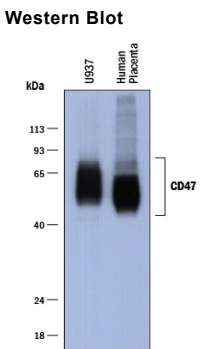
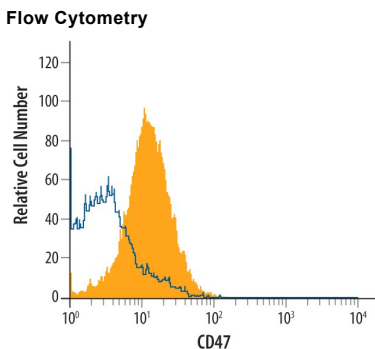
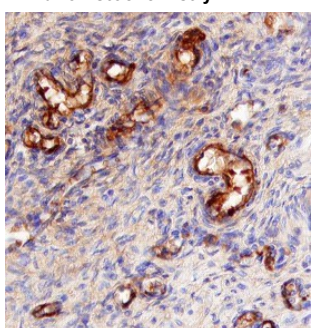
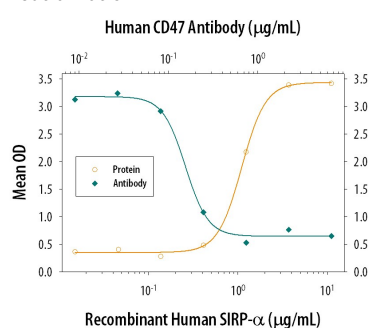


DESCRIPTION	
<b>Species Reactivity</b>	Human
<b>Specificity</b>	Detects human CD47 in direct ELISAs and Western blots. In direct ELISAs and Western blots, less than 5% cross-reactivity with recombinant mouse CD47 is observed.
<b>Source</b>	Polyclonal Sheep IgG
<b>Purification</b>	Antigen Affinity-purified
<b>Immunogen</b>	Mouse myeloma cell line NS0-derived recombinant human CD47 Gln19-Pro139 Accession # Q08722
<b>Endotoxin Level</b>	<0.10 EU per 1 µg of the antibody by the LAL method.
<b>Formulation</b>	Lyophilized from a 0.2 µm filtered solution in PBS with Trehalose. See Certificate of Analysis for details. *Small pack size (-SP) is supplied as a 0.2 µm filtered solution in PBS.

APPLICATIONS	
<b>Please Note:</b> Optimal dilutions should be determined by each laboratory for each application. <i>General Protocols</i> are available in the <i>Technical Information</i> section on our website.	
	<b>Recommended Concentration      Sample</b>
<b>Western Blot</b>	1 µg/mL      See Below
<b>Flow Cytometry</b>	2.5 µg/10 <sup>6</sup> cells      See Below
<b>Immunohistochemistry</b>	5-15 µg/mL      See Below
<b>CytoF-ready</b>	Ready to be labeled using established conjugation methods. No BSA or other carrier proteins that could interfere with conjugation.
<b>Neutralization</b>	Measured by its ability to neutralize SIRPα/CD172a-mediated adhesion of human erythrocytes. The adhesion of human erythrocytes to immobilized Recombinant Human SIRPα/CD172a/Fc Chimera (2 µg/mL, 100 µL/well) was maximally inhibited (70-100%) by 2 µg/mL of the antibody.

DATA	
<p><b>Western Blot</b></p>  <p><b>Detection of Human CD47 by Western Blot.</b> Western blot shows lysates of U937 human histiocytic lymphoma cell line and human placenta tissue, not heated to minimize aggregation. PVDF membrane was probed with 1 µg/mL of Sheep Anti-Human CD47 Antigen Affinity-purified Polyclonal Antibody (Catalog # AF4670) followed by HRP-conjugated Anti-Sheep IgG Secondary Antibody (Catalog # HAF016). Specific bands were detected for CD47 at approximately 45-70 kDa (as indicated). This experiment was conducted under reducing conditions and using Immunoblot Buffer Group 1.</p>	<p><b>Flow Cytometry</b></p>  <p><b>Detection of CD47 in Human Lymphocytes by Flow Cytometry.</b> Human whole blood lymphocytes were stained with Sheep Anti-Human CD47 Antigen Affinity-purified Polyclonal Antibody (Catalog # AF4670, filled histogram) or control antibody (Catalog # 5-001-A, open histogram), followed by Northern-Lights™ 557-conjugated Anti-Sheep IgG Secondary Antibody (Catalog # NL010).</p>
<p><b>Immunohistochemistry</b></p>  <p><b>CD47 in Human Placenta.</b> CD47 was detected in immersion fixed paraffin-embedded sections of human placenta using 5 µg/mL Sheep Anti-Human CD47 Antigen Affinity-purified Polyclonal Antibody (Catalog # AF4670) overnight at 4 °C. Tissue was stained with the Anti-Sheep HRP-DAB Cell &amp; Tissue Staining Kit (brown; Catalog # CTS019) and counterstained with hematoxylin (blue). View our protocol for Chromogenic IHC Staining of Paraffin-embedded Tissue Sections.</p>	<p><b>Neutralization</b></p>  <p><b>Cell Adhesion Mediated by SIRPα/CD172a and Neutralization by Human CD47 Antibody.</b> Recombinant Human SIRPα/CD172a (Catalog # 4546-SA), immobilized onto a microplate, supports the adhesion of the human erythrocytes in a dose-dependent manner (orange line). Adhesion elicited by Recombinant Human SIRP-α (2 µg/mL) is neutralized (green line) by increasing concentrations of Sheep Anti-Human CD47 Polyclonal Antibody (Catalog # AF4670). The adhesion was maximally inhibited (70-100%) by 2 µg/mL of the antibody.</p>

#### PREPARATION AND STORAGE

<b>Reconstitution</b>	Reconstitute at 0.2 mg/mL in sterile PBS.
<b>Shipping</b>	The product is shipped at ambient temperature. Upon receipt, store it immediately at the temperature recommended below. *Small pack size (-SP) is shipped with polar packs. Upon receipt, store it immediately at -20 to -70 °C
<b>Stability &amp; Storage</b>	<b>Use a manual defrost freezer and avoid repeated freeze-thaw cycles.</b> <ul style="list-style-type: none"><li>● 12 months from date of receipt, -20 to -70 °C as supplied.</li><li>● 1 month, 2 to 8 °C under sterile conditions after reconstitution.</li><li>● 6 months, -20 to -70 °C under sterile conditions after reconstitution.</li></ul>

#### BACKGROUND

CD47 (also integrin-associated protein/IAP and OA3) is a variably glycosylated, 40-60 kDa atypical member of the Ig-Superfamily. It is expressed on almost all cell types, including erythrocytes. CD47 binds to TSP-1 and SIRP $\alpha$ , and forms a membrane complex with CD36 and  $\alpha_v\beta_3$ . Mature human CD47 is a 305 amino acid (aa), five-transmembrane glycoprotein. It contains a 123 aa extracellular region (aa 19-141) that is characterized by the presence of a V-type Ig-like domain (aa 19-127), and a 34 aa C-terminal cytoplasmic tail that interacts with Gic subunits. Three splice variants occur over aa 293-323. Over aa 19-139, human CD47 shares 61%, 71% and 66% aa identity with mouse, porcine and canine CD47, respectively.