

Product Datasheet

BAK/BAK1 Mouse Monoclonal Antibody (orb570309)

Catalog Number	orb570309
Category	Antibodies
Description	Anti-BAK/BAK1 Antibody (monoclonal, B6E4). Tested in Flow Cytometry, IF, IHC, ICC, WB applications. This antibody reacts with Human, Mouse, Rat.
Target	Bcl-2 homologous antagonist/killer
Clonality	Monoclonal
Species/Host	Mouse
Isotype	Mouse IgG2b
Conjugation	Unconjugated
Reactivity	Human, Mouse, Rat
Form/Appearance	Lyophilized
Concentration	500 µg/ml
Buffer/Preservatives	Each vial contains 4mg Trehalose, 0.9mg NaCl, 0.2mg Na ₂ HPO ₄ , 0.05mg Na ₃ N.
Reconstitution	Add 0.2ml of distilled water will yield a concentration of 500µg/ml.
Purification	Immunogen affinity purified.
Immunogen	E.coli-derived human BAK/BAK1 recombinant protein (Position: A22-S211). Human BAK shares 78.3 % amino acid (aa) sequence identity with mouse BAK.
UniProt ID	Q16611

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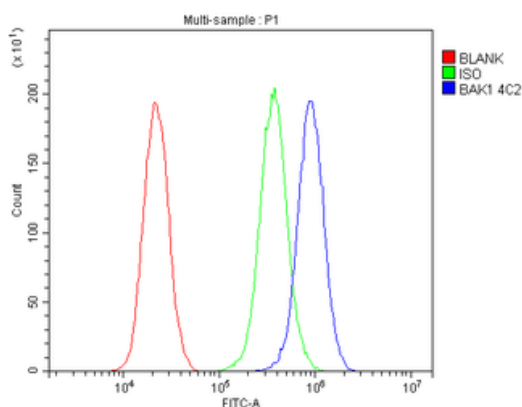
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MW	25 kDa
Tested applications	FC, ICC, IF, IHC, WB
Dilution range	Western blot, 0.1-0.5µg/ml Immunohistochemistry (Paraffin-embedded Section), 0.5-1µg/ml Immunocytochemistry/Immunofluorescence, 2µg/ml Flow Cytometry (Fixed), 1-3µg/1x10 ⁶ cells
Specificity	No cross reactivity with other proteins.
Cross Reactivity	No cross-reactivity with other proteins.
Antibody Type	Primary Antibody
Clone Number	B6E4
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
Expiration Date	12 months from date of receipt.



Flow Cytometry analysis of PC-3 cells using anti-BAK1 antibody. Overlay histogram showing PC-3 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 mouse anti-BAK1 Antibody (1 µg/1x10⁶ cells) for 30 min at 20°C. DyLight®488 conjugated goat anti-mouse IgG (5-10 µg/1x10⁶ cells) was used as secondary antibody for 30 minutes at 20°C. Isotype control antibody (Green line) was mouse IgG (1 µg/1x10⁶) used under the same conditions. Unlabelled sample without incubation with primary antibody and secondary antibody (Red line) was used as a blank control.

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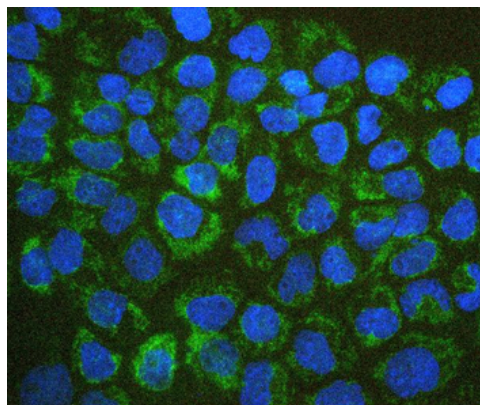
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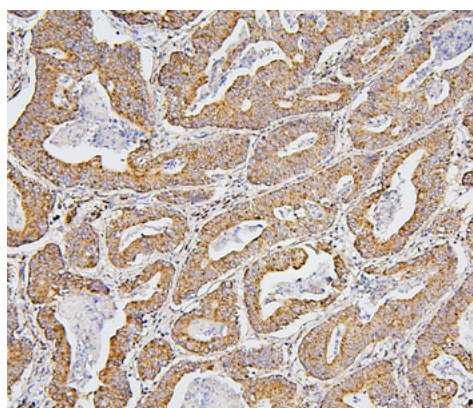
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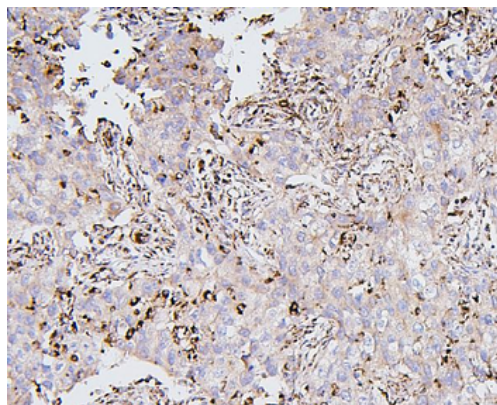
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IF analysis of BAK1 using anti-BAK1 antibody. BAK1 was detected in immunocytochemical section of A431 cell. Enzyme antigen retrieval was performed using IHC enzyme antigen retrieval reagent for 15 mins. The cells were blocked with 10% goat serum. And then incubated with 2 $\mu\text{g}/\text{mL}$ mouse anti-BAK1 Antibody overnight at 4°C. DyLight®488 Conjugated Goat Anti-mouse 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 BAK1 using anti-BAK1 antibody. BAK1 was detected in paraffin-embedded section of human intestinal cancer 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 $\mu\text{g}/\text{ml}$ mouse anti-BAK1 Antibody overnight at 4°C. Biotinylated goat anti-mouse IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using Streptavidin-Biotin-Complex (SABC) with DAB as the chromogen.



IHC analysis of BAK1 using anti-BAK1 antibody. BAK1 was detected in paraffin-embedded section of human lung cancer 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 $\mu\text{g}/\text{ml}$ mouse anti-BAK1 Antibody overnight at 4°C. Biotinylated goat anti-mouse IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using Streptavidin-Biotin-Complex (SABC) with DAB as the chromogen.

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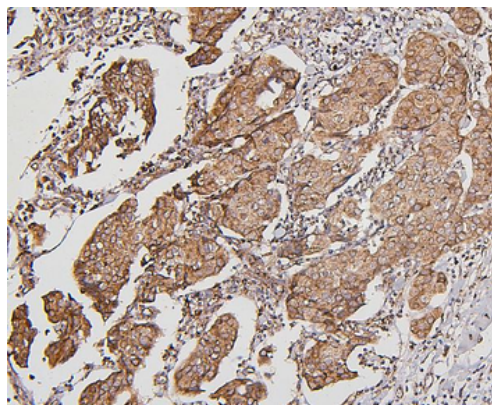
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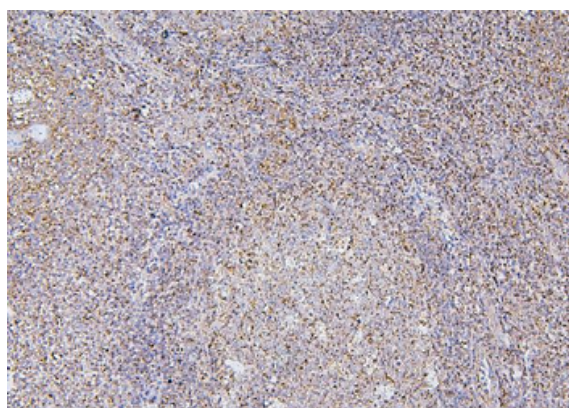
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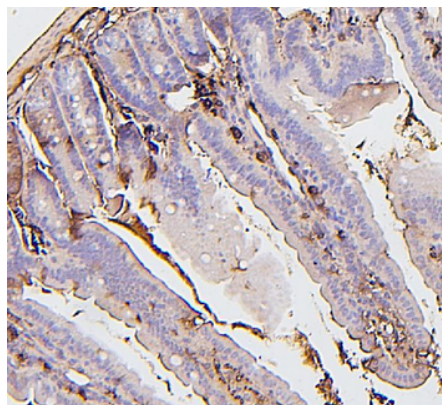
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IHC analysis of BAK1 using anti-BAK1 antibody. BAK1 was detected in paraffin-embedded section of human mammary cancer tissues. Heat mediated antigen retrieval was performed in citrate buffer (pH6, epitope retrieval solution) for 20 mins. The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1 μ g/ml mouse anti-BAK1 Antibody overnight at 4°C. Biotinylated goat anti-mouse IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using Streptavidin-Biotin-Complex (SABC) with DAB as the chromogen.



IHC analysis of BAK1 using anti-BAK1 antibody. BAK1 was detected in paraffin-embedded section of human tonsil 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 μ g/ml mouse anti-BAK1 Antibody overnight at 4°C. Biotinylated goat anti-mouse IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using Streptavidin-Biotin-Complex (SABC) with DAB as the chromogen.



IHC analysis of BAK1 using anti-BAK1 antibody. BAK1 was detected in paraffin-embedded section of mouse intestine 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 μ g/ml mouse anti-BAK1 Antibody overnight at 4°C. Biotinylated goat anti-mouse IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using Streptavidin-Biotin-Complex (SABC) with DAB as the chromogen.

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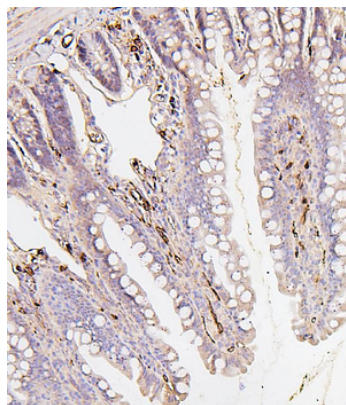
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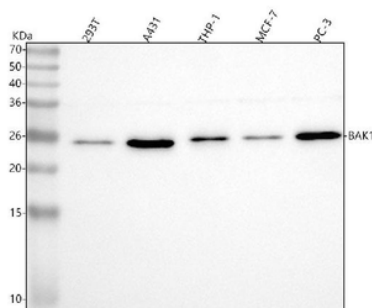
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IHC analysis of BAK1 using anti-BAK1 antibody. BAK1 was detected in paraffin-embedded section of rat intestine tissues. Heat mediated antigen retrieval was performed in citrate buffer (pH6, epitope retrieval solution) for 20 mins. The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1 μ g/ml mouse anti-BAK1 Antibody overnight at 4°C. Biotinylated goat anti-mouse IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using Streptavidin-Biotin-Complex (SABC) with DAB as the chromogen.



Western blot analysis of BAK/BAK1 using anti-BAK/BAK1 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 μ g of sample under reducing conditions. Lane 1: human 293T whole cell lysates, Lane 2: human A431 whole cell lysates, Lane 3: human THP-1 whole cell lysates, Lane 4: human MCF-7 whole cell lysates, Lane 5: human PC-3 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 mouse anti-BAK/BAK1 antigen affinity purified monoclonal antibody at 0.5 μ g/mL overnight at 4°C, then washed with TBS-0.1% Tween 3 times with 5 minutes each and probed with a goat anti-mouse IgG-HRP secondary antibody at a dilution of 1:10000 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 BAK/BAK1 at approximately 25 kDa. The expected band size for BAK/BAK1 is at 23 kDa.

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