AS5.1 CD11a cluster report

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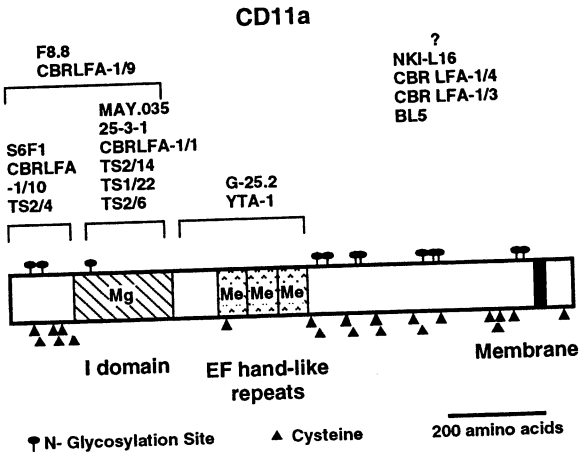

The CD11a cluster was designated during the First Workshop and distinguished from CD18 in the Third Workshop. CD11a complexes with the β_2 subunit of the integrin family, CD18, to form the cell surface heterodimer, LFA-1 [1]. It is a member of the leucocyte family of integrins that also includes Mac-1 (CD11b/CD18) and p150,95 (CD11c/CD18) [1]. LFA-1 binds to three cell surface proteins, ICAM-1 (CD54), ICAM-2 (CD102), and ICAM-3 (CD50), which are all members of the Ig supergene family [2–4]. Expression of CD11a on the cell surface requires the presence of an intact β_2 subunit [5]. Patients with leucocyte adhesion deficiency, who lack a functional CD18 molecule, do not express LFA-1 on their cell surface and have an abnormality in the function of their leucocytes [5,6]. The monoclonal antibodies (mAb) that clustered within CD11a include S125 (F8.8), S140 (NKI-L16), S145 (S6F1), S146 (MAY.035), S148 (25-3-1), S149 (CBR LFA-1/10), S150 (CBR LFA-1/9), S151 (CBR LFA-1/1), S152 (CBR LFA-1/4), S154 (CBR LFA-1/3), S158 (TS2/14), S159 (TS1/22), S160 (TS2/4), S161 (TS2/6), S163 (BL5), S167 (G-25.2), and S169 (YTA-1).

Molecular characterization

The molecular structure of CD11a has been determined [7] and a schematic diagram is shown at the beginning of this chapter. It is a transmembrane protein of M_r 180 000 on sodium dodecyl sulfate-polyacrylamide gel electrophoresis (SDS-PAGE) under reducing conditions [1,7]. The external portion of the molecule contains an I domain that plays a role in the ligand binding site and a metal-binding region that resembles the 'EF hand-like' loop found in a number of Ca^{2+} binding proteins [7]. The molecule contains a short cytoplasmic domain of 53 amino acids [7]. CD11a maps to chromosome 16 with the other two leucocyte integrin α subunits [8]. Antibodies within this group, with the exception of S150 (CBR LFA-1/9), S152 (CBR LFA-1/4), and S163 (BL5) (data not submitted), immunoprecipitate the heterodimer.

Cell surface expression

LFA-1 is expressed on lymphocytes, monocytes, and neutrophils [9]. The antibodies clustered here reacted with transfected cell lines expressing CD18/CD11a and not with cells expressing CD18/CD11b or CD18/CD11c. Epitope mapping [Huang and Springer, AS5.6] was performed using CD11a human × mouse constructs coexpressed with CD18 in COS cells. Flow cytometry of T- and B-cell lines as well as thymocytes yielded variable staining patterns among the antibodies in this subpanel. mAb S140 (NKI/L16) and S145 (S6F1) have previously been shown to recognize activation states of LFA-1 [10,11]. S140 (NKI-L16) stained only thymocytes, THP-1 cells, U937 transfectants, and thymocyctes from patients with severe combined immunodeficiency disease (SCID); mAb S145 (S6F1) exhibited a similar staining pattern except that it recognized THP-1 cells weakly and showed enhanced staining only with Raji–*tat* transfectants and fetal thymocytes. With the exception of S152 (CBR LFA-1/4), which was negative on all cell lines except for fetal thymocytes, the remaining antibodies stained most T- and B-cell lines and leucocytes.

Functional studies

Stimulation of cells with phorbol esters or activation of the T-cell receptor activates cellular adhesion

through LFA-1 [12,13]. mAb were examined for their ability to inhibit homotypic aggregation, cytotoxic cell–cell interactions, and binding to purified ICAMs or transfected cells [Petruzzelli *et al.*, AS5, Table 2]. mAb were also examined for stimulation of LFA-1 function [Petruzzelli *et al.*, AS5, Table 2]. mAb S140 (NKI-L16) has previously been shown to stimulate aggregation and activate binding of LFA-1 to ICAM-1 [1]; this was confirmed here by three investigators.

Immunohistochemistry

Immunohistochemical staining was carried out with this subpanel of antibodies on skin, tonsil, lymph nodes with and without follicular hyperplasia, small bowel with and without Crohn's disease, liver with and without hepatitis C infection, and synovial membranes with and without rheumatoid arthritis. Generally, tissue in the diseased state was somewhat more reactive and staining was found on lymphocytes, granulocytes, and macrophages. mAb S145 (MAY.035) was negative on all but synovial membranes from patients with rheumatoid arthritis, and S140 (NKI-L16) stained the synovial membranes and lymph nodes with follicular hyperplasia but was otherwise unreactive.

References

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