### DESCRIPTION

**Species Reactivity**  
Human

**Specificity**  
Selected for its ability to block the adhesion of HSB-2 cells to immobilized recombinant human (rh) ICAM-1. In direct ELISAs, approximately 5% cross-reactivity with rhICAM-3 is observed and less than 2% cross-reactivity with rhICAM-2, rhICAM-4, rhICAM-5, recombinant rat ICAM-1, recombinant mouse ICAM-1, and rhVCAM-1 is observed.

**Source**  
Polyclonal Sheep IgG

**Purification**  
Antigen Affinity-purified

**Immunogen**  
Chinese hamster ovary cell line CHO-derived recombinant human ICAM-1 Extracellular domain

**Endotoxin Level**  
<0.20 EU per 1 μg of the antibody by the LAL method.

**Formulation**  
Lyophilized from a 0.2 μm filtered solution in PBS with Trehalose. See Certificate of Analysis for details.

*Small pack size (-SP) is supplied either lyophilized or as a 0.2 μm filtered solution in PBS.

### APPLICATIONS

**Please Note:** Optimal dilutions should be determined by each laboratory for each application. General Protocols are available in the Technical Information section on our website.

<table>
<thead>
<tr>
<th>Application</th>
<th>Recommended Concentration</th>
<th>Sample</th>
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</thead>
<tbody>
<tr>
<td>Western Blot</td>
<td>0.2 μg/mL</td>
<td>See Below</td>
</tr>
<tr>
<td>Immunocytochemistry</td>
<td>1-15 μg/mL</td>
<td>See Below</td>
</tr>
<tr>
<td>Adhesion Blockade</td>
<td></td>
<td>The adhesion of HSB2 human peripheral blood acute lymphoblastic leukemia cells (5 x 10^4 cells/well) to immobilized recombinant human ICAM-1/Fc Chimera (12.5 μg/mL, 100 μL/well) was maximally inhibited (80-100%) by 25 μg/mL of the antibody.</td>
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</tbody>
</table>

### DATA

#### Western Blot

Detection of Human ICAM-1/CD54 by Western Blot.

Western blot shows lysates of HUVEC human umbilical vein endothelial cells and Ramos human Burkitt’s lymphoma cell line untreated (-) or treated (+) with 10 ng/mL Recombinant Human TNF-α (Catalog # 210-TA) for 24 hours. PVDF membrane was probed with 0.2 μg/mL of Sheep Anti-Human ICAM-1/CD54 Antigen Affinity-purified Polyclonal Antibody (Catalog # AF720) followed by HRP-conjugated Anti-Sheep IgG Secondary Antibody (Catalog # HAF016). A specific band was detected for ICAM-1/CD54 at approximately 90 kDa (as indicated). This experiment was conducted under reducing conditions and using Immunoblot Buffer Group 1.

#### Immunocytochemistry

ICAM-1/CD54 in A431 Human Cell Line. ICAM-1/CD54 was detected in immersion fixed A431 human epithelial carcinoma cell line (left panel; positive stain) and A549 human lung carcinoma cell line (right panel; negative stain) using Sheep Anti-Human ICAM-1/CD54 Antigen Affinity-purified Polyclonal Antibody (Catalog # AF720) at 1.7 μg/mL for 3 hours at room temperature. Cells were stained using the NorthernLights™ 557-conjugated Anti-Sheep IgG Secondary Antibody (red; Catalog # NL010) and counterstained with DAPI (blue). Specific staining was localized to plasma membrane. View our protocol for Fluorescent ICC Staining of Cells on Coverslips.

### PREPARATION AND STORAGE

**Reconstitution**  
Reconstitute at 0.2 mg/mL in sterile PBS.

**Shipping**  
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.

**Stability & Storage**  
Use a manual defrost freezer and avoid repeated freeze-thaw cycles.

- 12 months from date of receipt, -20 to -70 °C as supplied.
- 1 month, 2 to 8 °C under sterile conditions after reconstitution.
- 6 months, -20 to -70 °C under sterile conditions after reconstitution.

### BACKGROUND

Intercellular Adhesion Molecule-1 (ICAM-1) binds the leukocyte integrins LFA-1 and Mac-1. ICAM-1 expression is weak on leukocytes, epithelial and resting endothelial cells, as well as some other cell types, but expression can be boosted by IFN-γ, TNF-α, IL-1β and LPS. Soluble ICAM-1 is found in a biologically active form in serum, probably as a result of proteolytic cleavage from the cell surface, and is elevated in patients with various inflammatory syndromes such as septic shock, LAD, cancer and transplantation.