DEPLETION OF EPIDERMAL LANGERHANS CELLS AND Ia IMMUNOGENICITY FROM TAPE-STRIPPED MOUSE SKIN*

By J. WAYNE STREILEIN, LANYA WHITE LONSBERRY, AND PAUL R. BERGSTRESSER

From the Departments of Cell Biology and Internal Medicine, University of Texas Health Science Center at Dallas, Dallas, Texas 75235

The immunogenicity of solid tissue allografts resides among constitutive cells. The importance of this realization derives chiefly from recent information concerning the differential expression of transplantation alloantigens on various tissues. Class I major histocompatibility complex (MHC)[1] alloantigens are expressed by virtually all nucleated cells, whereas class II MHC determinants (Ia alloantigens of mice) have a restricted distribution, limited largely to B lymphocytes, macrophages, subpopulations of T lymphocytes, and specialized dendritic cells (1, 2). Ia antigens have been shown not to be expressed on the parenchymal keratinocytes of normal murine body wall skin, although Langerhans cells within the epidermis do express the gene products of the *I* region of *H-2* (3). We have previously shown (4) that allografts of cornea, an epidermal tissue devoid of Langerhans cells, fail to induce alloimmunity when donor and host differ only at the *I* region of *H-2*. We interpreted this result to mean that Langerhans cells might be the only cells in normal body wall skin responsible for the immunogenic expression of Ia alloantigens. However, there are differences between cornea and body wall skin other than differential content of Langerhans cells. For instance, the normal cornea also lacks vascular endothelial cells, a cell type that has been reported to express Ia antigens (5, 6).

In an effort to resolve this issue, we attempted to remove Langerhans cells from conventional body wall epidermis to test the ability of that skin to express Ia antigens. Ultraviolet light (UVL) irradiation of murine skin results in a transient loss of ATPase-positive, Ia-positive cells from the epidermis (7). However, we found that skin grafts prepared from UVL-treated skin are no less immunogenic for Ia antigens than grafts from unirradiated skin (8). It has recently been reported (9) that significant numbers of Langerhans cells, as detected by electron microscopy, may remain within the epidermis after UVL irradiation (9). Therefore, it is not surprising that UVL-treated grafts remain immunogenic for Ia antigens. Another method that has been reported to divest skin of Langerhans cells is repeated stripping with cellophane tape. Lessard, Wolff, and Winkelmann (10) demonstrated that repeated tape-stripping of guinea pig skin removes the stratum corneum, exposing the suprabasilar layer that contains Langerhans cells. Most of this suprabasal portion of the epidermis is then

* Supported by grants AI-10678, AI-17363, and CA-09082 from the U. S. Public Health Service.

[1] Abbreviations used in this paper: MHC, major histocompatibility complex; PBS, phosphate-buffered saline; UVL, ultraviolet light.

eliminated as a compact parakeratotic layer, carrying resident Langerhans cells with it. They reported that during the next few days the underlying epidermis was devoid of identifiable Langerhans cells. We used tape-stripping of murine body wall skin in an effort to remove resident Langerhans cells and to test the Ia antigenicity of skin grafts prepared therefrom. The results of these experiments form the basis of this report.

Materials and Methods

Mice. Animals used in these experiments were obtained from our domestic breeding facility and used between the ages of 3 and 5 mo. Ia-disparate combinations included A.TH and A.TL, C57BL/6 and H-2 bm12, and B10.AQR and B10.T (6R). *H-2 K*-disparate combinations included B10.A and B10.AQR, A.TL and A.AL, and C57BL/6 and H-2 bml. For specific haplotype designations, see Klein et al. (11).

Identification of Epidermal Langerhans Cells. Epidermal Langerhans cells were identified in *"en face"* whole mounts of epidermis by immunofluorescence microscopy and by cell surface ATPase activity, as described previously (12). To prepare specimens, each skin graft was excised from recipient animals, or normal body wall split-thickness skin samples were removed with a keratotome (12). Specimens were then cemented by the epidermal surface to glass cover slips, and the underlying dermis separated and discarded after a 4-h incubation in EDTA at 37°C (12). Tissues were fixed at 23°C in several changes of acetone over 30 min, a procedure that also removed the cyanoacrylic cement. They were then washed in three changes of phosphate-buffered saline (PBS) at 4°C. Commercial murine monoclonal anti-Ia^k with biotin conjugate (specificity 2; Becton, Dickinson & Co., Sunnyvale, CA) was diluted 1:20 in PBS, and aliquots of 200 μl were placed in small closed vials into which each specimen was placed for a 16-h incubation at 4°C. Specimens were then washed three times in PBS over 90 min, incubated for 120 min in fluorescein conjugated avidin (Becton, Dickinson & Co.), diluted 1:20 in PBS, and washed again three times in PBS. These final three procedures were performed at 23°C on a gently moving rocker table. Individual slides were prepared by mounting specimens, basal cell layer up, in 90% glycerol/10% PBS. These *en face* mounts of epidermis were examined with an Orthoplan fluorescence microscope equipped for epi-illumination (E. Leitz, Inc., Rockleigh, NJ). Background staining of epidermal nuclei, resulting from direct binding of avidin, was seen frequently, although the Ia-positive dendritic Langerhans cells were easily identified.

Tape-Stripping of Skin. Mice were anesthetized with chloral hydrate, restrained in a supine position, and their abdominal skins dry shaven with a razor blade. The shaved abdomen was then stripped by repeated application (15 times) of cellaphone tape (Scotch-brand magic transparent tape, 810; 3M Co., St Paul, MN). This number of tape applications was sufficient to cause the epidermal surface to glisten. Additional applications did not improve the removal of additional stratum corneum.

Skin Grafting. Full thickness skin grafts were prepared and grafted orthopically according to the method previously described (13). Plaster casts were removed 8 d later, and the grafts were observed daily for evidence of rejection. Rejection was judged to be complete when all evidence of epidermis was gone. Median survival times were calculated according to the method of Litchfield (14).

Results

Effect of Tape-Stripping on Cutaneous Langerhans Cells. Abdominal skin of adult B10.A, A/J, and C57BL/6 mice were dry shaved, then tape-stripped to glistening with cellophane tape. Immediately thereafter, as well as 24 h later, skin biopsies were taken and analyzed for the presence of epidermal Langerhans cells, using two techniques: cell surface ATPase, and, where possible, cell-surface expression of Ia^k alloantigens, using fluorescent labeled antisera. Each assay was performed on sheets of epidermis, prepared as described previously. The results of this study are presented in Table I.

TABLE I

Effect of Tape-Stripping on Epidermal Langerhans Cells in Murine Abdominal Wall Skin

Assay used to detect Langerhans cells	Appearance of Langerhans cells* (hours after tape-stripping)			
	0	N‡	24	N
ATPase				
Interfollicular	−	(2)	−	(2)
Follicular	±	(2)	−	(2)
Anti-Ia				
Interfollicular	−	(7)	++	(5)
Follicular	±	(7)	++	(5)

−, No Langerhans cells seen.

±. Focal areas of intact epidermis containing Langerhans cells.

++, Langerhans cells present with altered morphology (see text) at a surface density ranging from 100 to 400/mm^2.

* Abdominal wall skin from two or more mice examined at 0 and 24 h.

‡ Number of specimens examined.

Immediately after tape-stripping, ATPase-positive Langerhans cells disappeared from most portions of interfollicular epidermis, although rare, small islands of ATPase-positive dendritic cells could be seen adjacent to hair follicles. When tape-stripped skin was examined with this assay 24 h later, ATPase-positive cells were absent from well-stripped portions of the epidermis and from hair follicles as well. When similar preparations were studied by immunofluorescence with anti-Ia^k, it was found that very rare Ia-positive cells remained within the interfollicular epidermis immediately after cellophane tape-stripping. Chiefly, these cells were found near the surface openings of hair follicles, although small fields of apparently intact Langerhans cells were occasionally identified. Because these latter areas appeared to be covered with morphologically intact epidermal cells, we presume that they had been incompletely stripped. When stripped skin was assayed 24 h later, Ia-positive cells had reappeared within the epidermis, although few displayed typical dendritic configurations of normal Langerhans cells. It would appear that tape-stripping is an effective means of removing the vast majority of resident Langerhans cells from cutaneous surfaces. However, whether all Langerhans cells were completely removed could not be determined from this experiment. Although some Langerhans cells are though to be derived from bone marrow stem cells (15, 16), it has been reported (17) that epidermal Langerhans cells are also capable of self renewal within the epidermis. The following experiments were designed to determine whether Ia-positive cells that are identified within epidermis of skin stripped 24 h previously are of resident origin or come from the systemic circulation.

Identification of Source of Langerhans Cells in Tape-Stripped Skin Grafts. Abdominal wall skin of donor mice was tape-stripped as before and used to prepare full thickness skin grafts that were placed orthotopically on appropriate recipient mice. At periodic intervals thereafter, selected grafts were excised to assay for the appearance of Ia-positive (Langerhans) cells within graft epidermis. The results are summarized in Table II. In the first experiment, tape-stripped B10.A grafts were placed on syngeneic recipients. Immediately thereafter and throughout the next 3 d, the graft epidermis

TABLE II

Effect of Tape-Stripping on Langerhans Cells in Skin Grafts Placed on Syngeneic and Allogeneic Mice

Donor of tape-stripped Skin	Recipient	Appearance of Ia-positive Langerhans cells (days after grafting)*							
		0	1	2	3	5	7	10	14
B10.A	B10.A	−	−	−	−	+‡	+	++	+++
B10.A	C57BL/6	±	−	−	NT	−	−	−	(−)§
C57BL/6	B10.A	−	NT‖	−	NT	+‡	+	NT	NT

* Grafts from two recipient mice were examined at each time period. Results were similar in each instance. The method of scoring was as follows. −, no Ia positive Langerhans cells seen in epidermis; ±, focal areas of intact epidermis containing Langerhans cells; +‡, Langerhans cells seen at periphery of graft and in scattered focal areas; +, normal Langerhans cells present in reduced numbers throughout the specimen; ++, normal Langerhans cells present in normal numbers; +++, normal Langerhans cells present in increased numbers in portions, but not entire surface of specimen.

§ Graft epidermis destroyed by rejection.

‖ Not tested.

lacked Ia-positive cells. However, Ia-bearing cells appeared focally at 5 d and diffusely at 7 d.

Ia-positive putative Langerhans cells in tape-stripped grafts, examined on days 5 and 7, exhibited great variation in both morphology and distribution. This might be attributed to variations in the rate at which vascular supply returned to graft dermis, resulting in different rates of immigration into the epidermis. All four grafts placed on B10.A recipients and examined on day 5 contained Ia-positive dendritic cells of host origin. These cells usually exhibited dense surface staining, with brightly stained dendritic processes, although some cells appeared round, with no evidence of dendrites. One C57BL/6 graft exhibited a large peripheral area of normally distributed Langerhans cells. In the other three specimens, cells were observed in <10% of graft surfaces, where they were observed to form small clusters of regularly spaced, dendritic cells. By day 7, more normal appearing distributions could be seen. Ia-positve Langerhans cells were found in 20–50% of each graft, predominantly with normal, delicate surface staining. For all eight grafts examined on days 5 and 7, no distinction could be made between syngeneic and allogeneic recipients on the basis of morphology or distribution. On day 14, Langerhans cell numbers and appearance had returned essentially to normal; in some areas of these healed-in grafts, the concentration of Langerhans cells actually appeared to have increased above normal. Thus, tape-stripped skin grafts acquire Ia-positive cells much more slowly than does intact, stripped skin, suggesting that nonspecific injury to grafts delays recovery of resident cells or that repopulation from the systemic circulation accounts for the reappearance of these cells. When skin is grafted by this technique, ~ 48–72 h is required for the graft to establish an effective anastomosis with the systemic blood vasculature. To discriminate between these two possibilities, allografts of tape-stripped skin were used. These results are also presented in Table II.

Tape-stripped B10.A grafts placed on C57BL/6 recipients never reexpressed epidermal cells bearing Ia^k alloantigens. Moreover, tape-stripped skin from C57BL/6 donors that was placed on B10.A recipients acquired Ia^k-positive epidermal cells at 4 d; in fact, the appearance of these cells on day 7 was indistinguishable from that observed in stripped B10.A grafts on B10.A recipients.

We conclude from these experiments that tape-stripping irreversibly removes

Langerhans cells from the epidermis. When Ia-positive cells and/or Langerhans cells reappear in tape-stripped skin, the cells must come from the systemic circulation, rather then arising phoenixlike from the original resident population.

Alloantigenicity of Tape-Stripped Skin Grafts. Because tape-stripping effectively removes almost all Langerhans cells from the epidermis, it became possible to assess the contribution of donor Langerhans cells to the aggregate immunogenicity of skin allografts. In preliminary studies, it was determined that syngeneic tape-stripped skin grafts heal readily. In fact, union between graft and host epidermis is achieved more quickly than with unstripped skin grafts. In conducting these studies, we had two considerations in mind: (*a*) Langerhans cells, alone among epidermal cells, express the products of the *I* region of the *H-2* complex, and (*b*) Langerhans cells apparently possess the unique property of presenting antigens in a highly immunogenic form to immunocompetent lymphocytes.

Tape-stripped skin grafts were prepared from various donor strains and placed on histoincompatible recipients representing either Ia antigenic disparity or *H-2 K* disparity alone. The survival patterns of these grafts are presented in Table III. Control mice received unstripped skin from the same donors. In three of four strain combinations, differing only with respect to *I* region antigens, a minority of animals in each group retained their tape-stripped graft beyond 30 d. In these three groups, the median survival time of tape-stripped skin grafts was prolonged compared to controls. It would thus appear that Langerhans cell-depleted skin grafts express significantly less Ia antigen in immunogenic form than do normal skin grafts. Yet, other constituent cells bearing Ia antigens must still remain within these grafts because they are able to initiate alloimmunity. By contrast, the survival patterns of *H-2 K*-disparate stripped and nonstripped skin grafts were essentially identical, implying that donor epidermal Langerhans cells are of little consequence in the process by which mice become immunized to class I allodisparities of skin allografts. These data do not exclude a role for Langerhans cells in the process of allograft recognition, because host-derived Langerhans cells appear in grafted skin as early as 4 d after grafting.

TABLE III

Survival Patterns of Tape-Stripped Skin Allografts

Donor	Host	Skin	Days after grafting (number of surviving grafts)										Median survival time (95% confidence limits)
			8	9	10	11	12	13	14	15	20	30	
													d
A.T.L I^{a*}	A.TH I^a	Stripped	8	8	8	7	7	6	6	6	4	2	20.0 (14.0–28.6)
		Normal‡	9	8	7	5	3	1	0	0	0	0	11.4 (10.4–12.56)
A.TH I^a	A.TL I^b	Stripped	3	3	2	2	2	1	1	1	1	1	NB§
		Normal‡	4	4	2	2	1	1	0	0	0	0	NB
B10.AQR I^k	6R I^q	Stripped	4	4	3	1	1	1	1	1	1	1	NB
		Normal‡	3	3	2	2	2	2	0	0	0	0	NB
bm12 I^{bm12}	B6 I^b	Stripped	6	6	6	6	6	6	5	3	1	1	16.8 (14.9–18.9)
		Normal‡	7	7	6	6	2	1	1	1	0	0	12.0 (10.7–13.4)
A.AL K^k	A.TL K^a	Stripped	4	4	3	3	3	3	3	2	0	0	NB
		Normal‡	7	6	5	5	3	2	1	1	0	0	—
B6 K^b	Hzl K^{bml}	Stripped	8	8	5	4	4	4	4	3	0	0	12.7 (11.8–13.8)
		Normal‡	9	9	8	7	7	7	7	7	1	0	14.7 (13.6–15.6)

* Phenotype of disparity at *H-2*.

‡ Unstripped skin as control.

§ Numbers too few to calculate reliable median survival time.

Discussion

Langerhans cells are known to possess two attributes that bear on their role in immunologic reactions within the skin. First, they alone among epidermal cells express, on the cell surface, molecular products of the *I* region of the *H-2* complex in the mouse (3, 4). Second, they function as antigen-presenting cells to immunocompetent T lymphocytes, as measured in vitro and in vivo (18, 19). In the context of allograft immunity, the contribution of Langerhans cells to the immunogenicity of skin grafts is a matter of some controversy. We have been searching for an experimental maneuver that could rid body wall skin of all Langerhans cells so that we could test the immunogenicity of grafts prepared therefrom. As previously reported (4), allografts prepared from murine cornea, an epidermal surface devoid of Langerhans cells, fail to immunize against Ia alloantigens, but effectively immunize against *H-2 K/D* antigens. However, we have been unable to erase Ia alloantigenicity from murine skin by exposing donors to ultraviolet irradiation in doses sufficient to reduce identifiable Langerhans cells to <5% of normal levels within epidermis (8). Thus, we turned to cellophane tape-stripping, a technique used originally for this purpose by Lessard et al. (10). Although tape-stripping in our hands has not achieved our ultimate goal of removing all vestiges of Langerhans cells from skin, the technique is sufficiently effective to allow us to draw certain inferences about the alloimmunologic properties of these cells.

Langerhans Cells Make a Significant Contribution to the Ia Antigenicity of Skin Grafts. Tape-stripped skin grafts placed on recipients that differ only at the *I* region of *H-2* survived longer than their unstripped controls, and some grafts survived indefinitely (beyond 30 d). However, because the majority of these grafts did immunize their hosts and were rejected, we must conclude that either (*a*) dermal cells (macrophages, endothelial cells, etc.) that may express Ia antigens are sufficient to induce allograft immunity; and/or (*b*) the very few Langerhans cells that remain after skin stripping, especially those near the stomata of and within hair follicles are sufficient to immunize. Unfortunately, not enough glabrous skin is available on a mouse to examine this latter possibility.

Donor Langerhans Cells Are Not Essential for Effective Presentation of H-2 K/D Antigens in Skin Allografts. In fact, one panel of tape-stripped *H-2 K*-disparate grafts was rejected slightly more promptly than their unstripped controls. This result reaffirms our previous assertion that corneal allografts differing only at *H-2K* are effective immunogens in the absence of donor Langerhans cells. It is altogether possible that host Langerhans cells fulfill this processing role or that Langerhans cells are not needed for alloantigen presented through skin. If Langerhans cells are to be included among passenger cells, as originally identified by Steinmuller (20) and then incorporated by Lafferty (21) and Bach (22) into a hypothesis concerning allogeneic interactions, then their role in skin is not analogous to the role passenger cells play in thyroid grafts. It has been reported and confirmed (23, 24) that thyroid grafts, rendered free of syngeneic passenger cells by a variety of maneuvers, are unable to immunize to their constituent alloantigens. Perhaps skin grafts may be so immunogenic as to overshadow the passenger cell effects seen with other types of tissue allografts.

An unexpected bonus from these experiments was new insight into the kinetics of Langerhans cell turnover in skin. It should be pointed out that the few Langerhans cells that remain after tape-stripping are associated with hair follicles and do not

appear to migrate out into the epidermis proper, at least during the 14 d in which grafts of stripped skin were observed. Thus, we conclude that cellophane stripping removes essentially all intraepidermal Langerhans cells. In stripped but anatomically intact skin, Langerhans cells reappear (as detected by surface Ia antigens) within 24 h, but in grafted, stripped skin, they do not reappear until several days later. B10.A Langerhans cells appeared in stripped C57BL/6 skin allografts on day 4; moreover, B10.A Langerhans cells never reappeared in stripped B10.A grafts placed on C57BL/6 hosts. We conclude that, after skin stripping, Langerhans cells are replaced almost exclusively from a systemic pool. This realization is provocative on two accounts. First, there appears to be no inherent (genetic) barrier to the physiologic infiltration of epidermis by histoincompatible Langerhans cells. One might have expected that something akin to allogeneic inhibition would render this result unlikely (25). These data imply that histocompatibility determinants, even those encoded by the MHC, do not guide (positively or negatively) the migration of Langerhans cells. Second, a reservoir of Langerhans cells (or their immediate precursors) exists in normal animals, a reservoir that can be called upon to deliver into the epidermis within a day's time a fresh supply. Our data are consistent with the hypothesis that the progenitor of Langerhans cells resides among hematopoietic stem cells. However, the rapidity of infiltration of stripped, intact skin with dendritic Langerhans cells is also consistent with the hypothesis that they come from a pool of recirculating dendritic cells, as postulated by Anderson (26) and by Steinman (27), and may thus represent a newly appreciated, distinctive type of peripheral blood leukocyte.

Summary

To explore the relationships among Ia antigen expression, epidermal Langerhans cells, and the immunogenicity of skin allografts, cellophane tape-stripping was used in H-2 congenic and recombinant mice of defined immunogenetic disparity. Tape-stripping of murine abdominal wall skin achieved almost complete depletion of epidermal Langerhans cells within a few hours of application, as measured by cell surface ATPase and expression of Ia antigens. Tape-stripping also reduced, to a considerable degree (but not absolutely), the Ia immunogenicity of skin allografts prepared from stripped surfaces. No comparable reduction in immunogenicity of class I major histocompatibility determinants was observed, suggesting that Langerhans cells are relatively unimportant in the presentation of H-2K antigens in skin grafts.

Langerhans cells reappear within 24 h of tape-stripping to anatomically intact skin, but are detectable in orthotopically grafted skin only after the graft has been in residence for 4 d, i.e., shortly after it has acquired a blood supply. Repopulating Langerhans cells at that time and thereafter are exclusively of host origin. These results indicate that the traffic of Langerhans cells to the skin can be extremely dynamic, especially when the epidermal surface has been markedly disturbed, and the data imply that, under normal circumstances, large numbers of Langerhans cells can be mobilized readily from an available pool of precursors.

We greatly appreciate the technical assistance of Ms. Debbie Bate and Mr. Eric Garland. We thank Dr. R. E. Billingham for critical review of the manuscript and Ms. Sara Howard for preparing the typed manuscript.

Received for publication 31 August 1981 and in revised form 9 November 1981.

References

1. Klein, J. 1975. Biology of the mouse histocompatibility-2 complex Springer-Verlag, Heidelberg. 329–346.
2. Hammerling, G. J. 1976. Tissue distribution of Ia antigens and their expression on lymphocyte subpopulations. *Transplant. Rev.* **30:**64.
3. Rowden, G., T. M. Phillips, and T. L. Delovitch. 1978. Expression of Ia antigens by murine keratinizing epithelial Langerhans cells. *Immunogeneitcs.* **7:**465.
4. Streilein, J. W., G. B. Toews, and P. R. Bergstresser. 1979. Corneal allografts fail to express Ia antigens. *Nature (Lond.).* **282:**326.
5. Wernet, P. 1976. Human Ia-type alloantigens.: methods of detection, aspects of chemistry and biology, markers of disease states. *Transplant. Rev.* **30:**271.
6. Hirschberg, H., T. Moen, and E. Thorsby. 1971. Complement and cell-mediated specific destruction of human endothelial cells treated with anti-DRw antisera. *Transplant. Proc.* **11:** 776.
7. Bergstresser, P. R., G. B. Toews, and J. W. Streilein. 1980. Natural and perturbed distributions of Langerhans cells: responses to ultraviolet light, heterotopic skin grafting, and dinitrofluorobenzene sensitization. *J. Invest. Dermatol.* **75:**73.
8. Streilein, J. W., G. B. Toews, and P. R. Bergstresser. 1980. Langerhans cells: functional aspects revealed by in vivo grafting studies. *J. Invest. Dermatal.* **75:**17.
9. Aberer, W., G. Schuler, G. Stingl, H. Honigsmann, and K. Wolff. 1981. Ultraviolet light depletes surface markers of Langerhans cells. *J. Invest. Dermatol.* **76:**202.
10. Lessard, R. J., K. Wolff, and R. K. Winkelmann. 1968. The disappearance and regeneration of Langerhans cells following epidermal injury. *J. Invest. Dermatol.* **50:**171.
11. Klein, J., L. Flaherty, J. L. VandeBerg, and D. C. Shreffler. 1978. *H-2* haplotypes, genes, regions, and antigens: first listing. *Immunogenetics.* **6:**489.
12. Bergstresser, P. R., C. R. Fletcher, and J. W. Streilein. 1980. Surface densities of Langerhans cells in relation to rodent epidermal sites with special immunologic properties. *J. Invest. Dermatol.* **74:**77.
13. Streilein, J. W., and J. Klein. 1977. Neonatal tolerance induction across regions of H-2 complex. *J. Immunol.* **119:**2147.
14. Litchfield, J. T., Jr. 1949. A method for rapid graphic solution of time-percent effect curves. *J. Pharmacol. Exp. Ther.* **97:**399.
15. Katz, S. I., K. Tamaki, and D. H. Sachs. 1979. Epidermal Langerhans cells are derived from cells originating in bone marrow. *Nature (Lond.).* **282:**324.
16. Frelinger, J. A., and J. G. Frelinger. 1980. Bone marrow origin of Ia molecules purified from epidermal cells. *J. Invest. Dermatol.* **75:**68.
17. Giacometti, L., and W. Montagna. 1967. Langerhans cells: uptake of tritiated thymidine. *Science (Wash. D. C.).* **157:**439.
18. Stingl, G., S. I. Katz, I. Clement, I. Green, and E. M. Shevach. 1978. Immunologic functions of Ia-bearing epidermal Langerhans cells. *J. Immunol.* **121:**2005.
19. Toews, G. B., P. R. Bergstresser, and J. W. Streilein. 1980. Epidermal Langerhans cell density determines whether contact hypersensitivity or unresponsiveness follows skin painting with DNFB. *J. Immunol.* **124:**445.
20. Steinmuller, D. 1967. Immunization with skin isografts taken from tolerant mice. *Science (Wash. D. C.).* **158:**127.
21. Lafferty, K. J. 1980. Immunogenicity of foreign tissues. *Transplantation.* **29:**179.
22. Bach, J. F. 1980. *Comment. Transplantation.* **29:**182.
23. Lafferty, K. J., M. A. Cooley, and J. Woolnough. 1975. Thyroid allograft immunogenicity is reduced after a period in organ culture. *Science (Wash. D. C.).* **188:**259.
24. Gose, J. E., and F. J. Bach. 1981. Rejection crises that resolve can be triggered by passenger cells in otherwise syngeneic transplants. *Transplant. Proc.* **13:**1117.

25. Hellstrom, K. E., and I. Hellstrom. 1967. Allogeneic inhibition of transplanted tumor cells. *Prog. Exp. Tumor Res.* **9**:40.
26. Anderson, A. O., J. T. Warren, and D. L. Gasser. 1981. Presence of lymphoid dendritic cells in thoracic duct lymph from Lewis rats. *Transplant. Proc.* **13**:1460.
27. Steinman, R. M., and M. C. Nussenzweig. 1980. Dendritic cells, features and functions. *Immunol. Rev.* **53**:127.