BOSTER
antibody and ELISA experts

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Anti-Prohibitin/PHB Antibody Picoband™

Catalog Number: A01178-1

About PHB

PHB (Prohibitin), also known as PHB1, is a protein that in humans is encoded by the PHB gene. White et al. (1991) mapped the PHB gene to chromosome 17 by analysis of human-mouse somatic cell hybrid cell lines using a genomic fragment of human prohibitin DNA isolated from a library using the rat prohibitin cDNA clone. By in situ hybridization, they localized the gene to 17q21. Sato et al. (1992) isolated the human homolog of the rat prohibitin gene and mapped it to 17q12-q21 by in situ hybridization. Proliferation of tumor cells depends on new blood vessel formation (angiogenesis) that accompanies malignant progression. Anticancer therapies using angiogenesis inhibitors or cytotoxic agents targeted to the vasculature of tumors have been evaluated in clinical trials. Although white fat is a nonmalignant tissue, it has the capability to quickly proliferate and expand. Furthermore, it is highly vascularized. Rupnick et al. (2002) showed that nonspecific angiogenesis inhibitors can prevent the development of obesity of mice.

Overview

Product Name	Anti-Prohibitin/PHB Antibody Picoband™
Reactive Species	Human, Mouse, Rat
Description	Boster Bio Anti-Prohibitin/PHB Antibody Picoband™ catalog # A01178-1. Tested in ELISA, Flow Cytometry, IHC, WB applications. This antibody reacts with Human, Mouse, Rat.
Application	ELISA, Flow Cytometry, IHC, WB
Clonality	Polyclonal
Formulation	Each vial contains 4mg Trehalose, 0.9mg NaCl and 0.2mg Na2HPO4.
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	P35232

Technical Details

Immunogen	E.coli-derived human Prohibitin/PHB recombinant protein (Position: M1-I257).
Predicted Reactive Species	Human
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).
Cross Reactivity	No cross-reactivity with other proteins.
Isotype	Rabbit IgG

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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 Immunohistochemistry (Paraffin-embedded Section), 2-5ug/ml, Human, Mouse, Rat Flow Cytometry, 1-3ug/1x10^6 cells, Human, Rat Direct ELISA, 0.1-0.5ug/ml, Human

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Anti-Prohibitin/PHB Antibody Picoband™ (A01178-1) Images

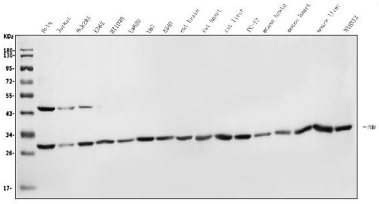

Figure 1. Western blot analysis of Prohibitin/PHB using anti-Prohibitin/PHB antibody (A01178-1).
Electrophoresis was performed on a 5-20% SDS-PAGE gel at 70V (Stacking gel) / 90V (Resolving gel) for 2-3 hours. The sample well of each lane was loaded with 30ug of sample under reducing conditions.
Lane 1: human Hela whole cell lysates,
Lane 2: human Jurkat whole cell lysates,
Lane 3: human Hek293 whole cell lysates,
Lane 4: human K562 whole cell lysates,
Lane 5: human HT1080 whole cell lysates,
Lane 6: human Sw620 whole cell lysates,
Lane 7: human U87 whole cell lysates,
Lane 8: human A549 whole cell lysates,
Lane 9: rat brain tissue lysates,
Lane 10: rat heart tissue lysates,
Lane 11: rat liver tissue lysates,
Lane 12: rat PC-12 whole cell lysates,
Lane 13: mouse brain tissue lysates,
Lane 14: mouse heart tissue lysates,
Lane 15: mouse liver tissue lysates,
Lane 16: mouse NIH/3T3 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-Prohibitin/PHB antigen affinity purified polyclonal antibody (Catalog # A01178-1) 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 Prohibitin/PHB at approximately 28-30KD. The expected band size for Prohibitin/PHB is at 28-30KD.

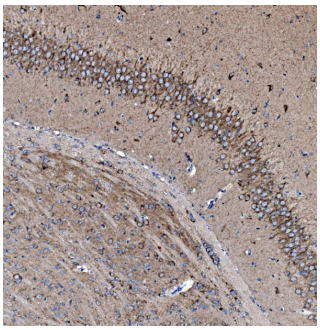

Figure 2. IHC analysis of Prohibitin/PHB using anti-Prohibitin/PHB antibody (A01178-1).
Prohibitin/PHB was detected in paraffin-embedded section of mouse brain tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2ug/ml rabbit anti-Prohibitin/PHB Antibody (A01178-1) 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 Strepavidin-Biotin-Complex (SABC) (Catalog # SA1022) with DAB as the chromogen.

Figure 3. IHC analysis of Prohibitin/PHB using anti-Prohibitin/PHB antibody (A01178-1).
Prohibitin/PHB was detected in paraffin-embedded section of

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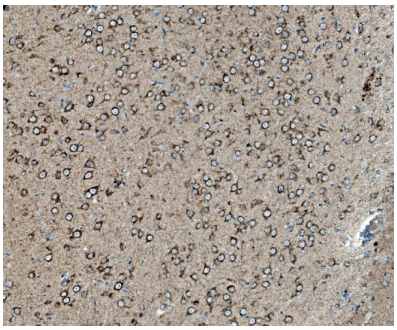

rat brain tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2ug/ml rabbit anti-Prohibitin/PHB Antibody (A01178-1) overnight at 4°C. Biotinylated goat anti-rabbit IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using Strepavidin-Biotin-Complex (SABC) (Catalog # SA1022) with DAB as the chromogen.

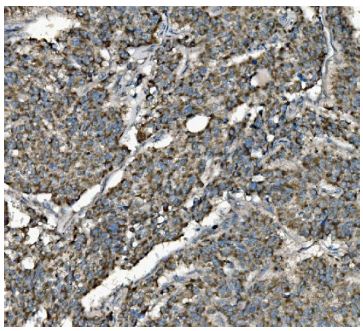

Figure 4. IHC analysis of Prohibitin/PHB using anti-Prohibitin/PHB antibody (A01178-1).
Prohibitin/PHB was detected in paraffin-embedded section of human adrenocortical adenoma tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2ug/ml rabbit anti-Prohibitin/PHB Antibody (A01178-1) 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 Strepavidin-Biotin-Complex (SABC) (Catalog # SA1022) with DAB as the chromogen.

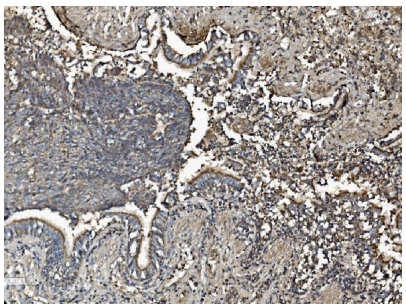

Figure 5. IHC analysis of Prohibitin/PHB using anti-Prohibitin/PHB antibody (A01178-1).
Prohibitin/PHB was detected in paraffin-embedded section of human lung cancer tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2ug/ml rabbit anti-Prohibitin/PHB Antibody (A01178-1) 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 Strepavidin-Biotin-Complex (SABC) (Catalog # SA1022) with DAB as the chromogen.

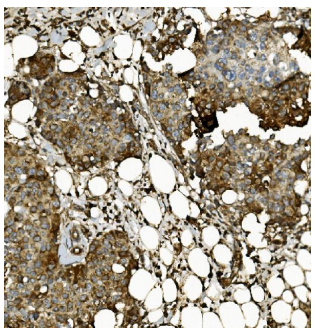

Figure 6. IHC analysis of Prohibitin/PHB using anti-Prohibitin/PHB antibody (A01178-1).
Prohibitin/PHB was detected in paraffin-embedded section of human breast cancer tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2ug/ml rabbit anti-Prohibitin/PHB Antibody (A01178-1) 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 Strepavidin-Biotin-Complex (SABC) (Catalog # SA1022) with DAB as the chromogen.

Figure 7. IHC analysis of Prohibitin/PHB using anti-Prohibitin/PHB antibody (A01178-1).
Prohibitin/PHB was detected in paraffin-embedded section of human liver cancer tissue. Heat mediated antigen retrieval

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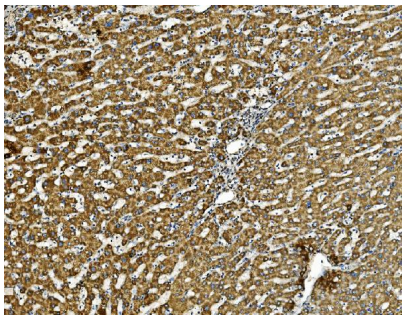

was performed in EDTA buffer (pH8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2ug/ml rabbit anti-Prohibitin/PHB Antibody (A01178-1) overnight at 4°C. Biotinylated goat anti-rabbit IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using Strepavidin-Biotin-Complex (SABC) (Catalog # SA1022) with DAB as the chromogen.

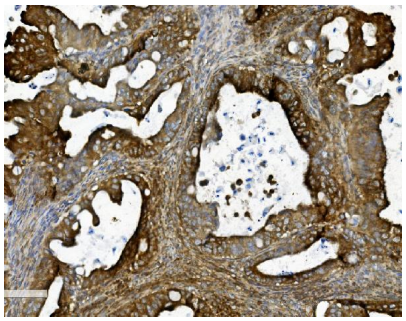

Figure 8. IHC analysis of Prohibitin/PHB using anti-Prohibitin/PHB antibody (A01178-1).
Prohibitin/PHB was detected in paraffin-embedded section of human ovarian cancer tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2ug/ml rabbit anti-Prohibitin/PHB Antibody (A01178-1) 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 Strepavidin-Biotin-Complex (SABC) (Catalog # SA1022) with DAB as the chromogen.

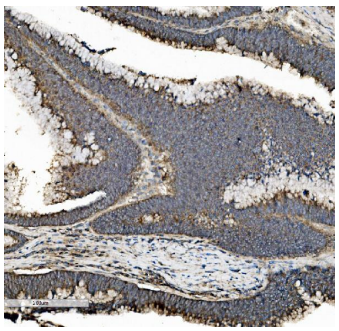

Figure 9. IHC analysis of Prohibitin/PHB using anti-Prohibitin/PHB antibody (A01178-1).
Prohibitin/PHB was detected in paraffin-embedded section of human rectal cancer tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2ug/ml rabbit anti-Prohibitin/PHB Antibody (A01178-1) 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 Strepavidin-Biotin-Complex (SABC) (Catalog # SA1022) with DAB as the chromogen.

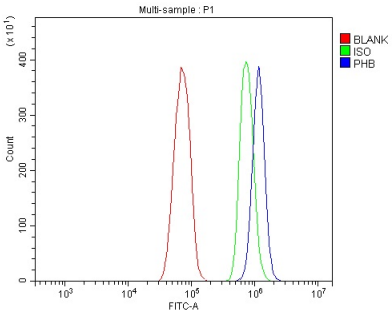

Figure 10. Flow Cytometry analysis of RH35 cells using anti-Prohibitin/PHB antibody (A01178-1).
Overlay histogram showing RH35 cells stained with A01178-1 (Blue line).The cells were blocked with 10% normal goat serum. And then incubated with rabbit anti-Prohibitin/PHB Antibody (A01178-1, 1ug/1x10^6 cells) for 30 min at 20°C. DyLight®488 conjugated goat anti-rabbit IgG (BA1127, 5-10ug/1x10^6 cells) was used as secondary antibody for 30 minutes at 20°C. Isotype control antibody (Green line) was rabbit IgG (1ug/1x10^6) used under the same conditions. Unlabelled sample (Red line) was also used as a control.

Figure 11. Flow Cytometry analysis of HL-60 cells using anti-Prohibitin/PHB antibody (A01178-1).
Overlay histogram showing HL-60 cells stained with A01178-1 (Blue line). The cells were blocked with 10%

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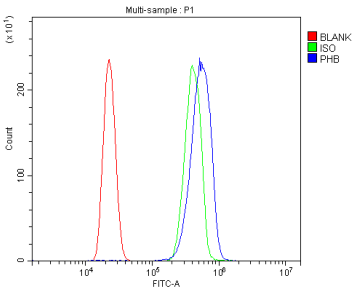

normal goat serum. And then incubated with rabbit anti-Prohibitin/PHB Antibody (A01178-1, 1 ug/1x10^6 cells) for 30 min at 20°C. DyLight®488 conjugated goat anti-rabbit IgG (BA1127, 5-10 ug/1x10^6 cells) was used as secondary antibody for 30 minutes at 20°C. Isotype control antibody (Green line) was rabbit IgG (1 ug/1x10^6) used under the same conditions. Unlabelled sample (Red line) was also used as a control.

Anti-Prohibitin/PHB Antibody