to LFA-1, *P. falciparum*-infected erythrocytes, and rhinoviruses.

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AS4.3 CD102 (ICAM-2) cluster report

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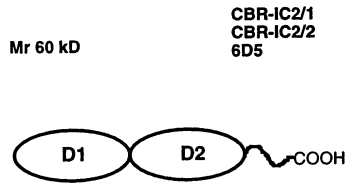
CD102 (ICAM-2)



The existence of a second ligand for LFA-1 was initially postulated based on the observation that some cell–cell interactions were blocked by monoclonal antibodies (mAb) to LFA-1, but not by mAb to ICAM-1. This activity was constitutively expressed on endothelium [1]. A cDNA for ICAM-2 was isolated from an endothelial library by screening for the ability of transfected COS cells to bind to LFA-1 [2]. deFougerolles *et al.* [3] obtained mAb to ICAM-2 expressed in COS-cell transfectants and Nortamo *et al.* [4] obtained mAb to ICAM-2 expressed in *Escherichia coli*. These mAb, submitted to the Fifth Workshop as S085 (CBR-IC2/1), S086 (CBR-IC2/2), and S099 (6D5), allowed ICAM-2 to be clustered as CD102. No ICAM-2 mAb were identified in the Endothelial Section [Klickstein *et al.*, E6.29] or in the Blind Panel [Shaw *et al.*, BP1.3].

Cellular expression

Immunohistochemical and flow cytometric studies [Autschbach *et al.*, AS2.5; Koretz *et al.*, AS4.16; Krajewski *et al.*, AS10.10; unpublished Workshop studies by Athanasou, Bene, Cerf-Bensussan, Malizia, Patarroyo, Soligo, and Timens] found high levels of ICAM-2 on all endothelium and lower levels on a subset of haemopoietic cells, as previously published [3]. Interestingly, ICAM-2 is expressed on platelets as the sole ICAM, and can function as an LFA-1 ligand on these cells [5]. ICAM-2 was also found on thymic stromal cells [Friedrich *et al.*, AS6.15], but in general was not present on non-haemopoietic cells other than endothelium. Unlike ICAM-1, ICAM-2 is constitutively expressed and not responsive to lipopolysaccharide (LPS) or cytokines.

Immunochemistry

Immunoprecipitation of ^{125}I-labelled human umbilical cord vein endothelial cells (HUVEC) revealed a broad 60-kDa band in sodium dodecyl sulfate-polyacrylamide gel electrophoresis (SDS-PAGE) with or without reduction. Treatment with N-glycanase decreased the apparent M_r of ICAM-2 to 31 kDa [3]. No other posttranslational modifications have been described.

Molecular cloning

The ICAM-2 cDNA reveals a type 1 integral membrane glycoprotein with two immunoglobulin superfamily (IgSF) repeats, a 26-amino-acid transmembrane region, and a 26-residue cytoplasmic domain (see introductory diagram) [2]. The IgSF repeats are most similar to those of CD50 and CD54, with 34 and 37 per cent amino acid identity in the first two IgSF domains, respectively [2].

Transfectant and epitope analysis

The specificity of the ICAM-2 mAb was confirmed on recombinant protein by at least two of the laboratories [Klickstein *et al.*, AS4, Table 1].

Function

LFA-1 (CD11a/CD18), is the only known counter-receptor for ICAM-2. S086 (CBR/IC2/2) and S099 (6D5) were found to efficiently block the LFA-1/ICAM -2 interaction, whereas S085 (CBR-IC2/1) did not.

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AS4.4 Interaction of the ICAM molecules with β_2 integrins on T cells and neutrophils

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The intercellular adhesion molecules, ICAM-1, -2, and -3, are the ligands for the leucocyte-restricted integrin LFA-1 and have important roles in cell–cell adhesion in the immune system. They belong to the immunoglobulin superfamily: ICAM-2 has two extracellular Ig-like domains; ICAM-1 and ICAM-3 both have five such domains. A combination of domain deletion constructs, chimeric proteins, and homologue scanning mutagenesis has been used in previous studies to locate the antibody epitopes and binding site for LFA-1 on ICAM-1 [1,2]. The results have been integrated into a model of the two N-terminal domains of ICAM-1 and suggest that E34 on the C strand and Q73 on the F/G loop of domain 1 are critical for binding to LFA-1. The role of domain 2 is not clear, but it appears to be important for the conformational integrity of domain 1 and may also participate in binding of LFA-1 in that two LFA-1 blocking monoclonal antibodies (mAb) have been mapped to this domain. In other studies peptides from domains 2, 4, and 5 interfere with cell aggregation, which leaves open the possibility that, although domains 1 and 2 have been shown to be sufficient for ligation to LFA-1, other domains may have a role in this ICAM-1 function [3,4]. Domain 3 has been shown to contain the binding site for another leucocyte integrin, Mac-1 [5].

To determine which ICAMs are recognized by the CAM panel (Subpanel 4), 35 (S083 was not received) of the 36 mAb were tested for reactivity with ICAM-1 Fc (5 domain), ICAM-2 Fc (2 domain), and ICAM-3 Fc