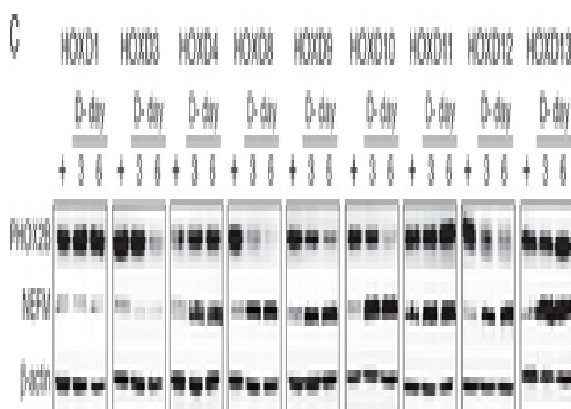
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Analysis of poly (ADP-ribose)-mediated caspase-independent cell death in breast adenocarcinoma cells after TRPM2 inhibition and chemotherapeutic treatments. Immunoblot detection of (A) apoptosis-inducing factor (AIF) and (B) poly(ADP-ribose) glycohydrolase (PARG) in MDA-MB-231 breast adenocarcinoma cells after RNAi silencing. Loading controls for immunoblots were provided by the immunodetection of β -actin. Con, untransfected cells; Scr, cells transfected with negative control scrambled siRNA oligos. (C) Quantification of cell death by flow cytometry was performed in MDA-MB-231 cells after RNAi knockdown of AIF or PARG, pretreatment with 20 μ m ACA for 30 min and treatment with 100 μ m MNNG. *P 0.05, one-way ANOVA and unpaired Student's t-test; error bars represent the SEM.



Distinct functions of HOXD proteins in regulation of neuronal differentiation. (A-B) Phase contrast imaging (A) and NEFM immunofluorescent staining (B) of BE(2)-C/Tet-Off/myc-HOXD cells cultured in the presence or absence of Doxy for 7 days. Scale bars, 50 μ m. Shown are representative of three independent experiments with similar results. (C) Immunoblot analysis of PHOX2B and NEFM protein levels in the same cell samples described in Figure 3. Beta-actin levels are shown as loading control. Shown are representative of two independent experiments with similar results. (D) Quantification of NEFM mRNA levels in BE(2)-C/Tet-Off/myc-HOXD cells cultured in the presence or absence of Doxy for 3 days. The NEFM mRNA levels in BE(2)-C/Tet-Off/myc-HOXD cells in the presence of Doxy were designated as 1.0 (dashed line). The data were from two independent samples with each being assayed in triplicates and analyzed using two-tailed Student's t-test with the p values indicated. Error bars, SD.

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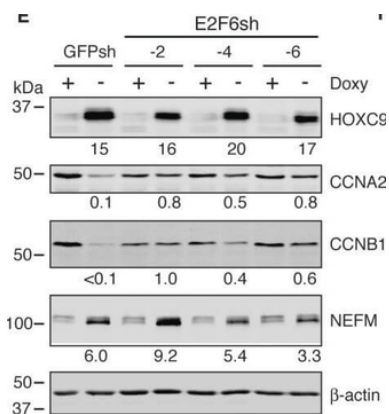
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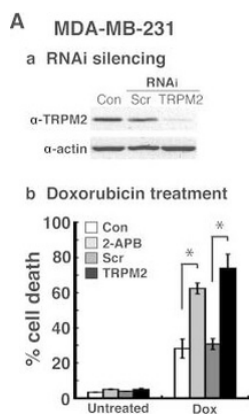
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E2F6 is essential for HOXC9 induction of G1 arrest and repression of cell cycle genes. (A) Immunoblot analysis of E2F6 levels in BE(2)-C/Tet-Off/myc-HOXC9 cells infected with lentiviruses expressing shRNA against GFP or various coding regions of E2F6. E2F6 levels were quantified against β-actin. (B-D) Phase contrast imaging and growth assay (B) and cell cycle analysis (C, D) showing E2F6 knockdown abrogated HOXC9-induced growth arrest. Error bars, SD (n = 4). (E-F) Immunoblot analysis (E) and quantification (F) showing that E2F6 knockdown abrogated HOXC9 repression of cyclins, but not HOXC9 induction of NEFM. HOXC9, CCNA2, CCNB1 and NEFM levels were quantified against β-actin with the protein levels in GFPsh-expressing cells cultured in the presence of doxycycline (Doxy+) were defined as 1.0 (dashed lines). Error bars, SD (n = 3). Data in (D) and (F) were analyzed with unpaired, two-tailed Student's t-test and p values are indicated.



Effects of TRPM2 inhibition and RNAi knockdown in cancerous and non-cancerous breast cell lines after chemotherapeutic treatments. Immunoblot levels of TRPM2 protein after TRPM2 RNAi silencing (a) and quantification of cell death by flow cytometry (b) in (A) MDA-MB-231 breast adenocarcinoma, (B) MCF-7 breast adenocarcinoma and (C) human mammary epithelial cells (HMECs), a non-cancerous primary breast cell line. For RNAi silencing, cells were transfected with 100 nM anti-TRPM2 duplex siRNA oligos (TRPM2) or 100 nM negative control scrambled duplex siRNA oligos (Scr). Loading controls for immunoblots were provided by the immunodetection of β-actin. For chemotherapeutic treatments, the cells were pretreated 30 min with the TRPM2 inhibitor, 2-APB, and then subsequently treated with (A-b) 2 μm doxorubicin (Dox) until analysis; (B-b) 7 μm tamoxifen (Tam) until analysis; and (C-b) 2 μm doxorubicin (Dox) until analysis. Con, untransfected negative control cells. All error bars represent the SEM. *P 0.05 via one-way ANOVA and unpaired Student's t-test.

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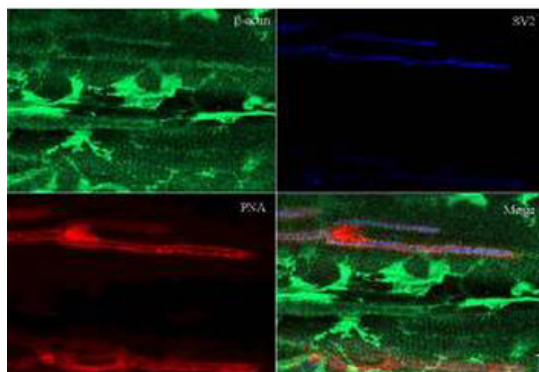
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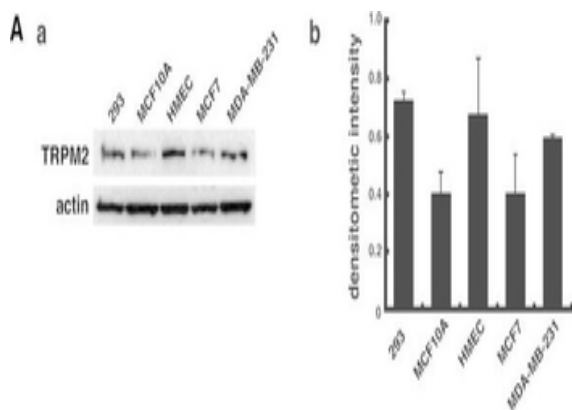
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Immunohistochemistry of Rabbit Anti-Beta Actin Antibody. Tissue: sections 4.2 mm thick of rana pipiens tissue. Fixation: formalin fixed paraffin embedded. Antigen retrieval: not required. Primary antibody: Beta Actin antibody at 1:200 for 1 h at RT. Secondary antibody: Peroxidase rabbit secondary antibody at 1:10000 for 45 min at RT. Localization: Beta Actin is at the neuromuscular junction. Staining: Beta Actin as precipitated green signal with red and blue counterstain.



TRPM2 levels in breast cell lines and knockdown of TRPM2 levels by RNAi silencing. (A) Immunoblot (a) and densitometric quantification (b) of TRPM2 protein levels in two noncancerous breast cell lines (HMEC, MCF-10A) and two cancerous breast cell lines (MCF-7, MDA-MB-231). Immunoblot detection of TRPM2 protein levels in human embryonic kidney cells (HEK 293) provided the positive control. (B) RNAi silencing of TRPM2 in noncancerous HMEC cells (a), and quantification of protein levels by densitometry (b). (C and D) RNAi silencing of TRPM2 in MCF-7 and MDA-MB-231 breast adenocarcinoma cells (a) and quantification of protein levels by densitometry (b). All densitometric values represent TRPM2:actin ratios. Con, untreated negative control cells; Scr, treatment with scrambled negative control siRNA oligos.

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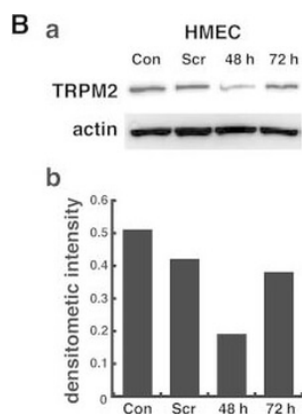
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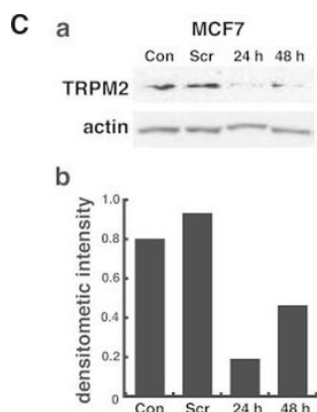
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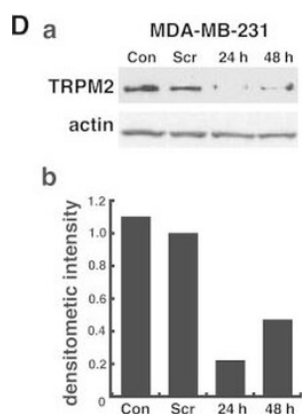
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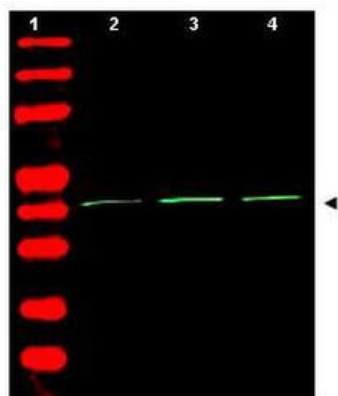
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Western Blot of Rabbit Anti-Beta Actin Antibody. Lane 1: molecular weight. Lane 2: human embryonic kidney 293 (p/n orb348669). Lane 3: human lung carcinoma A549 (p/n orb348675). Lane 4: mouse brain (p/n orb348715). Load: 35 µg per lane. Primary antibody: Beta Actin antibody at 1:1500 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: ~42 kDa corresponding to beta Actin (arrowhead). Other band(s): none.

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