CD31 Expressed on Distinctive T Cell Subsets Is a Preferential Amplifier of β1 Integrin-mediated Adhesion

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Summary

The CD31 (platelet endothelial cell adhesion molecule-1 [PECAM-1]/endothelial cell adhesion molecule [endoCAM]) molecule expressed on leukocytes, platelets, and endothelial cells is postulated to mediate adhesion to endothelial cells and thereby function in immunity, inflammation, and wound healing. We report the following novel features of CD31 which suggest a role for it in adhesion amplification of unique T cell subsets: (a) engagement of CD31 induces the adhesive function of β1 and β2 integrins; (b) adhesion induction by CD31 immunoglobulin G (IgG) monoclonal antibodies (mAbs) is sensitive, requiring only bivalent mAb; (c) CD31 mAb induces adhesion rapidly, but it is transient; (d) unique subsets of CD4+ and CD8+ T cells express CD31, including all naive (CD45RA+) CD8 T cells; and (e) CD31 induction is selective, inducing adhesive function of β1 integrins, particularly very late antigen-4, more efficiently than the β2 integrin lymphocyte function-associated antigen-1. Conversely, CD3 is more effective in inducing β2-mediated adhesion. Taken together, these findings indicate that unique T cell subsets express CD31, and CD31 has the capacity to induce integrin-mediated adhesion of T cells in a sensitive and selective fashion. We propose that, in collaboration with other receptors/ligands, CD31 functions in an “adhesion cascade” by amplifying integrin-mediated adhesion of CD31+ T cells to other cells, particularly endothelial cells.

Regulated adhesion is critical to virtually all the functions of T lymphocytes. These functions include both antigen-independent processes such as lymphocyte recirculation/homing and antigen-specific recognition events. Consequently, evolution has provided multiple modes of regulation of T cell adhesion. These include regulated expression of the T cell adhesion receptors such as very late antigen (VLA)1 integrins and L-selectin (LAM-1/Leu-8); regulated expression of the ligands such as intracellular adhesion molecule-1 (ICAM-1), vascular cell adhesion molecule-1 (VCAM-1), and E-selectin (endothelial leukocyte adhesion molecule-1 [ELAM-1]); and regulated function of the T cell adhesion receptors.

It is rapidly becoming apparent that this last mode of regulation, namely regulated function of the adhesion receptors, is powerful and widely used, not only by T cells, but by other cell types (1-10). Regulated function is a prominent characteristic of the integrin adhesion molecules, which are a diverse family of heterodimeric adhesion receptors used by virtually all cell types in adhesion to other cells and to extracellular matrix (11-14). Resting T cells express at least five integrins (11, 15). LFA-1 (αLβ2), the best known integrin on resting
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T cells, mediates primarily cell–cell interactions via at least three distinct ligands: ICAM-1, ICAM-2, and ICAM-3 (12, 16). In addition, there are four β1 integrins (VLA-3, VLA-4, VLA-5, and VLA-6) that mediate adhesion both to extracellular matrix via fibronectin (FN) and laminin (LN) (15) and to other cells via VCAM-1 (17). It is remarkable that integrins on circulating (“resting”) T cells do not mediate effective adhesion. Dustin and Springer (1) demonstrated that cross-linking of the CD3/TCR complex induces transient adhesion of regulated integrin function on T cells has been generalized by extensions in a variety of directions: (a) the β1 integrins show similar regulated function on resting T cells (15); (b) antigen-specific recognition induces β1 integrin function on T cells (18); and (c) integrin function on T cells is regulated not only by CD3 cross-linking (and the similar activation stimulus provided by pairs of CD2 mAb), but also by cross-linking of other receptors on the T cell surface, including CD7, CD28, and CD44 (1, 15, 19, 20). These findings for T cells have been both foreshadowed and complemented by a variety of findings regarding regulated function of integrins on other cell types including particularly platelets, granulocytes, and B cells (3–7, 10).

We use the terms adhesion “inducer” or “amplifier” to refer to a molecule on the T cell surface that augments integrin function. The nature of regulated adhesion makes the amplifier molecules as critical to the process as the adhesion molecule itself. In particular, differential expression of such amplifier molecules will be as important in T cell differentiation as differential expression of the adhesion molecule. The present report identifies CD31 as an amplifier molecule on unique subsets of T cells, and characterizes novel features of that adhesion induction. CD31 glycoprotein (also designated PECAM-1, platelet endothelial cell adhesion molecule) is an Ig superfamily member that is most similar in structure to classical adhesion molecules such as ICAM-1, VCAM-1, and neural cell adhesion molecule (NCAM) (21). It is expressed at high density on endothelium, platelets, granulocytes, and monocytes (22–24). It is also expressed by lymphocytes (23, 24). It has been implicated in cell–cell adhesion by a variety of findings. It accumulates at contact regions between endothelial cells (22), transfection of CD31 into L cells causes them to aggregate (25), and CD31 mAbs inhibit endothelial cell contact, as well as transfected L cell aggregation (25). CD31 has been postulated to bind to CD31 in homophilic interactions, as well as to participate in heterophilic interactions involving proteoglycans (22, 26). The present report demonstrates a role for CD31 in adhesion induction on T cells, and proposes a model of its potential involvement in an “adhesion cascade” on T cells.

Materials and Methods

Human T Cell Subsets. Highly purified CD4 T cells, CD8 T cells, and naive (CD45RA+CD45RO−) CD8 T cells were prepared from PBMC of volunteer research healthy donors by exhaus-

Adhesion Assay. Adhesion assays were performed essentially as previously described (15). Purified VCAM-1 (80 ng/ml), FN (1 μg/well), ICAM-1 (6 ng/well), collagen type I (1 μg/well), fibrinogen type I (1 μg/well), and control BSA (3% solution) were applied to 96-well microtiter plates (Costar, Cambridge, MA) in

Antibodies and other Reagents. The following mAbs were used as purified Ig: CD31-specific mAb NIH31-1 and NIH31-2 were generated and their specificity documented by binding to CD31-transfectants (data not shown); CD31 mAb PECAM-1.2 (K. Newman, unpublished observations); CD31 mAb 4G6 (S. M. Albelda, unpublished observations); CD31 mAb SGL34 (29) (S. Goyert, Cornell University Medical College, Manhasset, NY); CD31 mAb LAK1 (30) (M. Zocchi, Laboratory of Adoptive Immunotherapy, Milan, Italy); CD31 mAb L33 (D. Buck, Becton Dickinson & Co., San Jose, CA). Other mAbs are as follows: CD11b mAb NIH11b-1, CD49d mAb NIH49d-1, CD44 mAb NIH44-1 (31) and CD45 mAb NIH45-2 (generated locally), CD3 mAb OKT3, CD4 mAb OKT4, CD14 mAb 63D3, class II mAb IVA12, CD7 mAb 3A1, anticytotoxic mastocyte mAb 10F7 (all from American Type Culture Collection, Rockville, MD), CD2 mAb 95-5-49 (R. Queziones, Children’s Hospital Medical Center, Washington, DC), CD8 mAb B9.8.4 (B. Malissen, Centre National de la Recherche Scientifique, Marseilles, France), CD19 mAb FMC63, CD45RA mAb FMC71 (H. Zola, Flinders Medical Center, Bedford Park, Australia), CD45RO mAb UCHL1 (P. Beverley, Courtaulds Institute of Biochemistry, London, UK), CD18 mAb MM123 (J. E. Hildreth, Johns Hopkins Medical School, Baltimore, MD), CD49d mAb L25 (D. Buck), CD29 mAb MAB13, CD49e mAb MAB25 (both from Advanced Magnetics, Cambridge, MA) and/or Dynabeads Dynal Inc., Fort Lee, NJ), and a cocktail of mAbs consisting of MHC class II mAb IVA12, CD19 mAb FMC63, CD16 mAb CD11b mAb NIH11b-1, CD14 mAb 63D3, anticytotoxic mastocyte mAb 10F7, and CD45 mAb OKT4 or CD8 mAb B9.8.4 with or without CD45RA mAb FMC71 (to negatively isolate memory T cells), or CD45RO mAb UCHL1 (to negatively isolate naive T cells) (27). The anti–HLA-DR mAb IVA12 was included in the selection cocktail to exclude the normal low percentage of circulating activated T cells. Furthermore, the CD8 dull (dim) population which has NK-like features phenotypically and functionally (28), was also excluded from the CD8+ population by the use of a separation cocktail containing the CD16 mAb VD2 and the CD11b mAb NIH11b-1. The purity of T cell subsets were >96% CD4+ or >94% CD8+ and >99% CD45RA+ or >99% CD45RO+, as determined by flow cytometric analysis.

Adhesion Assay. Adhesion assays were performed essentially as previously described (15). Purified VCAM-1 (80 ng/ml), FN (1 μg/well), ICAM-1 (6 ng/well), collagen type I (1 μg/well), fibrinogen type I (1 μg/well), and control BSA (3% solution) were applied to 96-well microtiter plates (Costar, Cambridge, MA) in
Ca/Mg-free PBS at 4°C overnight. Binding sites on plastic were subsequently blocked with Ca/Mg-free PBS/3% BSA for 2–3 h at 37°C to reduce nonspecific attachment. Cells were plated onto 96-well plates (Costar) and cultured to confluence. Plates were washed three times with PBS before the addition of 50,000 51Cr-labeled T cells to each well in a final volume of 100 µl PBS/0.5% human serum albumin (HSA). mAbs (1 µg/well) were added to relevant wells. After a settling phase of 30 min at 4°C, which also allowed mAb binding, plates were rapidly warmed to 37°C for 15 min, and nonadherent cells were washed off. Well contents were lysed with 1% Triton X-100, and 3' emissions of well contents determined. Background binding of T cells to BSA or collagen was 1–7%. Data were expressed as mean percentage and SE of binding of T cell subsets from representative individuals. Crosslinking of CD3 and CD31 on T cells or T cell subsets was performed as described (15) by 30 min preincubation with relevant mAbs at 4°C and washing before addition to triplicate wells containing 0.05 µg goat anti–mouse Ig. When not being crosslinked, CD31 mAb was added at the beginning of the settling phase.

**Results and Discussion**

CD31 Is Expressed on Unique Subsets of T Cells. The complexities of T cell migration/homing are most readily understood in terms of regulated adhesion of different T cell subsets to different apposing surfaces, particularly endothelial cells. Our previous studies have emphasized differential regulation of adhesion molecules on different T cell subsets, and their relevance to cell–cell adhesion (2). Our interest in CD31 was first stimulated by observing that CD31 was differentially expressed on subsets of circulating T cells. Our comparisons between CD31 and other markers of T cell subsets indicate that CD31 is expressed on unique subsets of T cells (Fig. 1). CD31 heterogeneity does not correlate precisely with either of the two best understood dichotomies within T cells: CD4 vs. CD8 and CD45RA (“naive”) vs. CD45RO (“memory”). Nevertheless, there are biases towards higher frequency of CD31+ cells among CD8+ cells and among CD45RA+ cells. The conclusions from combined analysis of CD4/8 and CD45RA are that: among CD8+ cells, typically 90% express CD31, all of the naive (CD45RA+) cells and about half of the memory (CD45RA−) cells (Fig. 1 B); and among CD4 cells, typically 20% express CD31, about half of the naive (CD45RA+) cells and few of the memory (CD45RA−) cells (Fig. 1 A). This heterogeneity does not

**Figure 1.** Unique expression of CD31 antigens on resting peripheral T cell subsets. Histograms for CD31 on naive (CD45RA+) subpopulations (dotted line) and memory (CD45RA−) subpopulations (solid line) of purified resting peripheral CD4+ cells (A) and CD8+ T cells (B). Analyses were carried out with the CD45RA mAb Leu-18-FITC and the CD31 mAb SG134. The fraction of CD45RA+ cells is 34% for the CD4 and 65% for the CD8 preparation.
correspond to reactivity of any of the more than 50 molecules whose expression we have examined on T cells (data not shown). The bias toward CD31 expression on CD8 + cells and naive (CD45RA +) cells is remarkable since most adhesion molecules are similarly expressed on CD4 vs. CD8 cells (Y. Tanaka, unpublished observations), and of the many adhesion molecules differentially regulated on T cells, most are preferentially expressed on memory (CD45RA-) cells (2, 36-38).

CD31 mAbs Induce Integrin-mediated Adhesion of Resting Peripheral Human T Cells. Preliminary studies demonstrated that CD31 mAbs induced integrin-mediated adhesion. As expected, the induction of adhesion by CD31 is seen only in purified T cell fractions which include CD31 + T cells (data not shown). Since CD31 is uniformly positive on naive (CD45RA +) CD8 cells (Fig. 1), we undertook the most systematic analysis of adhesion induction on that subset of cells. Purified naive (CD45RA +) CD8 cells show augmented adhesion to the integrin ligands FN and VCAM-1 when CD31 mAbs are present during the assay (Fig. 2). The uniqueness of CD31 mAb-induced adhesion to each ligand is illustrated by the comparison with six different control mAbs, four of which (CD7, CD28, CD3, and CD44) have been described to be inducers of integrin-mediated adhesion of T cells. None of these control mAbs cause marked induction of adhesion in the absence of additional crosslinking (see below). In contrast, adhesion is augmented by most of the CD31 mAbs without additional crosslinking. The two CD31 mAbs that are least effective in induction of adhesion are the two mAbs that bind to the most membrane-proximal domains of CD31 (S. M. Albelda, unpublished observations).

To confirm that T cell binding to purified immobilized ligand is a valid model of integrin-mediated adhesion, the critical features of CD31-induced adhesion were reproduced for T cell binding to an L cell transfected with VCAM-1 (Fig. 3). About 20% of the resting CD8 naive cells bound to the VCAM-1 transfectant, and this binding was doubled by pretreatment with CD31 mAbs. Since VCAM-1 is expressed on the transfected cells at a level severalfold lower than on activated endothelial cells (data not shown), these data indicate that CD31-induced adhesion could be relevant to T cell adhesion to VCAM-1 expressing endothelium.

It is noteworthy that the adhesion induced by the best CD31 mAbs approaches or equals that of PMA (Figs. 2 and 3), which is generally the strongest pharmacologic inducer of T cell adhesion. To determine whether PMA and CD31 mAbs might activate cells in a complementary fashion, cells were activated by both CD31 and PMA (Fig. 4 A). The lack of demonstrable additive induction provided no evidence for distinct signaling pathways or activation of distinct subsets within this relatively homogenous population of CD8 naive (CD45RA +) cells. Additional controls in that experiment demonstrate that CD31-mediated induction does not nonspecifically alter adhesion of T cells to otherwise irrelevant extracellular matrix proteins (Figs. 4, B and C).

mAb blocking studies were performed to confirm that CD31-induced adhesion to the three ligands (FN, VCAM-1, and ICAM-1) was mediated by the integrin receptors on T cells which normally bind these ligands (Fig. 5). As expected, T cell binding to VCAM-1 and FN was mediated by integrins of the β1 family, and binding to ICAM-1 was mediated by integrins of the β2 family. More specifically, the VCAM-1 binding was mediated by VLA-4 and the FN binding...
was mediated primarily by VLA-5, with a small contribution from VLA-4. Functional inhibition by our newly generated NIH49d-1 mAb was identical to that of the reference VLA-4 mAb L25. Thus, the interactions of VLA-4/VCAM-1, VLA-4/FN, VLA-5/FN, and LFA-1/ICAM-1 induced by CD31 mAbs are consistent with those observed with other T cell populations and other inducing stimuli (19).

The capacity of CD31 IgG mAbs to induce adhesion in the absence of additional crosslinking by a polyvalent anti-Ig reagent (Fig. 2) seems to be a fundamental characteristic of CD31 IgG mAbs, which distinguish them from CD3 IgG mAbs, the prototypic inducers of T cell adhesion (1). When the issue of crosslinking is explored (Fig. 6), the results confirm that CD31 mAbs can induce in the absence of additional crosslinking, while CD3 IgG mAbs (and the other adhesion inducer molecules shown in Fig. 2) do not. CD31-induced adhesion is often augmented by crosslinking, but is almost always observed without it (Fig. 6, and data not shown). Thus, the CD31 "trigger" of adhesion appears to be a uniquely sensitive one requiring only dimer formation, since most CD31 IgGs tested induce adhesion (Fig. 2). Since Fab fragments of CD31 mAbs induce little adhesion (data not shown), the minimal stimulus in this system seems to be CD31 dimer formation, not receptor occupancy.

The kinetics of CD31 induction of adhesion to VCAM-1 were analyzed (Fig. 7). Induction by CD31 was rapid, regardless of the presence or absence of additional crosslinking. Induction by crosslinked CD3 was similar. The induced adhesion was gone by 60 min. Thus, the adhesion induced by CD31 resembles the rapid onset and decay of CD33-induced adhesion. This time course is consistent with models in which CD31-induced adhesion, like CD3-induced adhesion, plays a transient role in a coordinated sequence of events mediating T cell adhesion.

Differential Induction of Adhesion by CD31 vs. CD3. The fact that multiple surface molecules, including CD3, CD2, CD7, CD28, CD44, and now CD31 can regulate T cell adhesion, suggests that regulation of adhesion is a fundamental role served by a variety of cell surface molecules. Adhesion regulation would be most adaptive if different adhesion-inducing molecules preferentially regulated different adhesion receptors. We tested this possibility by comparing the ligand specificity of adhesion induced by crosslinked CD3 and CD31 in multiple donors and experiments (Fig. 8). Each point represents the differential binding of a particular preparation of T cells to two different ligands. When CD3 is used as the inducer (●), the T cells preferentially adhere to ICAM-1, as indicated by their position above the diagonal. Conversely,
preferentially induces LFA-1 function on some of these cells, and CD31 induces VLA-4 function in others.

Previous studies have fostered the concept of CD31 as an adhesion molecule per se. The present studies add a new perspective by showing that CD31 is an adhesion-inducing molecule. We favor the concept that CD31 mediates both weak and potent adhesion-induction, allowing it to serve as an adhesion amplifier. This kind of molecule would be the missing element in our understanding of T cell–endothelial cell adhesion as a cascade consisting of a coordinated series of receptor–ligand interactions which includes: (a) initial tenuous adhesion (tethering); (b) triggering; (c) integrin-mediated strong adhesion (glue); and (d) subsequent detachment (8). Studies of granulocytes indicate that the initial tethering is usually served by molecules of the selectin family (40). L-selectin may function in that role for many T cells (41). Strong adhesion is most likely mediated by the integrins LFA-1 and VLA-4 (2, 35, 42), however, this cannot occur on resting T cells in circulation until the integrins become functionally activated. The intervening step by which the integrins become activated is not understood. Obviously, the CD3/T cell receptor would not be expected to be involved in T cell–endothelial cell interactions. L-selectin is a good candidate, but has not been shown to induce integrin function.

CD31 is an excellent candidate for an adhesion amplifier in T cell–endothelial cell interactions, given the adhesion-inducing capacity of CD31 shown in the present studies, and its demonstrated role in cell interactions with endothelial cells (22). If analogous CD31 engagement occurs when CD31+ T cells bind to endothelium, then our results predict that such CD31 crosslinking will induce integrin-mediated adhe-
phasemal by the finding that dimer formation may be sufficient for triggering (Figs. 2 and 6) and that the adhesion induc-
its distal parts readily available early in cell-cell interaction. Our studies indicate that CD31 is particularly effective in
inducing cell interactions, since it is increasingly apparent how impor-
tant VLA-4 is in T cell interactions with endothelium in vitro, and ultimately in T cell recirculation in vivo (17, 35, 43–48).
We are currently testing the hypothesis that CD31 contributes to T cell–endothelial cell interactions.

Recent in vivo studies in the rat indicate effects of an anti-VLA-4 mAb on T cell movement into various sites, but
most dramatically into gut (48). Until now, there has been no explanation why there is such a predominance of CD8
rather than CD4 cells (90% CD8) among intraepithelial lymphocytes of the gut mucosa (49). We propose that the preferen-
tial expression of CD31 on CD8 cells rather than CD4 cells (typically 80 vs. 20%), together with its selective capacity
to induce VLA-4 function, may contribute to the preferential movement of CD8 cells into gut epithelium. In addition to
the conventional VLA-4 molecule (α4β1), there is an α4-containing integrin (α4β7) on gut-homing T cells which mediates interactions with a ligand on specialized gut endo-
thelium, Peyer’s patch high endothelial venule (HEV) (50, 51). It remains to be determined whether this integrin is also
activated functionally by engagement of CD31.

In the foregoing description of T cell–endothelial cell inter-
actions, the postulated adhesion-amplifying role for CD31 is
analogous to the adhesion–amplifying role played by CD3
in antigen-specific T cell interactions (1). We view these as
distinct but generally homologous adhesion cascades. More
generally, we expect that there will be multiple T cell adhe-
sion cascades. Each T cell will have the potential for many
adhesion cascades in its repertoire for use in interactions with
different cells or extracellular matrix. Different T cell subsets
will have different specialized repertoires of adhesion cascades.

Obviously, CD31 can be an inducer of adhesion only for those
unique subsets of T cells which express it. It is also apparent
that there must be and are other adhesion-inducing mole-
cules on resting T cells.

Although the foregoing data prompt us to propose a spe-
cial role for CD31 in T cell–endothelial cell interactions in
the gut, we suspect that it will also be important for T cells
in other contexts. The preferential expression of CD31 on
naïve cells raises the possibility that it may contribute to the
process of migration of naïve cells into lymph node (52). Three
lines of evidence indicate that VLA-4 ligands may exist in
lymph node HEV, and therefore are consistent with this hy-
thesis. First, VCAM-1 can be expressed on endothelium in
lymph nodes draining from sites of antigen stimulation (46). Second, VLA-4 mAb inhibits migration of several cate-
gories of T cells to peripheral lymph node (48). Finally, inhi-
bition studies with a peptide sequence from the III–CS site
of FN indicate that it inhibits lymphocyte binding to cul-
tured lymph node HEV (44). This suggests that VLA-4 may
be involved in binding to a ligand on these HEVs. Thus, CD31
and VLA-4 may be important for migration of some T cells
through lymph node HEV. Furthermore, given data that
CD31 may participate in homophilic interactions with CD31
(25), T cell CD31 may contribute to T cell interaction with
not only CD31+ lymph node HEVs, which express CD31
better than endothelial cells (53), but also with CD31+ APCs. Finally, Stockinger et al. (23) have demonstrated that
CD31 mAbs induce reactive oxygen metabolites from mono-
cytes. CD31 thus appears capable of contributing to the regu-
lation of cellular processes in addition to adhesion.

It is evident that CD31 does not act alone, but rather in
the context of other molecules which constitute the cascade.
T cells floating in a sea of CD31+ cells in circulation do not
have their integrins activated. Furthermore, only some of the
CD31+ T cells become detectably adhesive when exposed
to CD31 mAbs. Therefore, a permissive role for a suitable
tethering molecule and potentially other cofactors is likely
to be a prerequisite for physiologic CD31 triggering. We pre-
dict a role for CD31 in T cell subset adhesion to specialized
endothelial cells, where the combinatorial requirements of
tether, trigger, and glue offer enormous flexibility in an adhe-
sion cascade.

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References


by purified adhesion ligands VCAM-1, ICAM-1 but not ELAM-1. J. Exp. Med. 174:901.


