

## Product Information

### MONOCLONAL ANTI-HUMAN CD54 (ICAM-1), Clone 8.4A6

Purified Mouse Immunoglobulin

Product Number **C 7219**

#### Product Description

Monoclonal Anti-Human CD54 (ICAM-1) (mouse IgG1 isotype) is derived from the 8.4A6 hybridoma produced by the fusion of mouse myeloma cells and splenocytes from BALB/c mice immunized with a TNF- $\alpha$  activated human endothelial cells.<sup>1</sup> The isotype is determined using Sigma ImmunoType Kit (Product Code ISO-1) and by a double diffusion immunoassay using Mouse Monoclonal Antibody Isotyping Reagents (Product Code ISO-2). The antibody is purified from ascites fluid using protein A.

Monoclonal Anti-Human CD54 (ICAM-1) reacts specifically with CD54 (ICAM-1) expressed on the surface of activated endothelial cells, lymphocytes, monocytes and granulocytes. The epitope recognized by the antibody is localized in domain D2 of the CD54 (ICAM-1) molecule.<sup>2</sup> The product may be used in flow cytometry, ELISA,<sup>3</sup> and immunohistochemistry (frozen tissues). It inhibits leukocyte adhesion to activated endothelial cells,<sup>2</sup> but not adhesion of *Plasmodium falciparum*-infected erythrocytes.<sup>3</sup> The antibody stimulates homotypic aggregation of SKW3 cells.<sup>2</sup> It is also useful for studying soluble CD54 (ICAM-1) using capture ELISA.<sup>4</sup>

Human CD54, also known as intercellular adhesion molecule-1 (ICAM-1), is a 85-110 kDa single-chain type 1 integral membrane glycoprotein with an extracellular domain of five immunoglobulin superfamily repeats, a transmembrane region and a cytoplasmic domain. It shares considerable amino acid sequence homology with CD50 (ICAM-3) and with CD102 (ICAM-2).<sup>5,6</sup> CD54 (ICAM-1) is expressed by activated endothelial cells. It is detected on cells of many other lineages (e.g. epithelial cells, fibroblasts, chondrocytes, B lymphocytes, T lymphocytes (low), monocytes, macrophages, dendritic cells and neutrophils, with lower levels that increase in inflammation.

Also, CD54 (ICAM-1) is detected in some carcinoma and melanoma cells.<sup>2</sup> Soluble CD54 (ICAM-1) is detectable in the plasma and is elevated in patients with various inflammatory syndromes.<sup>4</sup> CD54 (ICAM-1) mediates myeloid cells adhesion to activated vascular endothelium at the vessel wall and to other leucocytes.<sup>1,2</sup> CD54 (ICAM-1) mediates T cell interactions with Antigen Presenting Cells or target cells and other T-T or T-B cell interactions. CD54 (ICAM-1) is the ligand for the integrin LFA-1 (CD11a/CD18).<sup>5</sup> In addition, CD54 has binding sites for Rhinovirus,<sup>6</sup> Mac-1(CD11b/CD18)<sup>7</sup> and *Plasmodium falciparum*-infected erythrocytes.<sup>3</sup> It seems to serve as an adhesion receptor for *Plasmodium falciparum*, thereby enabling binding of infected red blood cells to the vascular endothelia in selected organs. This causes the typical sequestration of severe malaria. *In vitro* upregulation of CD54

(ICAM-1) expression of cultured endothelial cells occurs upon activation by various inflammatory mediators, such as IL-1 $\beta$ , TNF- $\alpha$  or IFN- $\gamma$ . It is induced by phorbol esters, retinoic acid and lipopolysaccharide. Peak levels of CD54 (ICAM-1) usually occur within 10-24 hours.<sup>1</sup> The 8.4A6 producing hybridoma was developed by D.O. Haskard and coworkers<sup>1</sup> at the Rheumatology Unit, the Division of Medicine, Guy's Hospital, London.

#### Reagents

The product is supplied in 0.01 M phosphate buffered saline, pH 7.4, containing 1% BSA and 15 mM sodium azide as a preservative.

### Precautions and Disclaimer

Due to the sodium azide content a material safety sheet (MSDS) for this product has been sent to the attention of the safety officer of your institution. Consult the MSDS for information regarding hazardous and safe handling practices.

### Storage/Stability

Store at 2-8 °C. If slight turbidity occurs upon prolonged storage, clarify the solution by centrifugation before use.

### Product Profile

When assayed by flow cytometric analysis (FACScan) using 5µl of the antibody to stain  $2 \times 10^5$  cells (rhTNF- $\alpha$  stimulated cultured human umbilical vein endothelial cells, HUVEC), a fluorescence intensity is observed similar to that obtained with saturating monoclonal antibody levels. The percent population positive is also at the maximum percentage positive using saturating monoclonal antibody levels.

A minimum working dilution of 1:10 is determined by indirect immunofluorescent staining of acetone-fixed frozen sections of human tonsil.

### References

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5. Springer, T.A., et al., Nature **346**, 425 (1990).
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7. Diamond, M.S., et al., Cell **65**, 961 (1991).

Pcs/lpg/kaa 02/02

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