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The Role of E-Selectin, P-Selectin, and Very Late Activation Antigen-4 in T Lymphocyte Migration to Dermal Inflammation

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T lymphocyte infiltration into inflamed tissues is thought to involve lymphocyte rolling on vascular endothelial cells. Because both selectin and \( \alpha_4 \) integrin adhesion molecules can mediate leukocyte rolling, the contribution of these receptors to lymphocyte migration to inflammation was examined. The recruitment of \( ^{111} \)In-labeled spleen T cells to intradermal sites injected with IFN-\( \gamma \), TNF-\( \alpha \), LPS, polyinosine-cytosine, and Con A was measured in the rat, and the effect of blocking mAbs to E-selectin, P-selectin, very late activation Ag-4 (VLA-4), and LFA-1 was determined on this T cell migration in vivo. Anti-E-selectin and anti-P-selectin mAbs each inhibited 10–40 and 20–48%, respectively, of the T lymphocyte migration to the inflammatory sites, depending on the stimulus. Blocking VLA-4 inhibited 50% of the migration to all of the lesions except Con A. Treatment with both anti-VLA-4 and anti-E-selectin mAbs inhibited up to 85% of the lymphocyte accumulation, while P-selectin and VLA-4 blockade in combination was not more effective than VLA-4 blockade alone in TNF-\( \alpha \), IFN-\( \gamma \), LPS, and polyinosine-cytosine lesions. Inhibiting E-selectin, P-selectin, and VLA-4 together nearly abolished lymphocyte migration to all inflammatory sites. Anti-LFA-1 mAb strongly inhibited lymphocyte accumulation by itself, and this inhibition was not significantly further reduced by E- or P-selectin blockade. Thus, T cell migration to dermal inflammation is dependent on E-selectin, P-selectin, and VLA-4, likely because these three receptors are required for rolling of memory T lymphocytes, but VLA-4 and E-selectin are especially important for lymphocyte infiltration in these tissues.


Lymphocytes form an important part of the leukocyte infiltrate in many inflammatory conditions including cutaneous inflammation, such as psoriasis, contact dermatitis, and atopic dermatitis. The mechanisms of T cell migration from the blood into these tissues are incompletely understood. Four families of adhesion molecules, including the selectins, the integrins, sialomucins, and Ig supergene family molecules, are believed to mediate the capture and rolling of lymphocytes on the blood vessel endothelium and the firm adhesion and transendothelial migration steps (1, 2).

Based on previous studies, it is clear that the integrins LFA-1 (\( \alpha_4 \beta_2 \); CD11a/CD18) and very late activation Ag-4 (VLA-4)\(^3\) (\( \alpha_4 \beta_1 \); CD49d/CD29) mediate T lymphocyte recruitment to cutaneous inflammatory reactions involving delayed-type hypersensitivity (DTH) (1, 3–8). LFA-1 and VLA-4 can mediate adhesion of lymphocytes to endothelium, and \( \alpha_4 \) integrins have been shown to also mediate the rolling of lymphocytes on vascular endothelial cells in vitro and in postcapillary venules in vivo (9, 10). The members of the selectin family of adhesion molecules, including E- and P-selectin expressed on endothelium at sites of inflammation, have also been shown to mediate the rolling of lymphocytes on vascular endothelium in vitro (11–13). However, the importance of E- and P-selectin in recruitment of T lymphocytes to inflamed tissues is less well understood. E-selectin may be especially important for T lymphocyte migration to cutaneous inflammation, because a subset of T cells found enriched in human inflammatory dermatoses expresses a specific ligand for E-selectin, i.e., the cutaneous lymphocyte Ag (CLA) (14, 15). In vitro stimulated and adoptively transferred T cells of the Th1 phenotype in the mouse express ligands for P-selectin and/or E-selectin (16). These T cells have been shown in the mouse to use P-selectin for migration to inflammatory sites, such as DTH reactions and arthritis joints, and to peritoneal inflammation (16–19). A subset of unactivated T lymphocytes also expresses P-selectin glycoprotein ligand-1, which, when properly glycosylated, can bind to P-selectin (20).

Although the above studies suggest a role for these selectins in T lymphocyte or lymphoblast recruitment, the contribution of E- and P-selectin to resting T lymphocyte migration to sites of inflammation, such as the skin in vivo, in response to cytokines, cytokine inducers, and polyclonal activators has not been investigated. Furthermore, the relative contribution of adhesion molecules involved in rolling of lymphocytes, i.e., E- and P-selectin and the \( \alpha_4 \) integrin, VLA-4, has not been systematically examined or compared with the role of LFA-1, which mediates primarily firm adhesion and transmigration rather than leukocyte rolling. The studies presented in this work demonstrate a variable role in T cell recruitment for both E-selectin and P-selectin depending on the inflammatory stimulus, a marked contribution of the integrin VLA-4, and a substantially greater contribution for E-selectin in combination with VLA-4 than P-selectin in mediating T lymphocyte migration to dermal inflammation to most stimuli.

Materials and Methods

Animals and reagents

Male Lewis strain rats of 200–250 gm (Harlan Sprague-Dawley, Indianapolis, IN) were used in all experiments. Rat IFN-\( \gamma \) was generously provided by Dr. P. Van Der Meide (TNO Primate Center, Rijswijk, The Netherlands) and murine rTNF-\( \alpha \) was a kind gift from Genentech (South San Francisco, CA). Escherichia coli 011B4 LPS was from List Biologicals (Campbell, CA), and polyinosine-cytosine (poly(I:C)) and Con A were

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2 Abbreviations used in this paper: VLA-4, very late activation Ag-4; CLA, cutaneous lymphocyte Ag; DTH, delayed-type hypersensitivity; poly(IC), poly inosine-cytosine.
purchased from Sigma-Aldrich (St. Louis, MO). mAbs used included TA-2, a mAb to rat α4, and TA-3, a mAb to rat LFA-1 (α2 specific) (5, 21). RME-1 is a mAb against rat E-selectin and mAb RMP-1 recognizes rat P-selectin (22, 23); these are mouse IgG1 mAbs generated in our laboratory, and each of these blocks the adhesion function of its respective Ags based on in vitro and in vivo studies (22–24). Isotype control mAb was mAb B9 (IgG) reactive with pertussis toxin (25).

**In vivo migration assay**

Spleen T lymphocytes were isolated from donor rats as previously described (5). Briefly, the spleen was minced to obtain a cell suspension and RBCs were lysed with 0.84% NH₄Cl. T cells were purified by passage through a nylon wool column. The recovered cells were 91–93% CD3⁺, 2–4% surface Ig⁺, <2% neutrophils and monocytes, and contained 60–65% CD4⁺/8⁺ and 30–35% CD4⁺/8⁺ lymphocytes.

The T lymphocytes were labeled with ¹¹¹In-labeled oxine and their in vivo migration was measured as previously described (5). Briefly, 5 × 1⁰⁷ cells suspended in 0.5 ml of medium were labeled with 3.5 µCi of ¹¹¹In-labeled oxine for 10 min, washed, and resuspended for injection. Immediately afterward, where indicated, mAbs RME-1 and/or RMP-1 (1–2 mg), TA-2 (2 mg), TA-3 (3 mg), or B9 (2 mg), as a control, were administered i.v. The skin on the back of the animal was then shaved and 0.05 ml of test sample containing 300 U of IFN-γ, 10 ng of TNF-α, combinations of these two cytokines, 100 ng of LPS, 200 ng of poly(IC), or 10 µg of Con A were injected intradermally, using 30-gauge needles, into duplicate or triplicate skin sites. Each of these stimuli has been previously shown to recruit T lymphocytes to dermal sites over 6–20 h (26, 27). Diluent medium (RPMI 1640, 0.1% pyrogen free human serum albumin) was injected in control skin sites. Animals were sacrificed 20 h later, the skin on their backs was removed, and the injected areas were punched out with a leather punch. This produced a circular piece of tissue 12 mm in diameter with a standard surface area. At time of sacrifice, samples of blood, spleen, cervical, axillary, and mesenteric lymph nodes, and Peyers’ patches were taken. All samples were analyzed for ¹¹¹In content in a Wallac Wizard III gamma radioisotope counter (Wallac, Gaithersburg, MD). Results are expressed as cpm of ¹¹¹In accumulated per 20-h migration period per 10⁶ cpm injected on labeled cells.

**Immunofluorescence flow cytometry**

The expression of ligands for E-selectin and P-selectin on T cells was determined using mouse E-selectin and mouse P-selectin chimeras constructed fused to human μ-chain (kind gift from Drs. J. Lowe and L. Stoolman, University of Michigan, Ann Arbor, MI) as reported previously (28). Briefly, the purified T cells were incubated (45 min at 4°C) with either E- or P-selectin chimera constructs in immunofluorescence buffer. Binding was detected by using sequential incubation with biotin-labeled mouse anti-human μ-chain (Zymed Laboratories, San Francisco, CA) followed by washes and incubation with streptavidin-conjugated PE (Sigma-Aldrich). Controls included chimera plus 10 mM EDTA during the staining procedure to eliminate cation-dependent binding.

For double staining of IL-2R and selectin ligands, T cell IL-2R expression was determined using mAbs MRC OX-39 (Serotec, Oxford, U.K.), followed by FITC-labeled sheep anti-mouse IgG conjugated (Sigma-Aldrich). The sheep anti-mouse IgG was then blocked with 5% mouse serum and washing was conducted as above with the E-selectin and P-selectin chimeras. Samples were read immediately in a FACSCalibur flow cytometer (BD Biosciences, Mountain View, CA) with appropriate FL-1/FL-2 compensation. Twenty thousand lymphocytes were analyzed per sample.

**Statistics**

Data were expressed as means ± SEM of multiple animals. ANOVA and Student’s unpaired t test were used to compare the differences between means.

**Results**

**Effect of E- and P-selectin blockade on T cell migration to dermal inflammation**

The intensity of T lymphocyte accumulation in response to the different stimuli varied considerably, as shown in Fig. 1. The response to TNF-α and IFN-γ was the weakest, despite using an optimal dose for each stimulus. However, the combination of TNF-α with IFN-γ induced a synergistic increase in lymphocyte recruitment. The response to Con A was the greatest, with LPS and poly(IC) inducing intermediate responses. There was little effect of E-selectin or P-selectin mAb treatment on the TNF-α, IFN-γ, or TNF-α plus IFN-γ responses, or on the LPS or poly(IC) stimuli when only E- or P-selectin Ab was used. In contrast, the response to Con A was especially inhibited by anti-P-selectin treatment. In addition, the response to LPS, poly(IC), and especially Con A was inhibited by simultaneous blockade of E- and P-selectin with little effect of this combination of selectin blockade on the TNF-α and IFN-γ responses.

To more clearly visualize the effect of mAb treatment on the T cell recruitment induced by each stimulus, Fig. 2 shows the results normalized to the control response obtained with each stimulus. Treatment with mAb to E-selectin alone significantly inhibited the recruitment induced by TNF-α, LPS, and Con A, with no effect on the response to IFN-γ, TNF-α plus IFN-γ, or poly(IC). Ab to P-selectin, in contrast, inhibited the response to the combination of TNF-α plus IFN-γ and also significantly inhibited the response to LPS, poly(IC), and Con A. Blocking both E- and P-selectin also inhibited the response to all stimuli. The effect of mAb treatment on the T lymphocyte migration to cutaneous inflammatory sites is expressed as a percentage of the migration in control animals receiving isotype-matched mAb in response to the stimuli shown in Fig. 1. Each bar is the mean ± SEM of five to seven animals.

![FIGURE 1](http://www.jimmunol.org/) Effect of anti-E- and anti-P-selectin mAbs on T cell migration to dermal inflammation. Spleen T lymphocytes labeled with ¹¹¹In were injected i.v. together with mAbs to E- or P-selectin or an isotype-matched control mAb as indicated. Each animal also received intradermal injections of TNF-α (10 ng), IFN-γ (300 U), TNF-α (10 ng) plus IFN-γ (300 U), LPS (100 ng), poly(IC) (200 ng), Con A (10 µg), or diluent control in duplicate sites on the back. After 20 h the recipients were euthanized and the ¹¹¹In accumulation in each skin site was determined. Control skin sites accumulated 14 cpm. Each bar indicates the mean ± SEM of five to seven animals.

![FIGURE 2](http://www.jimmunol.org/) Effect of anti-E- and anti-P-selectin mAbs on T cell migration to dermal inflammation. The effect of mAb treatment on T lymphocyte migration to cutaneous inflammatory sites is expressed as a percentage of the migration in control animals receiving isotype-matched mAb in response to the stimuli shown in Fig. 1. Each bar is the mean ± SEM of five to seven animals.
had no effect on IFN-γ-induced migration, and there was no additive effect by dual selectin blockade on TNF-α and TNF-α plus IFN-γ responses, which were only modestly (30–40%) inhibited. T cell recruitment induced by LPS and Con A was especially strongly inhibited by E- plus P-selectin blockade, with a 60–70% reduction of the response to these stimuli.

**Effect of VLA-4 plus E- or P-selectin blockade on T cell migration to dermal inflammation**

VLA-4 has been reported to mediate capture and rolling of lymphocytes (9, 10). Therefore, the contribution of VLA-4 in E-selectin- and P-selectin-independent T lymphocyte migration was evaluated. As shown in Fig. 3, blocking VLA-4 significantly inhibited T cell recruitment induced by TNF-α, IFN-γ, TNF-α plus IFN-γ, LPS, and poly(I:C) by 42–56%. In contrast, VLA-4 blockade had no effect on the response to Con A. When anti-VLA-4 was combined with mAb to E-selectin, there was a marked reduction of the lymphocyte migration to all of the stimuli, with 80% inhibition of the recruitment to most of the inflammatory sites. In almost all cases a much greater inhibition was observed with this combination than with either anti-VLA-4 or anti-E-selectin alone. Once again the response to Con A was most resistant to inhibition with VLA-4 plus E-selectin blockade, inhibiting T cell recruitment by only 40%.

The role of P-selectin in conjunction with VLA-4 was also examined (Fig. 4). Blockade of P-selectin did not significantly potentiate the inhibition of migration observed in the presence of blocking VLA-4 to most of the stimuli and was substantially less effective than blockade of E-selectin plus VLA-4 (Fig. 3). Anti-P-selectin treatment did not potentiate the inhibition by anti-VLA-4 mAb on the response to TNF-α, IFN-γ, and poly(I:C) and had only a small effect on the response to LPS and TNF-α plus IFN-γ. Similarly, VLA-4 blockade, which did not affect the Con A response, also did not potentiate the inhibitory effect of P-selectin blockade.

**Effect of combined blockade of VLA-4 and E- and P-selectin on T cell migration to dermal inflammation**

The results above showed that VLA-4 contributed substantially to T lymphocyte migration to most of the inflammatory sites examined and that blocking E-selectin together with VLA-4 markedly inhibited, but did not abolish, T cell accumulation in these sites. To determine whether P-selectin contributed to this residual lymphocyte infiltration, the effect of blocking both E- and P-selectin plus VLA-4 was determined and compared with anti-VLA-4 and anti-VLA-4 plus anti-E-selectin treatment (Fig. 5). Blocking P-selectin, in addition to VLA-4 and E-selectin, was significantly more effective at inhibiting migration to all of the lesions than was blocking VLA-4 and E-selectin, and blocking P-selectin reduced T lymphocyte accumulation by >90%. In most of the inflammatory sites additional blockade of P-selectin decreased T cell migration from ~20% of control to <10%. Even the migration to Con A-induced inflammation was inhibited from 56% of control to <10% when all three receptors were blocked. Thus, P-selectin appeared to mediate a substantial part of the residual T cell migration that was independent of VLA-4 and E-selectin function.

**Effect of blocking LFA-1 and E- and P-selectin on T cell migration to dermal inflammation**

Both VLA-4 and LFA-1 are important integrins mediating T cell migration to dermal inflammation, and combined blockade of VLA-4 and LFA-1 essentially abolishes T cell migration to dermal inflammation (4). Therefore, the role of LFA-1 and E- and P-selectin in T cell recruitment was examined (Fig. 6). Blockade of LFA-1 inhibited T cell migration induced by all the stimuli by 75–80%. Treatment with anti-P-selectin mAb did not potentiate

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**FIGURE 3.** The effect of anti-VLA-4 Ab treatment on T cell migration to dermal inflammation. Rats were injected i.v. with 111In-labeled T lymphocytes and mAbs to α4 (TA-2), E-selectin (RME-1), a combination of both mAbs, or control mAb as indicated. The migration of T cells to the indicated dermal inflammatory reactions was determined after 20 h and expressed as a percentage of migration in control animals. Each bar represents the mean ± SEM of five to seven animals.

**FIGURE 4.** The effect of anti-VLA-4 and anti-P-selectin mAb on T cell migration to dermal inflammation. Rats were injected with the mAbs to α4, P-selectin, both α4 and P-selectin together, or control mAb as indicated. The migration of T cells to the indicated inflammatory reactions was determined after 20 h and expressed as a percentage of migration in control animals. Each bar represents the mean ± SEM of five to seven animals.

**FIGURE 5.** The effect of anti-VLA-4, anti-E- plus anti-P-selectin mAbs on T cell migration to dermal inflammation. Rats were treated with mAbs to the indicated adhesion molecules or control mAb as indicated. The accumulation of T lymphocytes in dermal sites was determined and values are expressed as a percentage of the response in animals receiving control mAb. Bars are mean ± SEM of five to seven animals per group.
this inhibition, and anti-E-selectin mAb substantially (>10%) inhibited the residual T cell recruitment to only two of the stimuli, LPS and Con A. Furthermore, the combination of E- and P-selectin blockade had a comparable effect to E-selectin blockade alone, and, in response to stimuli such as TNF-α, IFN-γ, TNF-α plus IFN-γ, and poly(I:C), selectin blockade did not enhance the inhibition of T cell migration observed with LFA-1 blockade alone. Thus, it would appear that E- and P-selectins do not mediate alternate pathways to LFA-1 in T cell recruitment to dermal inflammation.

**Effect of blocking E- and P-selectin and VLA-4 on T lymphocyte homing to lymphoid tissues**

The effect of mAbs to E- and P-selectin and VLA-4 on the level of the labeled T lymphocytes in the blood and on their migration to lymphoid tissues was determined in the above animals. As shown in Fig. 7, anti-P- and anti-E-selectin treatment had no effect on the accumulation of labeled T cells in the blood, spleen, cervical, axillary and mesenteric lymph nodes, and Peyer’s patches. However, mAb to VLA-4 either alone or in the presence of E- and P-selectin blockade partially inhibited T cell homing to the mesenteric lymph nodes and abolished migration to Peyer’s patches in accordance with the known dependence of migration to the gut-associated lymphoid tissues on the α4 integrin, α4β7 (6, 29, 30).

**Discussion**

Previous studies showed that E- and P-selectin and the integrin VLA-4 could mediate lymphocyte rolling and that T lymphocytes activated in vitro or in vivo could migrate to cutaneous contact sensitivity reactions using E- and P-selectin (16, 19). This report is the first to examine the relative contributions of E-selectin, P-selectin, and VLA-4 to the migration of T lymphocytes to a variety of inflammatory reactions in the skin. Our findings show that there are marked differences in the use of E- and P-selectin and VLA-4 in T lymphocyte recruitment to these stimuli, even within the same tissue. E- and P-selectin both mediated recruitment to LPS and to the polyclonal lymphocyte activating agent Con A, because blockade of either E- or P-selectin significantly inhibited T cell migration to these stimuli and blocking both selectins reduced T cell accumulation by 60–70%. However, T cell migration to other stimuli, such as IFN-γ and TNF-α plus IFN-γ, was not inhibited by E-selectin blockade alone and was either unaffected or reduced by only 30% when E- and P-selectin were blocked. Interestingly, these results contrast with previous findings using blood monocytes in this model, which showed that monocyte migration to IFN-γ and TNF-α are highly P-selectin dependent (31). Thus, the relative importance of E- and P-selectin to T lymphocyte recruitment in the skin is highly stimulus dependent as well as different from monocytes.

Our results showing a major E- and P-selectin dependence of the migration of T cells to Con A- and LPS-induced inflammation is similar to the findings reported in mice, in which activated T cells and Th1 lymphocyte accumulation in DTH and contact sensitivity reactions was E- and P-selectin dependent (16, 19). The cells in these lesions expressed ligands for P- and E-selectin which appear to be related to the sustained level of α(1,3)fucosyltransferase VII, an enzyme critical to selectin ligand synthesis in activated Th1 cells (32).

Our studies highlight several important features of T lymphocyte recruitment that extend these previous reports. In contrast to the results with activated T cells, in which almost all of the migration to the dermal inflammation was E- and P-selectin dependent (16, 19), a significant component of the T cell migration in these studies was E- and P-selectin independent to all the inflammatory stimuli examined, including Con A. As discussed below, this appears to be the result of lymphocyte migration mediated by VLA-4. This may be a reflection of the fact that the selectin ligands, although present on a few unactivated T cells (2.3–4.8% in
the spleen T cells used here; see Results), are induced or up-regulated following T cell activation by Ag or mitogen, while virtually all resting T cells express VLA-4. Furthermore, VLA-4-mediated rolling does not require Ag activation of the lymphocyte (6, 10). Overall, only 2.5% of the spleen T cells expressed ligands for E-selectin and 5.2% for P-selectin, and <10% of these cells expressed the CD25 activation marker, suggesting that most of the selectin ligand-positive cells were not lymphoblasts. Whether it is only these initial selectin ligand-positive cells which use E- and P-selectin for migration in our model or whether some T cells acquire selectin ligands during the migration period is not yet clear. However, our results demonstrate that, under the resting conditions used, it is clear that T cells can use E- and P-selectin-independent mechanisms for migration as shown especially by the recruitment to IFN-γ, TNF-α, TNF-α plus IFN-γ, and the IFN inducer poly(I:C) (Fig. 2).

Our results also demonstrate a major role for VLA-4 in T cell migration to dermal inflammation to most of the inflammatory sites examined, and a particular cooperative interaction between VLA-4 and E-selectin, rather than P-selectin, in T cell recruitment. Blocking VLA-4 inhibited 40–60% of the lymphocyte recruitment to TNF-α, IFN-γ, TNF-α plus IFN-γ, LPS, and poly(I:C). E-selectin blockade markedly potentiated the inhibitory effect of blocking VLA-4, even when E-selectin blockade alone had no effect, as with, e.g., IFN-γ, TNF-α plus IFN-γ, and poly(I:C) (Fig. 3). The combination of E-selectin and VLA-4 blocked 80–85% of the T cell migration to those lesions (Fig. 5). In contrast, anti-P-selectin treatment had only a small or no potentiating effect on the inhibition of lymphocyte recruitment to most of these sites (Fig. 4). Furthermore, blocking P-selectin, in addition to VLA-4 and E-selectin, reduced T cell migration by only an additional 7–10% to these stimuli, suggesting that P-selectin contributes a relatively much smaller component to the lymphocyte migration to these inflammatory sites than does E-selectin. However, the balance of these mechanisms appears to be influenced by the complexity of the reaction as the response to the potent mitogen and cytokine inducer Con A shows. T cell recruitment to this stimulus was dependent on both E- and P-selectin and VLA-4 (Figs. 2 and 5).

Although previous reports by our group and others have shown that VLA-4 mediated T cell recruitment to dermal inflammation both alone and together with LFA-1, a role for VLA-4 as a mediator of E- and P-selectin-independent migration of resting T cells to cutaneous inflammation has not been previously demonstrated. Our findings extend to skin inflammation the observation that in CNS inflammation T cells use VLA-4, rather than E- or P-selectin, to migrate out of the blood (33–35). They also suggest that in tissues other than the CNS, VLA-4 may substantially substitute for selectins in mediating lymphocyte recruitment to some inflammatory stimuli, as seen in pulmonary inflammation with T lymphoblasts (28).

Despite the important role of VLA-4 in the T cell migration to the skin, endothelial selectins contributed significantly to T cell recruitment in every inflammatory lesion examined. Even in the case of IFN-γ, which was not inhibited by blocking E- and P-selectin, a selectin component was revealed when VLA-4 was blocked (Fig. 4). The balance of VLA-4 vs selectin-mediated mechanisms appears to vary considerably and is quite stimulus dependent, even within a given tissue. This could be related to differences in the extent of E- and P-selectin or VCAM-1 up-regulation induced by these stimuli. However, it is also compatible with multiple mechanisms being available to a migrating T cell and with selective recruitment of T cell subpopulations, which preferentially use different adhesion receptors. Further studies are needed to define the relative contribution of these variables.

Our studies also demonstrate that E-selectin, in particular, functions in concert with VLA-4 in T cell recruitment to most of the reactions studied. These observations agree with an important role for E-selectin in T cell migration to skin lesions, as demonstrated for DTH reactions in the pig and macaque monkey (36, 37), and with results in human skin biopsies showing that most infiltrating T cells express CLA, a carbohydrate ligand for E-selectin (14, 15). Our results also show differences in the role of E-selectin in lymphocyte recruitment to various stimuli, and the role of VLA-4 in the E-selectin-independent migration. The basis for the relatively greater importance of E-selectin than of P-selectin in most of the inflammatory sites is not clear. The rodent homolog of CLA has not been identified, but recent studies indicate that in mouse contact sensitivity reactions P-selectin glycoprotein ligand-1 on T cells is a ligand, although not the sole ligand, for E-selectin-mediated T cell recruitment (38). Our studies emphasize a greater contribution of E-selectin to T cell recruitment to skin than P-selectin.

Our findings in this study once again contrast the mechanisms of T cell migration with those of monocyte migration, the latter being P-selectin, but not E-selectin, dependent, and in particular mediated by VLA-4 and P-selectin (31). The predominant role of the VLA-4 and E-selectin mechanisms of T cell migration shown in this study is also in contrast with the requirement for P-selectin in the recruitment of leukocytes to acute inflammation in P-selectin knockout mice, with relatively normal acute inflammatory responses in E-selectin-deficient mice (39, 40).

Our findings also emphasize the marked difference between the role of VLA-4 and LFA-1 function in vivo in dermal inflammation. E- and P-selectin blockade did not potentiate the inhibitory effect of LFA-1 in most lesions (Fig. 6), in contrast to the enhanced inhibition in the presence of anti-VLA-4 treatment (Fig. 5). This suggests that LFA-1 functions at a different step in the adhesion cascade in vivo than does VLA-4 or the selectins. E- and P-selectin are primarily involved in mediating leukocyte capture, tethering and rolling in the postcapillary venules, a prerequisite step to firm adhesion and transendothelial migration of leukocytes. VLA-4 has also been shown to mediate lymphocyte capture and rolling in vitro, in addition to mediating firm adhesion to the endothelium (1, 10, 41). LFA-1, in contrast, mediates primarily firm adhesion and transendothelial migration (1). Our results suggest that VLA-4, but not LFA-1, is an important mechanism which can substitute for E- and P-selectin-mediated capture and rolling in T cell migration to many inflammatory stimuli in the skin.

In summary, these studies demonstrate that resting T cells can use E- and P-selectin for rapid (<20 h) migration to a range of inflammatory stimuli. Furthermore, VLA-4 is the predominant E- and P-selectin-independent mechanism for migration to skin, and VLA-4 and E-selectin in particular are complementary mechanisms operative in resting T cell migration to inflamed skin.

References


