

Anti-ANAPC2 Antibody Picoband™

Catalog Number: A06153-1

About ANAPC2

Anaphase-promoting complex subunit 2 is an enzyme that in humans is encoded by the ANAPC2 gene. A large protein complex, termed the anaphase-promoting complex (APC), or the cyclosome, promotes metaphase-anaphase transition by ubiquitinating its specific substrates such as mitotic cyclins and anaphase inhibitor, which are subsequently degraded by the 26S proteasome. Biochemical studies have shown that the vertebrate APC contains eight subunits. The composition of the APC is highly conserved in organisms from yeast to humans. The product of this gene is a component of the complex and shares sequence similarity with a recently identified family of proteins called cullins, which may also be involved in ubiquitin-mediated degradation.

Overview

Product Name	Anti-ANAPC2 Antibody Picoband™
Reactive Species	Human, Monkey, Mouse, Rat
Description	Boster Bio Anti-ANAPC2 Antibody Picoband™ catalog # A06153-1. Tested in ELISA, Flow Cytometry, IF, IHC, ICC, WB applications. This antibody reacts with Human, Monkey, Mouse, Rat.
Application	ELISA, Flow Cytometry, IF, IHC, ICC, WB
Clonality	Polyclonal
Formulation	Each vial contains 4mg Trehalose, 0.9mg NaCl and 0.2mg Na ₂ HPO ₄ .
Storage Instructions	Store at -20°C for one year from date of receipt. After reconstitution, at 4°C for one month. It can also be aliquotted and stored frozen at -20°C for six months. Avoid repeated freeze-thaw cycles.
Host	Rabbit
Uniprot ID	Q9UJX6

Technical Details

Immunogen	E.coli-derived human ANAPC2 recombinant protein (Position: K51-R272).
Recommended Detection Systems	Boster recommends Enhanced Chemiluminescent Kit with anti-Rabbit IgG (EK1002) for Western blot, and HRP Conjugated anti-Rabbit IgG Super Vision Assay Kit (SV0002-1) for IHC(P) and ICC.
Cross Reactivity	No cross-reactivity with other proteins.
Isotype	Rabbit IgG
Form	Lyophilized
Concentration	Adding 0.2 ml of distilled water will yield a concentration of 500 ug/ml.

Purification	Immunogen affinity purified.
Suggested Dilutions	Dilute the sample so that the expected range of concentrations fall within the detection range of this kit. If the expected range of concentration is unknown, a pilot test should be conducted to decide the optimal dilution ratio for your samples. Some PubMed article(s) citing the expression level of this target are as follows: Boster Bio's internal QC testing used: Western blot, 0.25-0.5ug/ml, Human, Mouse, Rat, Monkey Immunohistochemistry (Paraffin-embedded Section), 2-5ug/ml, Human Immunocytochemistry/Immunofluorescence, 5ug/ml, Human Flow Cytometry, 1-3ug/1x10⁶ cells, Human Direct ELISA, 0.1-0.5ug/ml, Human

Anti-ANAPC2 Antibody Picoband™ (A06153-1) Images

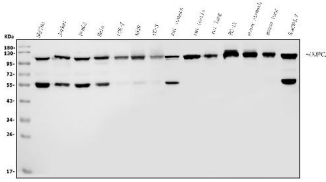


Figure 1. Western blot analysis of ANAPC2 using anti-ANAPC2 antibody (A06153-1). Electrophoresis was performed on a 5-20% SDS-PAGE gel at 70V (Stacking gel) / 90V (Resolving gel) for 2-3 hours. The sample well of each lane was loaded with 50ug of sample under reducing conditions.

Lane 1: human HEK293 whole cell lysates,
Lane 2: human Jurkat whole cell lysates,
Lane 3: human HEPG2 whole cell lysates,
Lane 4: human HELA whole cell lysates,
Lane 5: monkey COS-7 whole cell lysates,
Lane 6: human A549 whole cell lysates,
Lane 7: human PC-3 whole cell lysates,
Lane 8: rat stomach tissue lysates,
Lane 9: rat testis tissue lysates,
Lane 10: rat lung tissue lysates,
Lane 11: rat PC-12 whole cell lysates,
Lane 12: mouse stomach tissue lysates,
Lane 13: mouse lung tissue lysates,
Lane 14: mouse RAW264.7 whole cell lysates.

After Electrophoresis, proteins were transferred to a Nitrocellulose membrane at 150mA 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-ANAPC2 antigen affinity purified polyclonal antibody (Catalog # A06153-1) at 0.5 ug/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 (Catalog # EK1002) with Tanon 5200 system. A specific band was detected for ANAPC2 at approximately 110KD. The expected band size for ANAPC2 is at 94KD.

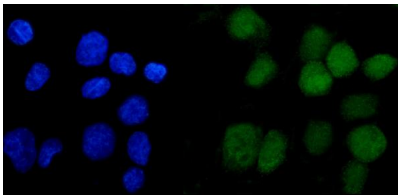
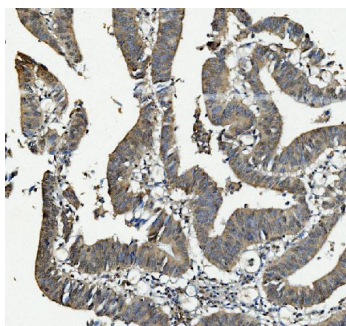


Figure 2. IF analysis of ANAPC2 using anti-ANAPC2 antibody (A06153-1).

ANAPC2 was detected in immunocytochemical section of A431 cells. Enzyme antigen retrieval was performed using IHC enzyme antigen retrieval reagent (AR0022) for 15 mins. The cells were blocked with 10% goat serum. And then incubated with 5ug/mL rabbit anti-ANAPC2 Antibody (A06153-1) overnight at 4°C. DyLight®488 Conjugated Goat Anti-Rabbit IgG (BA1127) 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.

Figure 3. IHC analysis of ANAPC2 using anti-ANAPC2 antibody (A06153-1).

ANAPC2 was detected in paraffin-embedded section of human rectal cancer tissue. Heat mediated antigen retrieval



was performed in EDTA buffer (pH8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2ug/ml rabbit anti-ANAPC2 Antibody (A06153-1) 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 Streptavidin-Biotin-Complex (SABC) (Catalog # SA1022) with DAB as the chromogen.

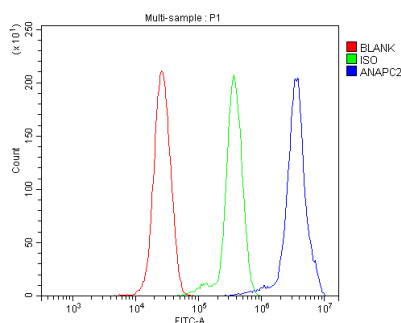


Figure 4. Flow Cytometry analysis of THP-1 cells using anti-ANAPC2 antibody (A06153-1).

Overlay histogram showing THP-1 cells stained with A06153-1 (Blue line). The cells were blocked with 10% normal goat serum. And then incubated with rabbit anti-ANAPC2 Antibody (A06153-1, 1ug/1x10⁶ cells) for 30 min at 20°C. DyLight®488 conjugated goat anti-rabbit IgG (BA1127, 5-10ug/1x10⁶ cells) was used as secondary antibody for 30 minutes at 20°C. Isotype control antibody (Green line) was rabbit IgG (1ug/1x10⁶) used under the same conditions. Unlabelled sample (Red line) was also used as a control.

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