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CHAPTER 29.1

IMMUNOSUPPRESSION AND THE BIDIRECTIONAL PARADIGM OF TRANSPLANTATION IMMUNOLOGY

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Acquired transplantation tolerance without immunosuppression was described by Billingham, Brent and Medawar[1,2] more than four decades ago following the injection of immune competent adult spleen cells into immunologically immature mice in utero or perinatally. The engrafted donor cells flourished, perpetuated themselves and were thought in effect to have endowed the recipient with the donor immune system. This explanation of tolerance by immunologic replacement was strengthened by Main and Prehn[3,4] who demonstrated the same outcome in irradiated adult mice, whose cytoablated hematolymphopoietic cells were reconstituted with donor bone marrow.

The anticipated clinical application of tolerance induction for transplantation was temporarily derailed in 1957 when Billingham and Brent showed in mice[5,6] and Simonsen in chickens[7] that graft-versus-host ("runt") disease (GVHD) was a danger in these immunologically "unbalanced" systems in rough proportion to the degree of MHC disparity. In spite of the same limitations as with the radiation chimera models, a strategy for clinical bone marrow transplantation could be assembled directly from the Main-Prehn experiments, with similar results. Stable drug free chimerism could be induced in irradiated animals using MHC matched donor marrow, but otherwise there was an intolerable incidence of GVHD.

Translation of these experiments into successful clinical bone marrow transplantation in 1968[8,9] was supremely gratifying because it had been so logical as Nobel Laureate Thomas has summarized.[10] However, the achievement effectively detached from a scientific base, surgeons who by this time had recorded thousands of successful whole organ transplantations under continuous immunosuppression—without host preconditioning, dependence on MHC matching or problems with GVHD. The avalanche of whole organ cases had begun in 1962 when azathioprine was combined with prednisone.[11] A characteristic cycle was identified in which rejection could be reversed surprisingly easily with prednisone. More importantly, the need later on for maintenance immunosuppression frequently declined. The same sequence has been seen since with all other organs transplanted and with all of the immunosuppressive regimens. Something had changed in the host, the graft or both. But what?

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Thirty years and a revolution in immunology later, the question was answered.[12-17] Donor leukocytes of bone marrow origin which are part of the structure of all complex grafts (multilineage "passenger leukocytes"[18,19]) had migrated from the organs and survived ubiquitously in the recipients. Thus, organ transplantation and acceptance was a cryptic example of cell transplantation whereby a small fragment of extramedullary donor bone marrow had migrated and become assimilated into the overwhelmingly larger immunologic network of the host (microchimerism) (Fig. 29.1.1). The cell movement was in both directions, the interstitial leukocyte constituency of the grafts becoming predominantly that of the recipient. In successful cases, graft and host were both genetic composites!!!

From this information, we proposed a two-way paradigm of transplantation immunology.[12-17] It holds that the immunologic reaction following whole organ transplantation is bidirectional (GVH as well as HVG) and mutually canceling, providing the two participants in the David/Goliath mismatch survive the initial confrontation. In clinical practice, this requires an umbrella of immunosuppression that covers both equally. Although the chimeric leukocytes are multilineage,[20,21] the dendritic cells of Steinman and Cohen[22,23] are thought to be key participants.[12-19,24]

With the two-way paradigm, virtually every previously enigmatic problem observation after whole organ transplantation can be explained: why organ grafts can be tolerogenic, why GVHD does not develop after the transplantation of immunologically active grafts such as the liver and intestine,[25] and why HLA matching is so poorly predictive of outcome.[26] It also has called into question the deeply ingrained assumption that tolerance induction for whole organ recipients requires host preconditioning in preparation for a variety of donor leukocyte preparations. Instead, the development of donor specific nonreactivity is the consequence of the reciprocal immunologic transaction that is merely chaperoned, not caused, by diverse techniques of immunosuppression with which this book is concerned. Thus, the various drugs which interrupt various stages of the immune reaction are merely permissive of a natural event that is ultimately the same with all successful agents, no matter what their site of action.

If this hypothesis were valid, it should be possible to augment the natural passenger leukocyte traffic in organ recipients. Historical efforts (of which

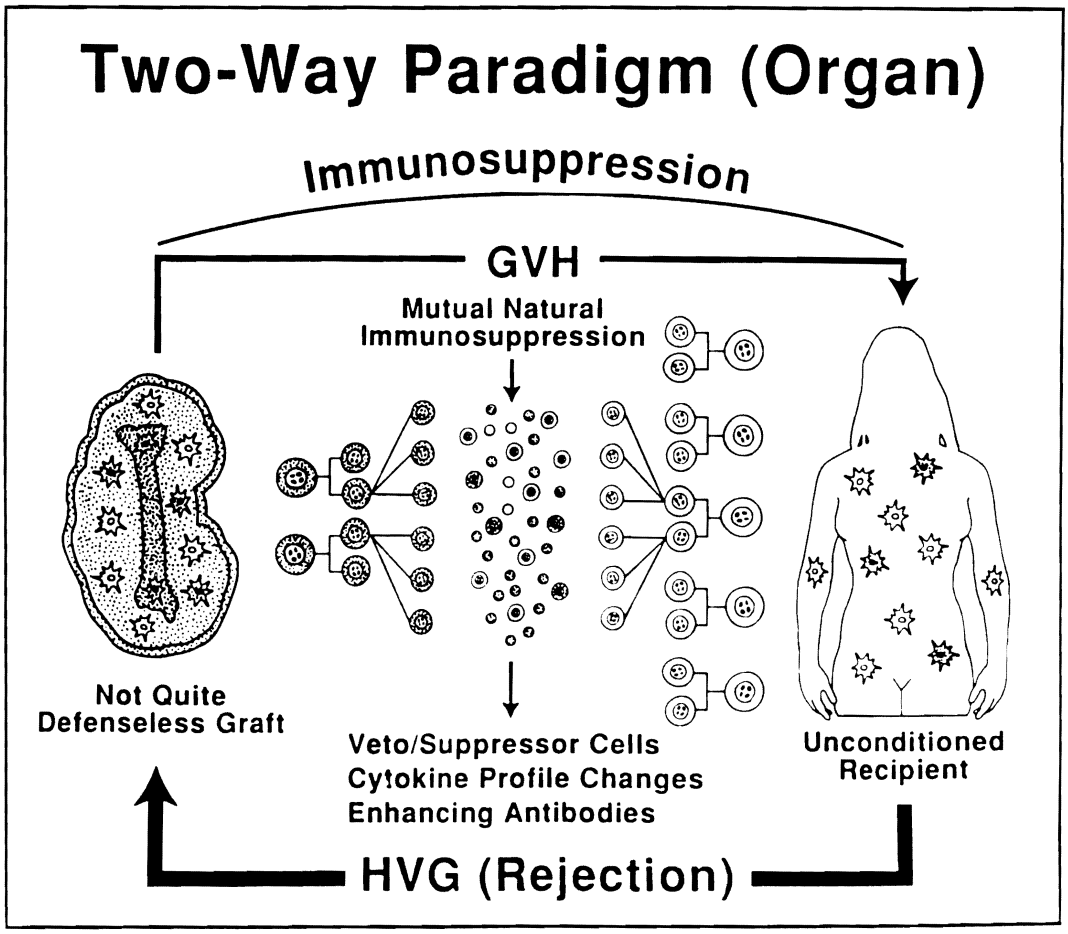

Fig. 29.1.1. Mechanism of whole organ graft acceptance involving a graft-versus-host (GVH) reaction by the bone marrow-derived donor leukocytes in the graft that are pitted against the whole recipient immunologic apparatus ([HVG] host-versus-graft, rejection). In standard clinical practice, the recipient is not preconditioned.

Monaco's were most noteworthy[27]) to give extra donor antigen in the form of bone marrow[27-32] or donor blood transfusions[33-36] were hampered in design or execution by the assumption that the infused cells would have a transient survival without preconditioning, justifiable anxiety about GVHD with any form of preconditioning and a lack of information about the appropriate timing of leukocyte infusions. Such trials have either failed outright or yielded equivocal results. Whether the administered donor leukocytes survived in these trials was unknown because no one looked for chimerism postoperatively.

The information that chimerism regularly follows successful whole organ transplantation exposed a perioperative window of opportunity during which unaltered MHC incompatible bone marrow or leukocyte donor specific blood could be given safely without recipient preparation or deviation from the standard clinical practices of immunosuppression that had empirically evolved.[37] In Pittsburgh, leukocyte augmentation (3×10^8/kg unaltered donor bone marrow perioperatively) has been carried out in 50 nonpreconditioned recipients of cadaveric kidneys, livers, hearts and lungs of whom only the first 18 have been recently reported; however, all 50 are well. Chimerism estimated to be >1000 times greater than that occurring spontaneously could be reliably produced and sustained under standard FK506-prednisone. Their persistent blood chimerism (usually >1%) trend toward donor specific nonreactivity and universal survival (along with the next 32 recipients) have marked them as an advantaged cohort[37] despite the fact that early (and easily controlled) rejection has been documented in more than half of the cases. Serious GVHD has not been observed. We have cautioned that chimerism is not synonymous with tolerance, but only a necessary condition for its attainment. The pace of drug weaning with the ultimate objective of drug discontinuance will have to be determined in each of these patients individually.

The bidirectional paradigm closes the vast gap between the bone marrow and organ transplant fields by showing how their perceived differences are direct reflections of treatment strategy—leaving intact the mutually censoring immunologic limbs in the whole organ recipient and deliberately trying to remove one of these in the conventional bone marrow recipient. However, in both kinds of patient, the appearance of MHC restricted veto and suppressor cells, enhancing antibodies and changes in cytokine profiles can be construed as by-products of and accessory to the seminal event of mixed chimerism. In the bone marrow recipient, the trace population is of host cells despite drastic cytoablation[38,39] and in the whole organ recipient the trace leukocyte population is of donor cells.

The fusion of whole organ and bone marrow transplantation into a unitarian world invites questions about the meaning of acquired transplantation tolerance. It seems obvious that essentially all examples of transplantation tolerance observed in either human whole organ or bone marrow recipients are independent of any single recipient organ or region and are thus "peripheral" rather than control. This insight about what is actually accomplished with the successful transplantation of either whole organs or bone marrow will require re-examination at many tenets of "classical" transplantation immunology, and should lead to improvements in therapy.

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REFERENCES

1. Billingham RE, Brent L, Medawar PB. "Actively acquired tolerance" of foreign cells. Nature 1953; 172:603-606.
2. Billingham R, Brent L, Medawar P. Quantitative studies on tissue transplantation immunity. III. Actively acquired tolerance. Philos Trans R Soc Lond (Biol) 1956; 239:357-412.
3. Main JM, Prehn RT. Successful skin homografts after the administration of high dosage X radiation and homologous bone marrow. J Natl Cancer Inst 1955; 15:1023-1029.
4. Main JM, Prehn RT. Fate of skin homografts in X-irradiated mice treated with homologous marrow. J Natl Cancer Instit 1957; 19:1053-1064.
5. Billingham R, Brent L. A simple method for inducing tolerance of skin homografts in mice. Trans Bull 1957; 4:67-71.
6. Billingham R, Brent L. Quantitative studies on transplantation immunity. IV. Induction of tolerance in newborn mice and studies on the phenomenon of runt disease. Philos Trans R Soc Lond (Biol) 1959; 242:439-477.
7. Simonsen M. The impact on the developing embryo and newborn animal of adult homologous cells. Acta Path Microbiol Scand 1957; 40:480.
8. Gatti RA, Meuwissen HJ, Allen HD, Hong R, Good RA. Immunological reconstitution of sex-linked lymphopenic immuno-logical deficiency. Lancet 1968; 2:1366-1369.
9. Bach FH. Bone-marrow transplantation in a patient with the Wiskott-Aldrich syndrome. Lancet 1968; 2:1364-1366.
10. Thomas ED. Allogeneic marrow grafting—A story of man and dog. In: Terasaki PI and Cecka JM, eds. Clinical Transplants. UCLA Press 1991:381-390.

11. Starzl TE, Marchioro TL, Waddell WR. The reversal of rejection in human renal homografts with subsequent development of homograft tolerance. Surg Gynecol Obstet 1963; 117:385-395.
12. Starzl TE, Demetris AJ, Murase N, Ildstad S, Ricordi C, Trucco M. Cell migration, chimerism, and graft acceptance. Lancet 1992; 339:1579-1582.
13. Starzl TE, Demetris AJ, Trucco M, Ramos H, Zeevi A, Rudert WA, Kocoua M, Ricordi C, Ildstad S, Murase N. Systemic chimerism in human female recipients of male livers. Lancet 1992; 340:876-877.
14. Starzl TE, Demetris AJ, Trucco M, Ricordi S, Ildstad S, Terasaki P, Murase N, Kendall RS, Kocoua M, Rudert WA, Zeevi A, Van Thiel D. Chimerism after liver transplantation for type IV glycogen storage disease and Type I Gaucher's disease. New Engl J Med 1993; 328:745-749.
15. Starzl TE, Demetris AJ, Trucco M, Murase N, Ricordi C, Ildstad S, Ramos H, Todo S, Tzakis A, Fung JJ, Nalesnik M, Rudert WA, Kocova M. Cell migration and chimerism after whole organ transplantation: The basis of graft acceptance. Hepatology 1993; 17: 1127-1152.
16. Starzl TE, Demetris AJ, Trucco M, Zeevi A, Ramos H, Terasaki P, Rudert WA, Kocova M, Ricordi C, Ildstad S, Murase N. Chimerism and donor specific nonreactivity 27 to 29 years after kidney allotransplantation. Transplantation 1993; 55:1272-1277.
17. Starzl TE, Demetris AJ, Murase N, Thomson AW, Trucco M, Ricordi C. Donor cell chimerism permitted by immunosuppressive drugs: A new view of organ transplantation. Immunol Today 1993; 14: 326-332.
18. Snell GD. The homograft reaction. Ann Rev Microbiol 1957; 11:439-458.
19. Steinmuller D. Immunization with skin isografts taken from tolerant mice. Science 1967; 158:127-129.
20. Demetris AJ, Murase N, Fujisaki S, Fung JJ, Rao AS, Starzl TE. Hematolymphoid cell trafficking, microchimerism, and GVHD reactions after liver, bone marrow, and heart transplantation. Transplantation Proc 1993; 25:3337-3344.
21. Qian S, Demetris AJ, Murase N, Rao AS, Fung JJ, Starzl TE. Murine liver allograft transplantation: Tolerance and donor cell chimerism. Hepatology 1994; 19: 916-924.
22. Steinman RM, Cohn ZA. Identification of a novel cell type in peripheral lymphoid organs of mice. I. Morphology, quantitation, tissue distribution. J Exp Med 1973; 137:1142.
23. Steinman RM. The dendritic cell system and its role in immunogenicity. Annu Rev Immunol 1991; 9:271-296.
24. Lu L, Woo J, Rao AS, Li Y, Watkins SC, Qian S, Starzl TE, Demetris AJ, Thomson AW. Propagation of dendritic cell progenitors from normal mouse liver using GM-CSF and their maturational development in the presence of type-I collagen. J Exp Med 1994; 179:1823-1834.
25. Starzl TE, Todo S, Tzakis A, Alessiani M, Casavilla A, Abu-Elmagd K, Fung JJ. The many faces of multivisceral transplantation. Surg Gynecol Obstet 1991; 172:335-344.
26. Starzl TE, Trucco M, Rao A, Demetris A. The blindfolding of HLA matching. In: Terasaki P, ed. Clinical transplants 1993-Current Opinions. Los Angeles, CA: UCLA Tissue Typing Laboratory, 1994:423-425.
27. Monaco AP, Clark AW, Brown RW. Active enhancement of a human cadaver renal allograft with ALS and donor bone marrow: Case report of an initial attempt. Surgery 1976; 79:384-392.
28. Najarian JS, Ferguson RM, Sutherland DER, Slavin S, Kim T, Kersey T, Simmons RL. Fractionated total lymphoid irradiation as preparative immunosuppression in high-risk renal transplantation. Ann Surg 1982; 196:442-452.
29. Kahn DR, Hong R, Greenberg AJ, Gilbert EF, Dacumos GC, Dufek JH. Total lymphatic irradiation and bone marrow in human heart transplantation. Ann Thorac Surg 1984; 38:169-171.
30. Myburgh JA, Meyers AM, Thomson PD, Botha JR, Margolius L, Lakier R, Smit JA, Stark JH, Gray C. Total lymphoid irradiation-current status. Transplant Proc 1989; 21:826-828.
31. Barber W H, Mankin J A, Laskow D A, Deierhol M H, Julian B A, Curtis J J, Diethelm AG. Long-Term results of a controlled prospective study with transfusion of donor specific bone marrow in 57 cadaveric renal allograft recipients. Transplantation 1991; 51:70-75.
32. Rolles K, Burrough AK, Davidson BR, Karatapanis S, Prentice HG, Hamon MD. Donor-specific bone marrow infusion after orthotopic liver transplantation. Lancet 1994; 343:263-265.
33. Anderson CB, Sicard GA, Etheredge EE. Pretreatment of renal allograft recipients with azathioprine and donor-specific blood products. Surgery 1982; 92: 315-341.
34. Sollinger HW, Burlingham WJ, Sparks EM, Glass NR, Belzer FO. Donor-specific transfusions in unrelated and related HLA-mismatched donor-recipient combinations. Transplantation 1984; 38:612-615.
35. Salvatierra O Jr, Melzer J, Potter D, Garovoy M, Vincenti F, Amend WJ, Husing R, Hopper S, Feduska NJ. A seven-year experience with donor-specific blood transfusions. Results and considerations for maximum efficacy. Transplantation 1985; 40:654-659.
36. Bean MA, Mickelson E, Yanagida J, Ishioka S, Brannen GE, Hansen JA. Suppressed antidonor MLC responses in renal transplant candidates conditioned with donor-specific transfusions that carry the recipient's noninherited maternal HLA haplotype. Transplantation 1990; 49:382-386.
37. Fontes P, Rao A, Demetris AJ, Zeevi A, Trucco M,

Carroll P, Rybka W, Ricordi C, Dodson F, Shapiro R, Tzakis A, Todo S, Abu-Elmagd K, Jordan M, Fung JJ, Starzl TE. Augmentation with bone marrow of donor leukocyte migration for kidney, liver, heart, and pancreas islet transplantation. Lancet 1994; 344: 151-155.

38. Hill RS, Petersen FB, Storb R, Appelbaum FR, Doney K, Dahlberg S, Ramberg R, Thomas ED. Mixed hematologic chimerism after allogeneic marrow transplantation for severe aplastic anemia is associated with a higher risk of graft rejection and a lessened indicence of acute graft-versus-host disease. Blood 1986; 67:811-816.
39. Petz LD. Documentation of engraftment and characterization of chimerism following marrow transplantation. In: Forman SJ, Blume KG, Thomas ED, eds. Bone Marrow Transplantation. Blackwell Scientific Publications, 1994:136-148.