Expression of Adhesion Molecules in Leprosy Lesions

LAURA SULLIVAN,1 SHIGETOSHI SANO,2 CLAUDE PIRMEZ,3 PADMINI SALGAME,2
CHRISTOPH MUELLER,4 FLORENCE HOFMAN,5 KOICHI UYEMURA,6
THOMAS H. REA,1 BARRY R. BLOOM,2 AND ROBERT L. MODLIN*6

Section of Dermatology1 and Department of Pathology,5 University of Southern California School of Medicine, Los Angeles, California 90033; Department of Microbiology and Immunology, Albert Einstein College of Medicine, Bronx, New York 104612; Evandro Chagas Hospital, Fundação Oswaldo Cruz, Rio de Janeiro, Brazil2; Department of Pathology, Universität Bern, Bern, Switzerland4; and Division of Dermatology and Department of Microbiology and Immunology, UCLA School of Medicine, Los Angeles, California 900336

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Leprosy presents as a clinical spectrum that is precisely paralleled by a spectrum of immunological reactivity. The disease provides a useful and accessible model, in this case in the skin, in which to study the dynamics of cellular immune responses to an infectious pathogen, including the role of adhesion molecules in those responses. In lesions characterized by strong delayed-type hypersensitivity against Mycobacterium leprae (tuberculoid, reversal reaction, and Mitsuda reaction), the overlying epidermis exhibited pronounced keratinocyte intracellular adhesion molecule 1 (ICAM-1) expression and contained lymphocytes expressing the ICAM-1 ligand, LFA-1. Conversely, in lesions in which delayed-type hypersensitivity was lacking (lepromatous), keratinocyte ICAM-1 expression was low and LFA-1* lymphocytes were rare. Expression of these adhesion molecules on the cells within the dermal granulomas was equivalent throughout the spectrum of leprosy. The percentage of lymphocytes in these granulomas containing mRNA coding for gamma interferon and tumor necrosis factor alpha, synergistic regulators of ICAM-1 expression, paralleled epidermal ICAM-1 expression. In lesions of erythema nodosum leprosum, a reactionary state of lepromatous leprosy thought to be due to immune complex deposition, keratinocyte ICAM-1 expression and gamma interferon mRNA* cells were both prominent. Antibodies to LFA-1 and ICAM-1 blocked the response of both αβ and γδ T-cell clones in vitro to mycobacteria. Overall, the expression of adhesion molecules by immunocompetent epidermal cells, as well as the cytokines which regulate such expression, correlates with the outcome of the host response to infection.

The disease spectrum of leprosy, which correlates with the degree of cell-mediated immunity, provides an opportunity to access and analyze immune responses to a single foreign pathogen, Mycobacterium leprae. At one pole of the spectrum, patients with tuberculoid leprosy have one or several skin lesions in which bacilli can only rarely be identified; at the opposite pole, patients with lepromatous leprosy have diffuse infiltration of skin and nerves with bacilli-laden macrophages (28, 29). The immunologic state of these patients is generally assessed by using the lepromin skin test (Mitsuda reaction), which is a granulomatous response that develops 3 to 4 weeks after intradermal injection of M. leprae. The test is positive in tuberculoid patients and negative in lepromatous individuals.

Imposed upon this spectrum are the so-called reactional states, i.e., reversal reaction and erythema nodosum leprosum (ENL), two conditions of particular interest in understanding mechanisms of immunoregulation in humans. The reversal reaction appears to be a naturally occurring delayed-type hypersensitivity response to M. leprae. Clinically, it is characterized by upgrading of the clinical picture toward the tuberculoid pole, including reduction of the bacillary load. Immunologically, a strong skin test reactivity to M. leprae antigens develops, as does lymphocyte responsiveness to M. leprae in vitro (3, 6, 12, 25, 39). The pathogenesis of ENL reactions is enigmatic, thought to be due to immune complex deposition in the lesions (5, 9, 40) and/or increases in cell-mediated immunity (23, 37).

The immune interactions of T cells with antigen-presenting cells are known to be dependent on the expression and interaction of cell surface molecules called adhesion molecules (4, 14, 18, 36). For example, it is known that lymphocytes express LFA-1, which interacts with intracellular adhesion molecule 1 (ICAM-1) on accessory cells to promote adhesion and signaling. Similarly, the CD2 molecule of T lymphocytes is the ligand for the LFA-3 molecule of antigen-presenting cells. The present study was undertaken to determine the expression of adhesion molecules in leprosy lesions across the spectrum and to try to correlate that expression with resistance.

MATERIALS AND METHODS

Patients. Patients with leprosy were classified by using the clinico morphological criteria of Ridley and Jopling (26, 28, 29). Clinical criteria for the diagnosis of reversal reactions included a change in lesions from torpid to erythematous and tumid; the abrupt onset of new erythematous, tumid lesions; and the sudden onset of neuritis. Histological changes were not uniform but included edema in the granulomas, increased numbers of lymphocytes, the presence of giant cells, and desmoplasia of the connective tissue. The diagnosis of ENL was made by using clinical and histopathological criteria. Patients presented with the distinctive picture of crops of painful and tender erythematous nodules in association with fever, malaise, and arthralgias. Histopathological

* Corresponding author.
examination of lesions revealed the presence of neutrophils and lymphocytes superimposed over a lepromatous infiltrate. In addition, lepromin skin tests (3-week Mitsuda reactions) from tuberculoid patients were studied (10). All specimens were obtained with informed consent. The patients were distributed among the different diagnostic groups and showed no segregation according to sex, race, or age.

Tissues. Skin biopsy specimens, obtained by the punch or scalpel technique at the time of diagnosis, were embedded in OCT medium (Ames Co., Elkhart, Ind.) and snap-frozen in liquid nitrogen. The tissues were stored at −70°C until sectioning.

Immunohistochemical studies. (i) Monoclonal antibodies. Primary mouse anti-human lymphocyte monoclonal antibodies were used at optimal concentrations as determined by titration on reactive tonsil. Markers included LFA-1 (TS 1/22 at 1:7,000; a gift of T. Springer, Harvard Medical School); CD2 (TS 2.18 at 1:7,000; provided by C. Clayberger and A. Krenski, Stanford University), LFA-3 (TS 2.9 at 1:4,000; provided by C. Clayberger and A. Krenski, Stanford University) and ICAM-1 (RR 1/1 at 1:10,000; a gift of T. Springer).

(ii) Immunoperoxidase staining. All sections were acetone fixed before being incubated with the monoclonal antibodies for 45 min. Immunostaining was performed by using the alkaline phosphatase anti-alkaline phosphatase technique (Dako Corp., Carpinteria, Calif.). Slides were sequentially incubated with rabbit anti-mouse antibody for 30 min and with alkaline phosphatase anti-alkaline phosphatase immune complex for 30 min and then developed with Vector Red substrate for 30 min (Vector Laboratories, Burlingame, Calif.). Slides were washed in Tris buffer between incubations. Endogenous alkaline phosphatase was blocked with 125 mM levamisole (Vector). Slides were counterstained with hematoxylin and then mounted in glycerine-gelatin. Some slides were stained by using the ABC Elite system (Vector Laboratories). In controls, the primary monoclonal antibody was omitted and isotype control antibodies of irrelevant specificity were used.

(iii) Quantiﬁcation of stained cells. The LFA-1- and CD2-bearing lymphocytes in the epidermis were enumerated and expressed as the number of positive cells per square millimeter of epidermis. For ICAM-1 and LFA-3, the percentage of the epidermis staining positive was estimated by two independent observers; their readings invariably agreed within 10%.

In situ hybridization. The in situ hybridization technique described below was performed as previously described (8, 22).

(i) Tissue preparation. Cryostat sections (3 to 5 μm) were placed on poly-l-lysine hydrobromide-coated slides (Sigma, St. Louis, Mo.) and air dried for 2 h. Dried sections were fixed for 20 min in 4% paraformaldehyde–phosphate-buffered saline (PBS) (pH 7.4), rinsed through several changes of PBS (Flow Laboratories), and dehydrated through graded alcohols. Sections were stored desiccated at 4°C until used.

(ii) Probe preparation. Sections were probed with antisense tumor necrosis factor alpha (TNFα). The data for gamma interferon (IFN-γ) are derived from a previous study (8). TNFα cDNA, a 600-bp fragment, was cloned into pGEM-1 (Promega Corp., Madison, Wis.) to allow for the generation of both sense and antisense 35S-labeled RNA probes. Template DNA was prepared by linearizing plasmid DNA with restriction enzymes that cut once at the distal end of the cloned insert; this restricted in vitro RNA synthesis to probe-specific sequences. The insert was cloned in the 5′-to-3′ direction so that RNA transcripts generated in vitro by transcription from the 5′ SP6 promoter generated sense-strand RNA while the 3′ T7-directed synthesis produced antisense RNA. 35S-labeled transcripts were reduced to an average of 200 bp by alkaline hydrolysis (1) and then ethanol precipitated after adding 7.5 μg of yeast tRNA per 106 cpm. The probe was subsequently resuspended at 1 × 106 to 2 × 106 in 10 mM Tris with 1 mM EDTA (pH 8) and stored at −70°C until hybridization to tissue sections.

(iii) Hybridization. Sections were digested in 1 μg of proteinase K per ml in 100 mM Tris-50 mM EDTA (pH 8) at 37°C for 30 min and then refixed for 20 min in 4% paraformaldehyde-PBS to stabilize cellular mRNA within the proteolyzed matrix. Free amino groups on tissue sections were acetylated by treatment with 0.25% acetic anhydride in 0.1 M triethanolamine (pH 8) for 10 min; the tissues were rinsed briefly in water and then air dried while awaiting hybridization. For hybridization, probe (denatured at 80°C for 3 min) was mixed with formamide (final concentration, 50%), NaCl (300 mM), Tris-HCl (pH 7.5; 20 mM), EDTA (5 mM), Denhardt’s solution (1 x), dextran sulfate (10%), and dithiothreitol (100 mM) at a final activity of 106 cpm per μl of hybridization mix. Then 10 μl of this hybridization solution containing 106 cpm of probe was added to each slide, and the tissue was covered with a siliconized coverslip and sealed with rubber cement. Slides were hybridized for 17 to 20 h at 47°C. Unhybridized probe was washed from the slides in a solution of 50% formamide, 2 × SS (1 × SSC is 0.15 M NaCl plus 0.015 M sodium citrate), and 5 mM EDTA in four changes for a total of 2 h at 56°C. After the second wash, the slides were digested with RNase A (20 μg/ml) and RNase T1 (1 U/ml) for 30 min at 37°C. After the last wash, slides were dehydrated through graded alcohols containing 400 mM ammonium acetate and air dried. For autoradiography, slides were dipped into Kodak NTB-2 emulsion, diluted 1:2 with 400 mM ammonium acetate, and exposed at 4°C for 10 to 28 days. The slides were then developed by using Dektol developer (1:2 in water) for 2 min and fixer for 5 min (photography supplies from Eastman Kodak, Rochester, N.Y.) and counterstained with hematoxylin and eosin. Each tissue was hybridized with sense-strand probes (homologous to cellular mRNA) as well as antisense strand probes (complementary to cellular mRNA); known positive and negative control tissues were included in each run.

(iv) Evaluation of slides. Cells were considered to be positive for the expression of cytokine mRNA when they contained more than twice the number of grains of the most positive cell on the control (sense) slide for the same tissue. Positive cells were enumerated over the entire section (8). The number of grains per average positive cell was constant for the TNFα slides but varied according to patient classification for the IFN-γ slides. Therefore, for IFN-γ, the number of grains per square millimeter of granuloma was determined by multiplying the number of positive cells per square millimeter by the grain count for average positive cell, thus giving an approximation for the total mRNA level in the tissue. For TNFα, the data are expressed as the percent positive cells in the section.

T-cell clones. (i) T-lymphocyte clones. Clones were derived from peripheral blood mononuclear cells of normal donors by the method of limiting dilution (16). T cells were seeded into round-bottom microtiter wells at a concentration of 0.3 cell per well. These cells were cultured and expanded in the presence of 10% AB serum, 20 U of recombinant interleukin-2 per ml (Genzyme, Boston, Mass.), 100 irradiated feeders, and antigen. Clones were expanded with antigen...
and feeder cells biweekly. Clones were phenotyped by using anti-TCR81 (kindly provided by M. Brenner), which identifies all cells using the TCR8 chain, and BMA 031 (kindly provided by R. Kurrle, Marburg, Germany), which identifies αβ T cells.

(ii) Measurement of antigen-induced proliferation of lines and clones. T-cell lines or clones, washed free of interleukin-2, were cultured in the presence of irradiated feeders as antigen-presenting cells. For αβ T-cell clones, triplicate 200-μl flat-bottom wells contained cells at a density of 10^4 cloned cells per 10^5 autologous feeders in the presence of M. leprae sonic extract. For the γδ T-cell clone, 3 x 10^4 cloned cells were plated in triplicate with 4 x 10^4 allogeneic feeders in the presence of M. tuberculosis H37Ra (Difco). Control cultures received interleukin-2 alone to demonstrate the viability of clones or lines, no antigen to detect background proliferation, or an irrelevant antigen such as tetanus toxoid. For αβ T-cell clones, cultures were pulsed with [3H]thymidine at 3 days and harvested 4 h later. For the γδ T-cell clone, cultures were pulsed at 2 days and harvested 6 h later. Blocking was attempted by adding anti-LFA-1 (1:500) and anti-ICAM-1 (1:1,000) antibodies at the beginning of the experiment. These concentrations were determined by titration on antigen reactivity of peripheral blood mononuclear cells using the lowest dilution to achieve maximal inhibition of proliferation. In all experiments, a mouse immunoglobulin G control at comparable concentrations was used, which did not inhibit proliferation.

RESULTS

Immunolabeling studies. To ascertain the distribution of adhesion molecules in leprosy lesions throughout the spectrum of leprosy, we performed immunolabeling on skin biopsy specimens by using monoclonal antibodies against ICAM-1, LFA-1, CD2, and LFA-3. The adhesion molecules were all strongly expressed on cells within dermal granulomas in all forms of leprosy (data not shown). However, striking differences were noted in the expression of these adhesion molecules on cells within the epidermis overlying these granulomas.

In Mitsuda reactions (n = 10), which serve as a standard measure of cutaneous hypersensitivity to challenge with M. leprae, more than 80% of keratinocytes expressed ICAM-1 (Fig. 1). Similarly, in immunologically active forms of leprosy characterized by strong delayed-type hypersensitivity against M. leprae, specifically tuberculoid lesions (n = 9), and reversal reactions (n = 5), more than 60% of keratinocytes were ICAM-1 positive. In contrast, in the epidermis overlying lepromatous lesions (n = 6), fewer than 10% of keratinocytes expressed ICAM-1. However, in lepromatous patients with ENL (n = 7), strong keratinocyte ICAM-1

FIG. 1. ICAM-1 expression by epidermal keratinocytes in leprosy. (A) The percentages of epidermal keratinocytes (mean ± standard error) expressing ICAM-1 are shown throughout the spectrum of leprosy. Keratinocyte cell surface staining by ICAM-1 is pronounced in reversal reaction (B) and ENL (C) epidermis. ICAM-1 staining is absent in lepromatous epidermis, but positive in dermal granulomas (D). Immunoperoxidase is counterstained with hematoxylin (40× objective). Abbreviations: LT, lepromin test; RR, reversal reaction; T, tuberculoid; L, lepromatous.
staining was visualized. ICAM-1 is not expressed on keratinocytes of normal skin (13). The distribution of LFA-1 immunolabeling was similar to that of its ligand ICAM-1 (Fig. 2). LFA-1-positive lymphocytes were most numerous in the epidermis of Mitsuda reactions, tuberculoid lesions, reversal reactions, and ENL lesions, but rare in the epidermis overlying nonreactional lepromatous lesions. These data indicate a correlation in DTH against M. leprae antigens between expression of the adhesion molecules ICAM-1 and LFA-1 in the epidermis overlying lesions. The data provide further evidence that ENL, in light of strong epidermal adhesion molecule expression, represents a state of increased cell-mediated immunity against M. leprae.

LFA-3, expressed by normal epidermis, was expressed by keratinocytes in all forms of leprosy (data not shown). CD2, the ligand for LFA-3, was found on a similar number of epidermal lymphocytes as LFA-1 (data not shown).

In situ hybridization. The induction of ICAM-1 expression is regulated chiefly by IFN-γ in synergy with TNFα. We hypothesized that ICAM-1 expression within the epidermis was chiefly regulated by the large numbers of immune T cells present within the dermal granulomas. To ascertain the presence of these cytokines in lesions, we performed in situ hybridization to detect cellular mRNA by using IFN-γ and TNFα RNA probes.

(i) IFN-γ gene expression in lesions. Previous analysis of IFN-γ mRNA revealed that the intensity of positive cells varied according to patient classification (8). Because of the possible importance of lymphokines on expression of adhesions in lesions, we summarize these published data in a single figure in terms of the number of grains in positive cells per square millimeter of epidermis (mean ± standard error). (B) LFA-1+ lymphocytes in the epidermis of a reversal reaction lesion (immunoperoxidase counterstained with hematoxylin; 40× objective). Abbreviations are as in Fig. 1.

FIG. 2. LFA-1-positive lymphocytes in the epidermis of leprosy lesions. (A) The number of epidermal lymphocytes which stained positive for LFA-1 per square millimeter of epidermis (mean ± standard error). (B) LFA-1+ lymphocytes in the epidermis of a reversal reaction lesion (immunoperoxidase counterstained with hematoxylin; 40× objective). Abbreviations are as in Fig. 1.

DISCUSSION

Lymphocyte adhesion is a necessary event for the cell-cell interactions required to produce an immune response (4, 14, 18, 36). The present study focused on detection of adhesion molecules in the epidermis overlying leprosy lesions, as well as on the immune events within dermal granulomas which influence adhesion molecule expression. The results for the different forms of leprosy reactions demonstrated that the expression of adhesion molecules was related to the level of cell-mediated immunity against M. leprae. Furthermore, both ICAM-1 and LFA-1 expression appeared to be necessary for the ability of αβ and γδ T-cell clones to respond to mycobacterial antigens. The principal molecular pathways involved in the preliminary, nonspecific adhesion interactions which facilitate antigen recognition include CD2 and its ligand LFA-3, and LFA-1 and its ligand ICAM-1. Indeed, antigen-independent cell-cell adhesion can be inhibited with monoclonal antibodies directed against either LFA-1 or CD2 present on T cells or against LFA-3 and sometimes ICAM-1 on target cells (14, 17, 31, 36). After preliminary contact, antigen-specific recognition in association with the major histocompatibility complex occurs via the T-cell receptor–CD3 complex. Our...
data indicate that the ability of both αβ and γδ T-cell receptor types to proliferate to mycobacterial antigens is dependent on adhesion resulting from the interaction of the LFA-1 and ICAM-1 molecules.

The adhesion molecules studied were expressed in the dermal granulomas of all lesions studied, the site of predominant disease activity at the time of biopsy. However, we found marked differences in the expression of adhesion molecules on cells within the epidermis across the spectrum of disease. Keratinocyte expression of ICAM-1 paralleled LFA-1 expressed by epidermal lymphocytes, pronounced in those types of lesions that are characterized by strong DTH

**TABLE 1. Antibodies to LFA-1 and ICAM-1 block T-cell clone reactivity to mycobacterial antigens**

<table>
<thead>
<tr>
<th>Clone</th>
<th>Expt no.</th>
<th>Media</th>
<th>Antigen</th>
<th>Antigen + anti-ICAM-1</th>
<th>Antigen + anti-LFA-1</th>
</tr>
</thead>
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<tr>
<td>12G12 (γδ)</td>
<td>1</td>
<td>1,084</td>
<td>7,440</td>
<td>88</td>
<td>3,755</td>
</tr>
<tr>
<td></td>
<td>2</td>
<td>227</td>
<td>5,782</td>
<td>NT*</td>
<td>1,395</td>
</tr>
<tr>
<td></td>
<td>3</td>
<td>645</td>
<td>5,757</td>
<td>60</td>
<td>NT</td>
</tr>
<tr>
<td>59.3 (αβ)</td>
<td>1</td>
<td>75</td>
<td>4,440</td>
<td>2,784</td>
<td>1,213</td>
</tr>
<tr>
<td>59.5 (αβ)</td>
<td>1</td>
<td>48</td>
<td>9,327</td>
<td>5,852</td>
<td>5,846</td>
</tr>
</tbody>
</table>

* The data are shown in counts per minute of [3H]thymidine incorporation for T-cell clones reactive to mycobacteria. A mouse immunoglobulin G control had no effect on the clones proliferative response.

* NT, not tested.

against *M. leprae*: lepromin skin tests, reversal reactions, and tuberculoid lesions. In the epidermis overlying lepromatous lesions, which are lesions from patients specifically unresponsive to *M. leprae* antigens, few keratinocytes expressed ICAM-1 and epidermal lymphocytes expressing LFA-1 were rarely detected. The number of epidermal lymphocytes expressing CD2 paralleled the numbers expressing LFA-1. The CD2 ligand, LFA-3, was normally expressed by all keratinocytes in lesions. However, the expression of major histocompatibility complex class II antigens by keratinocytes, as well as the number of epidermal Langerhans cells, has been shown to correlate with the level of delayed-type hypersensitivity against *M. leprae* (24). Therefore the conditions for interaction of epidermal T cells with antigen-presenting cells (Langerhans cells and/or ICAM-1+ HLA-DR+ keratinocytes) are more favorable in lepromin skin tests, reversal reactions, and tuberculoid lesions than in lepromatous lesions.

The expression of ICAM-1 by keratinocytes is known to be regulated by lymphokines, particularly the synergistic action of IFN-γ and TNFα (2, 11, 13). IFN-γ alone can exert an effect on ICAM-1 gene induction that is significant with respect to degree and persistence. We detected both IFN-γ (8) and TNFα mRNAs in leprosy lesions by in situ hybridization, and the relative levels of these mRNAs were correlated with the degree of epidermal ICAM-1 expression. Overall, the lowest percentages of TNFα-positive cells were found in lepromatous leprosy patients, with three- to fivefold-greater numbers in lepromin skin tests, tuberculoid lesions, and reversal reaction specimens. Similarly, the
lowest levels of IFN-γ mRNA were found in lepromatous lesions, with 10- to 1,000-fold greater levels in the more immunologically reactive forms of the disease.

In developing an understanding of the immunological mechanisms which cause leprosy, it is noteworthy that lepromatous patients show a curious, often spontaneously occurring reaction (ENL). ENL is thought to be due to immune-complex deposition within tissues containing bacilli and antigens of lepromatous patients. There is some evidence that changes in cell-mediated immune responses could facilitate antibody production in this reactive state. For example, there is a decrease in suppressor T-cell activity (20) and an influx of CD4+ memory T cells into ENL lesions (8, 19). The present data provide further evidence for immune activation in ENL lesions: (i) keratinocyte ICAM-1 expression and LFA-1+ epidermal lymphocytes were prominent; and (ii) IFN-γ mRNA expression was of a magnitude similar to that in tuberculosis lesions and lepromin skin tests and greater than in nonreactional lepromatous biopsies. The percentage of TNFα-positive cells was low, as in nonreactional lepromatous lesions, so that either the level of IFN-γ alone or the combined levels of TNFα and IFN-γ are likely to be sufficient to induce keratinocyte ICAM-1 expression.

The epidermis may have a key immunological role in leprosy. Not only are the earliest skin lesions of tuberculoid leprosy thought to arise within the epidermis (27), but also the presence of lymphocytic infiltration in the epidermis is a histological criterion for the classification of leprosy at or near to the tuberculoid pole (29). Furthermore, the epidermis overlying the granulomatous infiltrate appears to reflect the immunological characteristics of the dermal infiltrate (24). The epidermis contains keratinocytes and Langerhans cells, unique immunocompetent cells which may influence the outcome of an immune response by releasing cytokines and/or by presenting antigen to T cells (15, 30, 34). The migration and adhesion of these T cells to the epidermis may be mediated by interaction of ICAM-1 and LFA-1 (11, 32). Furthermore, the ability of keratinocytes to induce T-cell proliferation is dependent on expression of ICAM-1 (33). Both αβ and γδ T cells have been reported to be present in the epidermis of leprosy lesions (38), where they may interact with resident immune cells. The present data suggest that such an immune process may be dependent on the interaction of LFA-1 and ICAM-1. To our knowledge, these are the first data implicating LFA-1 and ICAM-1 in the ability of γδ T cells to proliferate in response to soluble antigen. Anti-LFA-1 and anti-ICAM-1 antibodies also block the cytotoxicity of mycobacterium-specific γδ T-cell clones (21a). Previously, the binding of γδ T cells to thymic epithelial cells has been shown to be dependent on both LFA-1 and ICAM-1 molecules (35). In addition, the cytotoxicity of a γδ T-cell clone recognizing major histocompatibility complex class I was blocked by anti-LFA-1 (7).

The data presented provide an immunological framework within which to develop an understanding of the pathogenesis of leprosy. Tuberculoid lesions, as well as lepromin skin tests, are characterized by both ICAM-1 expression by keratinocytes and LFA-1+ epidermal lymphocytes. This is paralleled by pronounced IFN-γ and TNFα mRNA levels in lesions. In striking contrast, the epidermis of lepromatous lesions is characterized by the absence of these markers of immune activation, although these molecules are strongly expressed in the dermal granulomas (41). The reactive states of leprosy represent an alternation in the immune response to M. lepra in both magnitude and effect. Reversal reactions appear to be hyperimmune responses with markedly increased cytokine production, causing upgrading of the clinical picture, often long term and often with concomitant tissue damage (8). On the other hand, ENL lesions exhibit features of a transient rise in cell-mediated immunity to M. leprae, with increased T-cell responses able to induce ICAM-1 expression by keratinocytes, but insufficient to reduce the bacillary load in lesions. Of interest, TNFα is not expressed in this reaction, suggesting that its expression may be required for long-term killing or growth inhibition of M. leprae. The changes in expression of these adhesion molecules and the dynamics of cytokine expression correlate with the outcome of infection. It is, however, unclear which is the initial event, and the present study does not directly address this question, yet we hypothesize that the initial interaction of T cells with antigen-presenting cells occurs in the absence of cytokine-induced ICAM-1 expression. Later, the release of cytokines with subsequent induction of ICAM-1 serves to amplify nascent cellular immunity. We believe that further characterization of adhesion molecules in leprosy lesions, as well as the cytokines which regulate their expression, will prove to be general and useful in defining mechanisms of resistance to intracellular pathogens.

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