

# Rabbit Anti-Nicotinic Acetylcholine Receptor alpha 3/CHRNA3

(100 µg/vial, Lyophilized)

**Catalog Number: BPAB2670**

FOR RESEARCH USE ONLY

**Detroit R&D, Inc.**

<b>Product Name</b>	Anti-Nicotinic Acetylcholine Receptor alpha 3/CHRNA3 Antibody
<b>Reactive Species</b>	Human, Mouse, Rat
<b>Description</b>	Rabbit IgG polyclonal antibody for CHRNA3 detection. Tested with WB, FCM in Human;Mouse;Rat.
<b>Application</b>	Flow Cytometry, WB
<b>Clonality</b>	Polyclonal
<b>Formulation</b>	Each vial contains 4mg Trehalose, 0.9mg NaCl, 0.2mg Na <sub>2</sub> HPO <sub>4</sub> , 0.05mg NaN <sub>3</sub> .
<b>Storage Instructions</b>	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.
<b>Host</b>	Rabbit
<b>Uniprot ID</b>	P32297
<b>Immunogen</b>	A synthetic peptide corresponding to a sequence of human CHRNA3.
<b>Cross Reactivity</b>	No cross reactivity with other proteins.
<b>Form</b>	Lyophilized
<b>Concentration</b>	Western blot, 0.1-0.5µg/ml Flow Cytometry, 1-3µg/1x10 <sup>6</sup> cells, Human

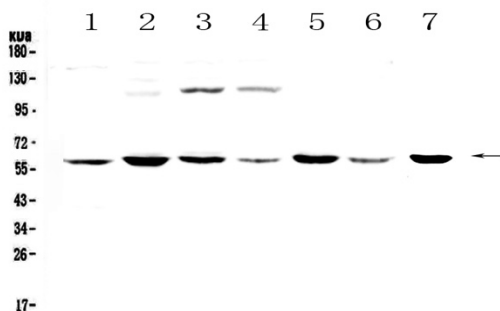


Figure 1. Western blot analysis of CHRNA3 using anti-CHRNA3 antibody. Electrophoresis was performed on a 5-20% SDS-PAGE gel at 70V (Stacking gel) / 90V (Resolving gel) for 2-3 hours. The sample well of each lane was loaded with 50ug of sample under reducing conditions.

Lane 1: human Hela whole cell lysates,  
Lane 2: human MDA-MB-453 whole cell lysates,  
Lane 3: human Jurkat whole cell lysates,  
Lane 4: human HepG2 whole cell lysates,  
Lane 5: human SK-OV-3 whole cell lysates,  
Lane 6: human PANC-1 whole cell lysates,  
Lane 7: mouse thymus tissue 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-CHRNA3 antigen affinity purified polyclonal antibody at 0.5 µg/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 1:10000 for 1.5 hour at RT. The signal is developed using an Enhanced Chemiluminescent detection (ECL) kit. A specific band was detected for CHRNA3 at approximately 60KD. The expected band size for CHRNA3 is at 57KD.

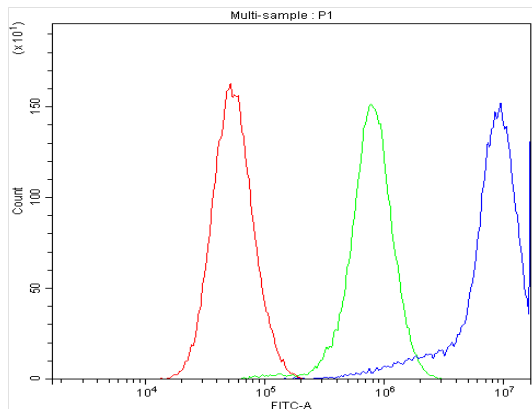


Figure 2. Flow Cytometry analysis of U251 cells using anti-CHRNA3 antibody. Overlay histogram showing U251 cells stained with the antibody (Blue line). The cells were blocked with 10% normal goat serum. And then incubated with rabbit anti-CHRNA3 Antibody (1,1µg/1x10<sup>6</sup> cells) for 30 min at 20°C. DyLight®488 conjugated goat anti-rabbit IgG (5-10µg/1x10<sup>6</sup> cells) was used as secondary antibody for 30 minutes at 20°C. Isotype control antibody (Green line) was rabbit IgG (1µg/1x10<sup>6</sup>) used under the same conditions. Unlabelled sample (Red line) was also used as a control.

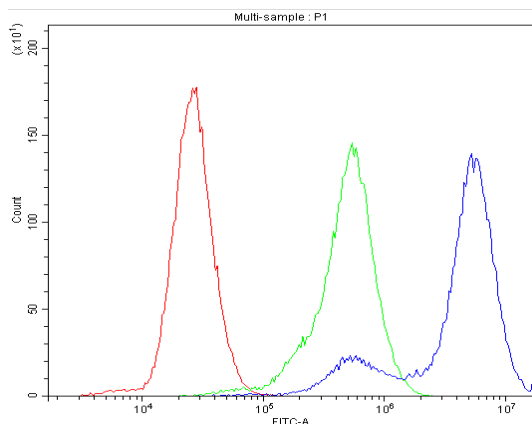


Figure 3. Flow Cytometry analysis of U-87 cells using anti-CHRNA3 antibody. Overlay histogram showing U-87 cells stained with the antibody (Blue line). The cells were blocked with 10% normal goat serum. And then incubated with rabbit anti-CHRNA3 Antibody (1,1µg/1x10<sup>6</sup> cells) for 30 min at 20°C. DyLight®488 conjugated goat anti-rabbit IgG (5-10µg/1x10<sup>6</sup> cells) was used as secondary antibody for 30 minutes at 20°C. Isotype control antibody (Green line) was rabbit IgG (1µg/1x10<sup>6</sup>) used under the same conditions. Unlabelled sample (Red line) was also used as a control.