Rabbit Anti-Nicotinic Acetylcholine Receptor alpha 5/CHRNA5



(100 μ g/vial, Lyophilized)

Catalog Number: BPAB2672 FOR RESEARCH USE ONLY

Product Name	Anti-Nicotinic Acetylcholine Receptor alpha 5/CHRNA5 Antibody
Reactive Species	Human, Mouse, Rat
Description	Rabbit IgG polyclonal antibody for CHRNA5 detection. subunit alpha-5(CHRNA5) detection. Tested with WB, IHC-P, IHC-F, ICC, FCM in Human;Mouse;Rat.
Application	Flow Cytometry, IHC, ICC, WB
Clonality	Polyclonal
Formulation	Each vial contains 4mg Trehalose, 0.9mg NaCl, 0.2mg Na ₂ HPO ₄ , 0.05mg NaN ₃ .
Storage Instructions	At -20°C for one year. After reconstitution, at 4°C for one month. It can also be aliquotted and stored frozen at -20°C for a longer time. Avoid repeated freezing and thawing.
Host	Rabbit
Uniprot ID	P30532
Immunogen Cross Reactivity Form	A synthetic peptide corresponding to a sequence of human CHRNA5. No cross reactivity with other proteins. Lyophilized
Concentration	Western blot, 0.1-0.5µg/ml, Human, Rat
	Immunohistochemistry (Frozen Section), 0.5-1µg/ml, Human Immunocytochemistry, 0.5-1µg/ml, Human
	Flow Cytometry, 1-3µg/1x10 ⁶ cells, Human

1	2
130KD -	
100KD —	Figure 1. Western blot analysis of CHRNA5 using anti-CHRNA5 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 50ug of sample under reducing conditions. lane 1: rat skeletal muscle tissue lysates,
70KD -	
55KD	lane 2: HEPG2 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-CHRNA5 antigen affinity purified polyclonal antibody at 0.5 µg/mL
35KD —	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 1:10000 for 1.5 hour at RT. The signal is developed using an Enhanced Chemiluminescent detection (ECL) kit. A specific band was detected for
25KD —	CHRNA5 at approximately 53KD. The expected band size for CHRNA5 is at 53KD.
15KD -	



Figure 2. IHC analysis of CHRNA5 using anti-CHRNA5 antibody.

CHRNA5 was detected in paraffin-embedded section of <u>mouse intestine tissues</u>. 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 1µg/ml antibody 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) with DAB as the chromogen.



Figure 3. IHC analysis of CHRNA5 using anti-CHRNA5 antibody. CHRNA5 was detected in paraffin-embedded section of <u>mouse cardiac muscle tissues</u>. 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 1µg/ml rabbit anti-CHRNA5 Antibody 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-B iotin-Complex (SABC) with DAB as the chromogen.



Figure 4. IHC analysis of CHRNA5 using anti-CHRNA5 antibody. CHRNA5 was detected in paraffin-embedded section of <u>rat intestine tissues</u>. 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 1µg/ml antibody 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) with DAB as the chromogen.



Figure 5. IHC analysis of CHRNA5 using anti-CHRNA5 antibody. CHRNA5 was detected in paraffin-embedded section of <u>rat cardiac muscle tissues</u>. 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 1µg/ml antibody 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) with DAB as the chromogen.



Figure 6. IHC analysis of CHRNA5 using anti-CHRNA5 antibody. CHRNA5 was detected in paraffin-embedded section of <u>human prostatic cancer</u> <u>tissues</u>. 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 1µg/ml antibody 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) with DAB as the chromogen.



Figure 7. Flow Cytometry analysis of U251 cells using anti-CHRNA5 antibody. Overlay histogram showing U251 cells stained with the antibody (Blue line). The cells were blocked with 10% normal goat serum. And then incubated with rabbit anti-CHRNA5 antibody ($1\mu g/1x10$ cells) for 30 min at 20°C. DyLight®488 conjugated goat anti-rabbit IgG (5-10 $\mu g/1x10$ cells) was used as secondary antibody for 30 minutes at 20°C. Isotype control antibody (Green line) was rabbit IgG ($1\mu g/1x10$) used under the same conditions. Unlabelled sample (Red line) was also used as a control.

Figure 8. Flow Cytometry analysis of A549 cells using anti-CHRNA5 antibody. Overlay histogram showing A549 cells stained with the antibody (Blue line). The cells were blocked with 10% normal goat serum. And then incubated with rabbit anti CHRNA5 Antibody (1 μ g/1x10 cells) for 30 min at 20°C. DyLight®488 conjugated goat anti-rabbit IgG (5-10 μ g/1x10 cells) was used as secondary antibody for 30 minutes at 20°C. Isotype control antibody (Green line) was rabbit IgG (1 μ g/1x10) used under the same conditions. Unlabelled sample (Red line) was also used as a control.

Figure 9. Flow Cytometry analysis of MCF-7 cells using anti-CHRNA5 antibody. Overlay histogram showing MCF-7 cells stained with the antibody (Blue line). The cells were blocked with 10% normal goat serum. And then incubated with rabbit anti-CHRNA5 Antibody (1 μ g/1x10· cells) for 30 min at 20°C. DyLight®488 conjugated goat anti-rabbit IgG (5-10 μ g/1x10· cells) was used as secondary antibody for 30 minutes at 20°C. Isotype control antibody (Green line) was rabbit IgG (1 μ g/1x10·) used under the same conditions. Unlabelled sample (Red line) was also used as a control.