© 2000 Nature America Inc. • http://medicine.nature.com

recipient NK cells. In this strain combination, radiation-sensitive T cells of the recipient can remain functional after sublethal total body irradiation and can cause rejection by responding to $H2^b$ alloantigens of the donor. It is not known whether recipient T cells can cause rejection after the lethal doses of irradiation used in Gandy's experiments. Thus, the authors have not definitively demonstrated that $TCR^-/CD8^+$ cells can overcome the engraftment barrier created by recipient T cells. In humans, T cells are involved in bone marrow graft rejection, wheras the role of NK cells remains uncertain. In mice, NK-mediated rejection can be overcome by treating the recipient with cyclophosphamide[8], an agent that is frequently included as part of the pretransplant regimen for humans.

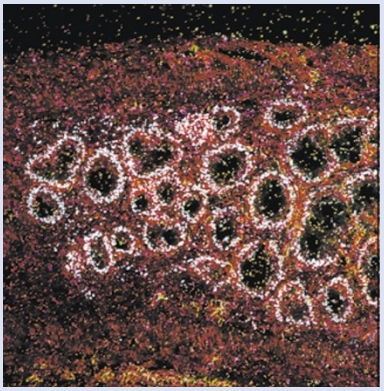
Gandy *et al.*[1] showed that $TCR^-/CD8^+$ cells were most effective in facilitation when limited numbers of highly enriched hematopoietic stem cells were used for transplantation. The effects were less profound when larger number of stem cells were given to recipients. In humans, the number of hematopoietic cells that can be obtained by aspiration of marrow is limited. Much larger numbers of stem cells can be obtained by apheresis of peripheral blood leukocytes from donors treated with a hematopoietic growth factor such as granulocyte colony-stimulating factor. With apheresis products used as a source of stem cells instead of aspirated marrow, donor T cells are not required for facilitation of engraftment when potent immunosuppressive regimens are given before transplantation[9]. In theory, less-intensive and less-toxic pre-transplant conditioning regimens would be sufficient if $TCR^-/CD8^+$ cells could be used to facilitate engraftment when T cells are depleted from the graft in order to prevent GVHD. In practical terms, however, it will be necessary to develop simple and convenient methods for isolating $TCR^-/CD8^+$ cells before any clinical trials can be undertaken.

The experiments by Gandy *et al.*[1] used highly enriched hematopoietic stem cells as a scientifically rigorous way to identify populations that facilitate engraftment. From a medical point of view, however, the use of stem cell populations exhaustively depleted of T cells for hematopoietic engraftment can cause highly undesirable effects in humans. Donor T cells mediate a 'graft-versus-leukemia' effect that helps to eliminate any malignant cells that could otherwise cause recurrent malignancy after the transplant. In addition, adults have reduced thymic function, and T-cell reconstitution after transplantation originates from T cells in the graft[10]. Hence, the use of purified stem cells together with $TCR^-/CD8^+$ cells for transplantation will require additional measures to prevent recurrent malignancy and promote immune reconstitution as compensation for the absence of T cells in the graft. Considerable development will be needed before $TCR^-/CD8^+$ cells can be used clinically in marrow transplantation. Nonetheless, the observations by Gandy *et al.*[1] have opened the way to an interesting new approach to regulate immune responses against alloantigens.

1. Gandy, K.L., Domen, J., Aguila, H., & Weissman, I. L. $CD8^+$ TCR^+ and $CD8^+TCR^-$ cells in whole bone marrow facilitate the engraftment of hematopoietic stem cells across allogeneic barriers. *Immunity* **11**, 579–590 (1999).
2. Martin, P.J. Prevention of allogeneic marrow graft rejection by donor T cells that do not recognize recipient alloantigens: potential role of a veto mechanism. *Blood* **88**, 962–969 (1996).
3. Martin, P.J., Akatsuka, Y., Hahne, M. & Sale, G. Involvement of donor T-cell cytotoxic effector mechanisms in preventing allogeneic marrow graft rejection. *Blood* **92**, 2177–2181 (1998).
4. Martin, P.J. Donor CD8 cells prevent allogeneic marrow graft rejection in mice: potential implications for marrow transplantation in humans. *J. Exp. Med.* **178**, 703–712 (1993).
5. Martin, P.J. *et al.* A phase I-II clinical trial to evaluate removal of CD4 cells and partial depletion of CD8 cells from donor marrow for HLA-mismatched unrelated recipients. *Blood* **94**, 2192–2199 (1999).
6. Kaufman, C. L. *et al.* Phenotypic characterization of a novel bone marrow-derived cell that facilitates engraftment of allogeneic bone marrow stem cells. *Blood* **84**, 2436–2446 (1994).
7. Asiedu, C. *et al.* Immunoregulatory role of CD8α in the veto effect. *Transplantation* **67**, 372–380 (1999).
8. Cudkowicz, G. & Bennett, M. Peculiar immunobiology of bone marrow allografts. I. Graft rejection by irradiated responder mice. *J. Exp. Med.* **134**, 83–102 (1971).
9. Aversa, F. *et al.* Treatment of high-risk acute leukemia with T-cell-depleted stem cells from related donors with one fully mismatched HLA haplotype. *N. Engl. J. Med.* **339**, 1186–1193 (1998).
10. Mackall, C.L., Hakim, F.T., & Gress, R.E. Restoration of T-cell homeostasis after T-cell depletion. *Sem. Immunol.* **9**, 339–346 (1997).

University of Washington School of Medicine
Fred Hutchinson Cancer Research Center
1100 Fairview Ave. North, D2-100
P.O. Box 19024
Seattle, Washington 98109-1024, USA
Email: pmartin@fhcrc.org

A new match-maker



Repair of DNA mismatches, and of small insertions and deletions, is a fundamental element of genome stability. In humans, these processes are accomplished by various combinations of two classes of factors, MSH (homologs of bacterial MutS) and MLH (homologs of bacterial MutL), some of which are known as PMS, because certain yeast proteins of this class were originally identified as modifiers of post-meiotic segregation. Mutations in some, but not all, of these factors are strongly correlated with hereditary non-polyposis colon cancer (HNPCC) and can cause a variety of defects in DNA repair, which can be confined to either mismatch repair, the repair of small insertions and deletions, or both. Deficiencies in the latter process are manifested as microsatellite instability (MSI), a marker for early-onset HNPCC.

In the January issue of *Nature Genetics*, Lipkin *et al.* have reported the identification of another repair factor, MLH3, that interacts with the previously characterized MLH1. Like many of its relatives, MLH3 is closely related to a known yeast protein, Mlh3p. Expression of a mutated dominant negative form of MLH3 induces MSI in cell lines, making it a strong candidate for cancer susceptibility. Moreover, the mouse homolog, *Mlh3*, maps to the complex trait locus Ccs1, which may be involved in susceptibility to colon cancer and produces an MSI phenotype. Additionally, *in situ* hybridization data (see picture) indicates that Mlh3 is expressed on epithelial cells of the colon.

But do mutations in *MLH3* underlie human colon cancer susceptibility? A paper published by Weber *et al.* in *Cytogenetics and Cell Genetics* **86**, 142–147 (1999) reports loss of heterozygosity in the region of chromosome 14q containing *MLH3* in approximately 30% of human colorectal cancers. Lipkin's group is also investigating whether specific mutations in *MLH3* are associated with susceptibility to colon and other types of human cancers.

Michael Ronemus

© 2000 Nature America Inc. • http://medicine.nature.com