



1921

Hematopoietic Reconstitution by Transplanted Grafts in Lethally Irradiated Recipients

Q. Ye, S.H. Chia, S. Doughton, T. Sakamoto, F. Galvao, A.J. Demetris, T.E. Starzl, and N. Murase

PERSISTENCE of multilineage microchimerism has been shown in recipients of successful solid organ transplantation.^{1,2} Continuous existence for decades of a trace number of donor cells implicates long-lasting engraftment of donor hematopoietic progenitor cells originating from the transplanted organs. We have recently showed that syngeneic organ transplantation was able to protect animals from lethal irradiation.³ This study further describes multilineage differentiation and self-renewal capacity of hematopoietic stem cells existing in solid organs.

MATERIALS AND METHODS

Inbred Lewis rats (Harlan Sprague Dawley, Indianapolis, Ind) were used for all experiments. Bone marrow cells (BMC) were obtained by flushing the medullary cavity of the tibias and femurs with RPMI medium. Heterotopic heart (HTx) and orthotopic liver transplantation (OLTx) in rats were performed according to previous publications.³

RESULTS

When Lewis rats received 9.5 Gy irradiation, all animals died in 12 days. Radioprotective effect of syngeneic BMC was cell dose dependent. Injection of 0.5×10^6 unfractionated BMC was not successful in rescuing irradiated animals and all died within 17 days. On the other hand, 1×10^6 or more BMC rescued all irradiated animals. After HTx, survival of irradiated recipients was significantly prolonged by a median of 15 days and one of six survived for 100 days. Syngeneic OLTx to irradiated recipients allowed five of six to survive for more than 100 days. Partial or complete protection of animals from lethal irradiation with organ transplantation strongly supports the existence of hematopoietic stem cells in these organs. Sequential analysis of total leukocytes and hematocrit values in these animals revealed that hematopoietic recovery after syngeneic OLTx was similar to that of 1×10^6 BMC infusion, suggesting that the hematopoietic function of the whole liver is comparable to that of 1×10^6 BMC. Recovery of granulocytes and lymphocytes (T, B, and NK cells) was also seen at a similar tempo to that of BMC. When splenocytes obtained from irradiated female recipients of male liver grafts were sorted

with monoclonal antibodies specific for T (R7.3), B (OX33), and NK (NK3.2.3.) cells and analyzed by polymerase chain reaction (PCR) using a Y-chromosome-specific probe, repopulating male cells were found in all fractions, suggesting multilineage reconstitution by OLTx. Furthermore, frequencies of hematopoietic progenitor cells (colony-forming unit in culture [CFU-C] per 5×10^6 nylon wool nonadherent BMC) in the bone marrow of reconstituted animals were 138 ± 8 and 128 ± 13 with OLTx and BMC (1×10^6), respectively. These results were equivalent to CFU-C in normal animals (165 ± 50).

DISCUSSION

These results implicate that hematopoietic progenitor cells normally exist in solid organs. It is not unreasonable to expect that these progenitor cells play a role in maintaining microchimerism after allogeneic transplantation. Primarily, organ transplantation transfers normal physiologic function of organs to recipients, however, at the same time miniature donor immune system in grafts may be carried into recipients. Multilineage composition of chimerism and existence of donor hematopoietic progenitor cells in the graft support that the establishment of donor immune system occurs in transplant recipients. Mechanisms by which two immune systems of donor and recipient cooperate in transplant recipients are currently under investigation.

REFERENCES

1. Starzl TE, Demetris AJ, Murase N, et al: *Lancet* 239:1579, 1992
2. Starzl TE, Demetris AJ, Trucco M, et al: *Hepatology* 17:1127, 1993
3. Murase N, Starzl TE, Ye Q, et al: *Transplantation* 61:1, 1996

From the Thomas E. Starzl Transplantation Institute, Departments of Surgery (Q.Y., S.H.C., S.P., F.G., T.E.S., N.M.) and Pathology (T.S., A.J.D.), University of Pittsburgh Medical Center, Pittsburgh, Pennsylvania.

Address reprint requests to Q. Ye, Thomas E. Starzl Transplantation Institute, Department of Surgery, University of Pittsburgh Medical Center, Pittsburgh, PA 15261.