The LFA-1, Mac-1 glycoprotein family and its deficiency in an inherited disease[1]

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ABSTRACT

A family of functionally important, high-molecular-weight glycoproteins with identical β subunits has recently been defined on leukocyte cell surfaces. Soon after these molecules and at least some of their functions had been defined with monoclonal antibodies, an inherited disease, LFA-1, Mac-1 deficiency, was discovered in humans. This deficiency has confirmed that this glycoprotein family is of central importance in leukocyte cell surface adhesion reactions.—**Springer, T. A.** The LFA-1, Mac-1 glycoprotein family and its deficiency in an inherited disease. *Federation Proc.* 44: 2660–2663; 1985.

The properties of the Mac-1, LFA-1 family are summarized in Table 1. The lymphocyte function-associated 1 (LFA-1) molecule was first defined in the mouse by rat monoclonal antibodies (MAb) that inhibited T lymphocyte-mediated killing (15, 16, 25, 29, 30, 34, 36). Subsequently, human LFA-1 was defined by mouse MAb and was found to have the same functional properties (7, 17, 23, 28, 31). Anti-LFA-1 MAb inhibit the first step in T cell-mediated killing, i.e., the adherence of the killer cell to the target cell (9, 16, 22, 36). Non-antigen-specific natural killing (7, 17, 23, 35, 36) and antibody-dependent cellular cytotoxicity (21, 28) are also inhibited. These and other studies (36) suggest that LFA-1 acts as a cell adhesion molecule that can synergize with antigen-specific, Fc, or other receptors to increase the avidity of cell-to-cell interactions. The importance of LFA-1 in leukocyte adhesion is not limited to immune interactions. LFA-1 MAb inhibit spontaneous (27, 29) and phorbol ester-induced (38) homotypic aggregation of B and T cell lymphoblastoid lines.

Mac-1 was described as a mouse differentiation antigen (37) 2 years before the discovery of LFA-1. Subsequently, human Mac-1 was defined by cross-reaction of MAb with mouse Mac-1 (6), and the OKM1 and Mo1 MAb to other determinants on human Mac-1 were obtained (10, 40). Mouse and human Mac-1 are present on macrophage/monocytes, granulocytes, and large granular lymphocytes, but not on lymphocytes (6, 10, 18, 20, 37, 40). After it was realized that Mac-1 and LFA-1 share a common β subunit (24), the hypothesis that Mac-1 might also have an adherence function was investigated by testing anti-Mac-1 MAb for inhibition of macrophage functions. MAb to Mac-1 selectively blocked the complement receptor type 3 (CR3) on mouse and human macrophages and granulocytes (8). The CR3 mediates adherence and phagocytosis of particles opsonized with iC3b, a cleaved, hemolytically inactive form of the third component of complement. $F(ab')_2$ anti-Mac-1 does not inhibit macrophage Fc receptors or the complement receptor type 1 (CR1). MAb to the Mo1, OKM10, and in some cases the OKM1 determinants on the human Mac-1 molecule also inhibit the CR3 (5, 13, 32, 42). Noninhibitory MAb binding to other determinants on Mac-1 have been obtained (33, 42). Such MAb-Mac-1 antigen complexes bound to *Staphylococcus aureus* bacteria specifically agglutinate iC3b-opsonized erythrocytes (42), which confirms that the Mac-1 molecule has CR3 activity. An interesting parallel between CR3 adherence and the adhesion events in which LFA-1 participates is their mutual dependence on divalent cations (36, 43). This is in contrast to CR1, complement receptor type 2, and Fc receptor adherence.

The members of the LFA-1, Mac-1 glycoprotein family contain identical β subunits noncovalently associated with different α subunits (Table 1).

* PHENOTYPING OF MACROPHAGES, DENDRITIC CELLS, AND NK CELLS—Minisymposium presented at the 68th Annual Meeting of The American Association of Immunologists, St. Louis, Missouri, June 4, 1984. Chaired by T. A. Springer. Accepted for publication July 30, 1984.

[1] Studies from the author's laboratory supported by National Institutes of Health grants CA 31798 and CA 31799 and by Council for Tobacco Research grant CTR 1307.

[2] Recipient of an American Cancer Society Faculty Award.

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Cross-linking experiments have shown the quaternary structure of each antigen is $\alpha_1\beta_1$ (26, 32). The β subunits are identical by peptide mapping (24, 41) in isoelectric focusing (32) and by complete immunological cross-reactivity (33). The α subunits differ by the same criteria.

Studies of dissociated subunits have clarified both MAb specificity and the interrelationships of this glycoprotein family in mouse and human (32, 33). Antibodies specific for LFA-1 react with the αL subunit; those specific for Mac-1 react with αM. Antibodies to the β subunit react with LFA-1, Mac-1, and a third member of the family, p150,95. p150,95 contains the αX subunit for which specific MAb have not yet been obtained.

The NH_2-terminal amino acid sequences of the murine Mac-1 and LFA-1 α subunits show 33% identity (39). This homology suggests that their genes arose by a duplication event. The identity between the β subunits suggests that the divergence between the α subunits confers adhesive specificity. Blocking experiments with subunit-specific MAb are in accord with this idea (32, 33, 42).

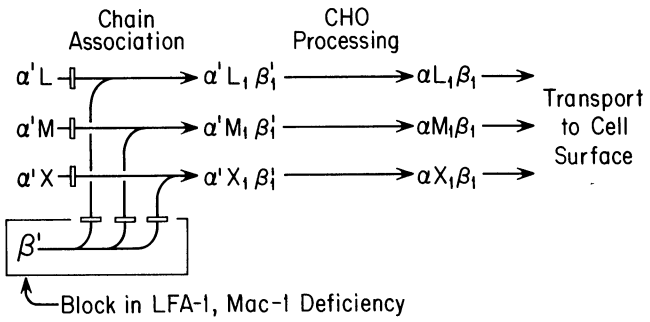

Figure 1. Biosynthesis of the LFA-1, Mac-1 family and postulated block in LFA-1, Mac-1 deficiency.

The first clue to LFA-1, Mac-1 deficiency came when total, surface-labeled granulocyte proteins from certain patients with recurring bacterial infections were subjected to sodium dodecyl sulfate–polyacrylamide gel electrophoresis. A lack was noted of a glycoprotein variously estimated as 110 (12), 180 (11), 150 (4), or 138 kilodaltons (1). Deficiency of a specific surface molecule was pinpointed in this laboratory by using ^{125}I-labeled anti-Mac-1 MAb and patients' granulocytes provided by Drs. J. Pitt and M. A. Arnaout (Table 2). Subsequently, similar results were obtained with Mo1 and OKM1 MAb, which bind to other determinants on the Mac-1 molecules, and with the TS1/18 anti-β subunit MAb (1, 14). The common β subunit suggested that LFA-1 should be tested, and it also was found to be deficient (see refs 2, 3). The lack of the entire LFA-1, Mac-1, p150,95 glycoprotein family on the surface of these patients' cells suggests that the primary defect may be in the common β subunit.

Previous work on the biosynthetic pathway of the LFA-1, Mac-1 family (19, 32), summarized in Fig. 1, is important in understanding the molecular basis of the inherited disease. The subunits are synthesized as separate precursors denoted α' and β'. Precursors associate into $\alpha'\beta'$ complexes. The subunits increase in M_r during processing, because of the conversion of high mannose to complex-type carbohydrate (T. Kishimoto, L. Sastre, and T. Springer, unpublished). The mature $\alpha\beta$ complexes are then transported from inside the cell to the surface.

It appears that the primary deficiency in patients' cells is of the β subunit. Normal α' precursor is made, but is not processed or transported to the cell surface in the absence of β, i.e., $\alpha\beta$ complementation is required for surface expression (2).

The accompanying articles by Anderson et al. (2) and Arnaout et al. (3) not only describe a novel molecular immunodeficiency disease, but also dramatically support and extend previous conclusions about the structural interrelationships and function of these molecules on normal cells, and emphasize their general importance in the biology of leukocyte adhesion reactions.

TABLE 1. The LFA-1, Mac-1 family

Mouse Human	LFA-1 LFA-1	Mac-1 Mac-1 (OKM1, Mo1)	p150,95
Subunits[a]	αL; β	αM; β	αX; β
$M_r \times 10^{-3}$	180; 95	170; 95	150; 95
Function	Adhesion	Complement receptor type 3	?
Cell distribution	Lymphocytes, monocytes,[b] granulocytes, large granular lymphocytes	Macrophages, monocytes, granulocytes, large granular lymphocytes	Monocytes, granulocytes, others?

[a] Noncovalent $\alpha_1\beta_1$ association; β subunits are identical. [b] Positive on blood monocytes and weakly positive on some macrophage lines; negative on thioglycolate-elicited macrophages.

TABLE 2. Anti-Mac-1 ^{125}I-labeled MAb binding to normal and patient granulocytes[a]

Cells	Preincubation with unlabeled MAb	^{125}I-labeled anti-Mac-1, counts/min bound	Specific binding, counts/min	Binding, % normal
Patient	Control	256	48	2
Patient	Anti-Mac-1	208		
Control	Control	2611	2348	100
Control	Anti-Mac-1	263		

[a] Granulocytes (6×10^5 in 50 μl) were preincubated with 20 μl of 100 μg/ml M1/70 anti-Mac-1 MAb or control M1/69HK MAb; ^{125}I-labeled M1/70 MAb was then added. After 0.5 h at 4 C, cells were washed and γ-radiation was counted.

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