

# Anti-CD27 Antibody [PSH08-90] - BSA and Azide free

## HA751255



<b>Product Type:</b>	Recombinant Rabbit monoclonal IgG, primary antibodies
<b>Species reactivity:</b>	Human
<b>Applications:</b>	WB, IF-Cell, IHC-P, FC, IP
<b>Molecular Wt:</b>	Predicted band size: 29 kDa
<b>Clone number:</b>	PSH08-90

**Description:** The tumor necrosis factor (TNF) receptor family is composed of several type I integral membrane glycoproteins that exhibit homology in their cysteine-rich extracellular domains. Members of this family include FAS, OX40, CD27 and CD30. Ligands for these receptors are often type II transmembrane glycoproteins, as is the case for CD27 and CD30. CD27 is a homodimeric lymphocyte-specific surface antigen present on T and B lymphocytes. Activation of the CD3 complex via the T cell receptor for antigen leads to an increase in CD27 expression. Together, CD27 and its ligand, CD27L, generate co-stimulatory signals required for complete T cell activation. CD30 is a surface marker for neoplastic cells of the Hodgkin's lymphoma and related hematologic malignancies. CD30L has been shown to enhance the proliferation of the Hodgkin's cell line HDLM-2, but exerts antiproliferative effects on large cell anaplastic lymphoma cell lines.

**Immunogen:** Recombinant protein within human CD27 aa 1-191.

**Positive control:** Raji cell lysate, Ramos cell lysate, Daudi cell lysate, Raji, human tonsil tissue, Human peripheral blood lymphocytes.

**Subcellular location:** Cell membrane.

**Database links:** SwissProt: P26842 Human

**Recommended Dilutions:**

<b>WB</b>	1:2,000
<b>IF-Cell</b>	1:100
<b>IHC-P</b>	1:200
<b>FC</b>	1:1,000
<b>IP</b>	1-2µg/sample

**Storage Buffer:** 1\*PBS (pH7.4).

**Storage Instruction:** Store at +4°C after thawing. Aliquot store at -20°C. Avoid repeated freeze / thaw cycles.

**Purity:** Protein A affinity purified.

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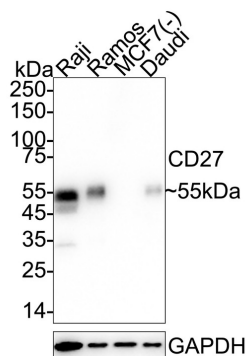
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## Images



**Fig1:** Western blot analysis of CD27 on different lysates with Rabbit anti-CD27 antibody (HA751255) at 1/2,000 dilution.

Lane 1: Raji cell lysate  
 Lane 2: Ramos cell lysate  
 Lane 3: MCF7 cell lysate (negative)  
 Lane 4: Daudi cell lysate

Lysates/proteins at 20 µg/Lane.

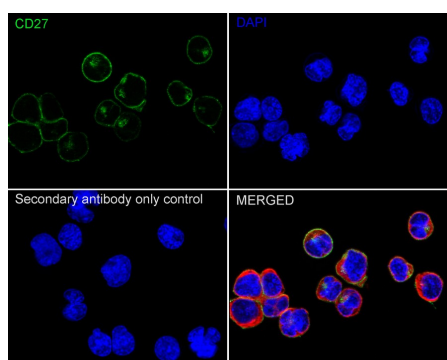
Predicted band size: 29 kDa  
 Observed band size: 55 kDa

Exposure time: 30 seconds; ECL: K1801;

4-20% SDS-PAGE gel.

Proteins were transferred to a PVDF membrane and blocked with 5% NFDM/TBST for 1 hour at room temperature. The primary antibody (HA751255) at 1/2,000 dilution was used in 5% NFDM/TBST at 4°C overnight. Goat Anti-Rabbit IgG - HRP Secondary Antibody (HA1001) at 1/50,000 dilution was used for 1 hour at room temperature.

**Fig2:** Immunocytochemistry analysis of Raji cells labeling CD27 with Rabbit anti-CD27 antibody (HA751255) at 1/100 dilution.



Cells were fixed in 4% paraformaldehyde for 15 minutes at room temperature, permeabilized with 0.1% Triton X-100 in PBS for 15 minutes at room temperature, then blocked with 1% BSA in 10% negative goat serum for 1 hour at room temperature. Cells were then incubated with Rabbit anti-CD27 antibody (HA751255) at 1/100 dilution in 1% BSA in PBST overnight at 4 °C. Goat Anti-Rabbit IgG H&L (iFluor™ 488, HA1121) was used as the secondary antibody at 1/1,000 dilution. PBS instead of the primary antibody was used as the secondary antibody only control. Nuclear DNA was labelled in blue with DAPI.

Beta tubulin (HA601187, red) was stained at 1/100 dilution overnight at +4°C. Goat Anti-Mouse IgG H&L (iFluor™ 594, HA1126) was used as the secondary antibody at 1/1,000 dilution.

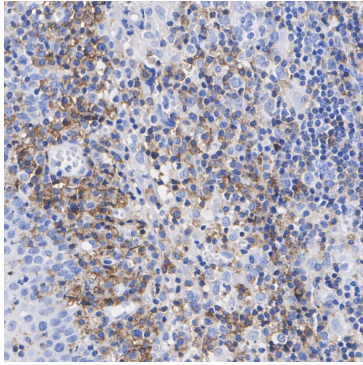
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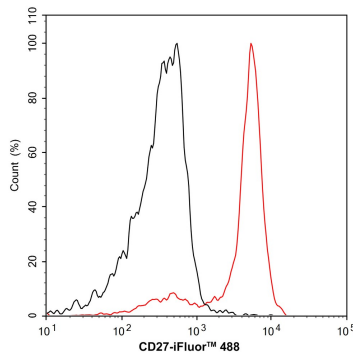
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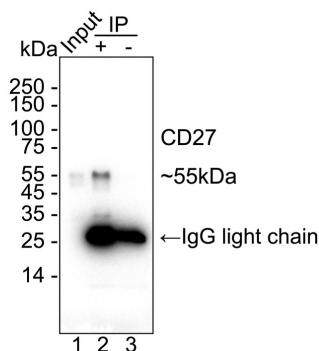
**Fig3:** Immunohistochemical analysis of paraffin-embedded human tonsil tissue with Rabbit anti-CD27 antibody (HA751255) at 1/200 dilution.

The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 20 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH<sub>2</sub>O and PBS, and then probed with the primary antibody (HA751255) at 1/200 dilution for 1 hour at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.



**Fig4:** Flow cytometric analysis of human peripheral blood lymphocytes labeling CD27.

Cells were washed twice with cold PBS and resuspend. Then stained with the primary antibody (HA751255, 1 $\mu$ g/mL) (red) compared with Rabbit IgG Isotype Control (black). After incubation of the primary antibody at +4 $^{\circ}$ C for an hour, the cells were stained with a iFluor™ 488 conjugate-Goat anti-Rabbit IgG Secondary antibody (HA1121) at 1/1,000 dilution for 30 minutes at +4 $^{\circ}$ C.



**Fig5:** CD27 was immunoprecipitated from 0.2 mg Raji cell lysate with HA751255 at 2  $\mu$ g/10  $\mu$ l beads. Western blot was performed from the immunoprecipitate using HA751255 at 1/1,000 dilution. Mouse Anti-Rabbit IgG kappa light chain secondary antibody (M1208-2) at 1/5,000 dilution was used for 1 hour at room temperature.

Lane 1: Raji cell lysate (input)  
Lane 2: HA751255 IP in Raji cell lysate  
Lane 3: Rabbit IgG instead of HA751255 in Raji cell lysate

Blocking/Dilution buffer: 5% NFDM/TBST  
Exposure time: 3 minutes; ECL: K1801

**Note:** All products are "FOR RESEARCH USE ONLY AND ARE NOT INTENDED FOR DIAGNOSTIC OR THERAPEUTIC USE".

## Background References

1. Grimsholm O. CD27 on human memory B cells-more than just a surface marker. Clin Exp Immunol. 2023 Jul
2. Starzer AM et al. New emerging targets in cancer immunotherapy: CD27 (TNFRSF7). ESMO Open. 2020 Mar

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