

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.

	Recommended Concentration	Sample
Western Blot	0.2 µg/mL	See Below
Immunocytochemistry	1-15 µg/mL	See Below
Adhesion Blockade	The adhesion of HSB2 human peripheral blood acute lymphoblastic leukemia cells (5×10^4 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.	

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. <ul style="list-style-type: none"> • 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 stimulated 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.