

## Basic Information

<b>Product Name</b>	Anti-Beclin 1/BECN1 Antibody	
<b>Gene Name</b>	BECN1	
<b>Source</b>	Rabbit	
<b>Isotype</b>	IgG	
<b>Species Reactivity</b>	human, mouse, rat	
<b>Tested Application</b>	WB, IHC, ICC/IF, FCM	
<b>Contents</b>	500 ug/ml antibody with PBS , 0.02% NaN3 , 1 mg BSA and 50% glycerol.	
<b>Immunogen</b>	E.coli-derived human Beclin 1 recombinant protein (Position: M1-S354). Human Beclin 1 shares 97% amino acid (aa) sequence identity with both mouse and rat Beclin 1.	
<b>concentration</b>	500 ug/ml	
<b>Purification</b>	Immunogen affinity purified.	
<b>Observed MW</b>	52-60KD	
<b>Dilution Ratios</b>	Western blot(WB): 1:500-2000 Immunohistochemistry in paraffin section (IHC): 1:50-400 Immunocytochemistry/Immunofluorescence(ICC/IF): 1:50-400 Flow cytometry (FCM): 1-3 $\mu$ g/1x10 <sup>6</sup> cells (Boiling the paraffin sections in 10mM citrate buffer,pH6.0,or PH8.0 EDTA repair liquid for 20 mins is required for the staining of formalin/paraffin sections.) Optimal working dilutions must be determined by end user.	

## Storage

12 months from date of receipt, -20°C as supplied. 6 months 2 to 8°C after reconstitution. Avoid repeated freezing and thawing.

## Background Information

Beclin-1, also known as also known as ATG6 or VPS30 is a protein that in humans is encoded by the BECN1 gene. Beclin-1 and its binding partner class III phosphoinositide 3-kinase (PI3K), also named Vps34, are required for the initiation of the formation of the autophagosome in autophagy. This gene participates in the regulation of autophagy and has an important role in development, tumorigenesis, and neurodegeneration. Schizophrenia is associated with low levels of Beclin-1 in the hippocampus of the affected which causes diminished autophagy which in turn results in increased neuronal cell death. It has been found that beclin-1 can promote autophagy in autophagy-defective yeast with a targeted disruption of *apg6/vps30*, and in human MCF7 breast carcinoma cells.

## Reference

Anti-Beclin 1/BECN1 Antibody被引用在1文献中。

## Selected Validation Data

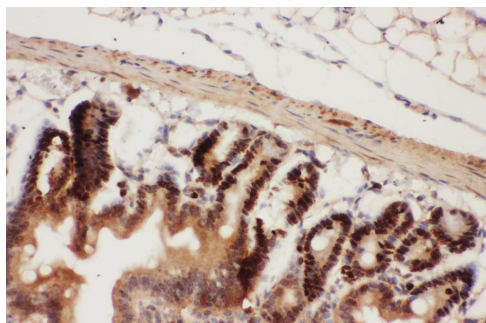


Figure 1. IHC analysis of Beclin-1 using anti-Beclin-1 antibody (PB9076). Beclin-1 was detected in paraffin-embedded section of Mouse Intestine Tissue. Heat mediated antigen retrieval was performed in citrate buffer (pH6, epitope retrieval solution) for 20 mins. The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1 $\mu$ g/ml rabbit anti-Beclin-1 Antibody (PB9076) 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.

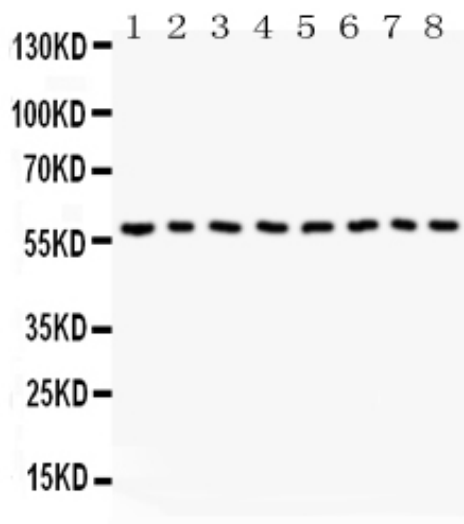


Figure 5. Western blot analysis of Beclin-1 using anti-Beclin-1 antibody (PB9076). Electrophoresis was performed on a 5-20% SDS-PAGE gel at 70V (Stacking gel) / 90V (Resolving gel) for 2-3 hours. The sample well of each lane was loaded with 50ug of sample under reducing conditions. Lane 1: Colo320 Whole Cell Lysate Lane 2: HepG2 Whole Cell Lysate Lane 3: PANC Whole Cell Lysate Lane 4: A431 Whole Cell Lysate Lane 5: Smmc Whole Cell Lysate Lane 6: Jurkat Whole Cell Lysate Lane 7: SW620 Whole Cell Lysate Lane 8: U87 Whole Cell Lysate 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-Beclin-1 antigen affinity purified polyclonal antibody (Catalog # PB9076) at 0.5  $\mu$ 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-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 (Catalog # EK1002) with Tanon 5200 system. A specific band was detected for Beclin-1 at approximately 60KD. The expected band size for Beclin-1 is at 52KD.

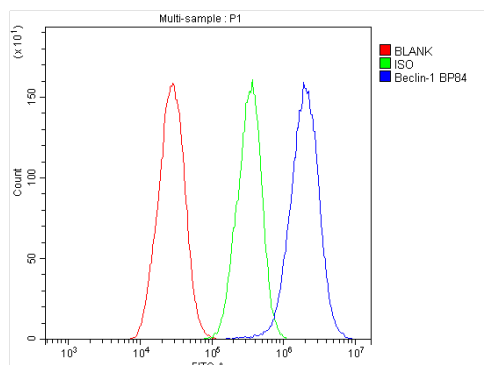


Figure 6. Flow Cytometry analysis of HepG2 cells using anti-Beclin-1 antibody (PB9076).

Overlay histogram showing HepG2 cells stained with PB9076 (Blue line). The cells were blocked with 10% normal goat serum. And then incubated with rabbit anti- Beclin-1 Antibody (PB9076,  $1\mu\text{g}/1 \times 10^6$  cells) for 30 min at  $20^\circ\text{C}$ . DyLight488 conjugated goat anti-rabbit IgG (BA1127,  $5\text{-}10\mu\text{g}/1 \times 10^6$  cells) was used as secondary antibody for 30 minutes at  $20^\circ\text{C}$ . Isotype control antibody (Green line) was rabbit IgG ( $1\mu\text{g}/1 \times 10^6$ ) used under the same conditions. Unlabelled sample (Red line) was also used as a control.

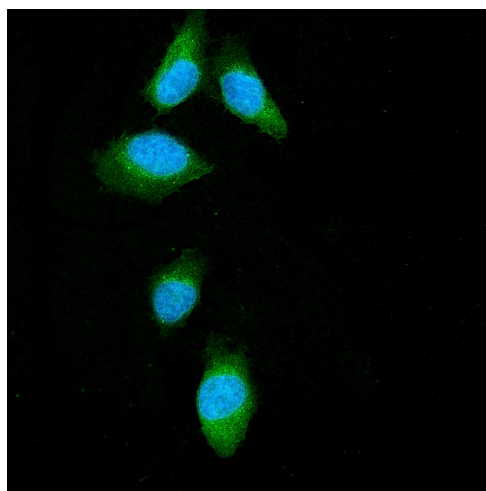


Figure 7. ICC analysis using anti- Beclin-1 antibody (PB9076) was detected in immersion fixed U2OS cell line . Cells were stained using the Dylight488-conjugated Anti-rabbit IgG Secondary Antibody (green)(Catalog#BA1127) and counterstained with DAPI (blue).