

Hamster-to-Rat Liver Xenografts Protect Extrahepatic Organs From Rejection

L.A. Valdivia, A.J. Demetris, J.J. Fung, S. Celli, C. Frye, N. Murase, and T.E. Starzl

THE liver allograft is relatively resistant to the hyperacute rejection caused by preformed antibodies,¹ and in some species and strain combinations it is accepted without immunosuppression.^{2,3} We have shown a similar resistance of the liver to humoral rejection after hamster to rat xenotransplantation.^{4,5} Moreover, it has been shown that the liver allograft can induce tolerance to other organs from the same donor strain² and that it also can shield these organs from hyperacute rejection in a presensitized recipient.⁶ Thus, in this study we tested if hamster livers previously transplanted under FK 506 would ameliorate the otherwise intractable rejection of hamster heart and skin xenografts in rats.

MATERIALS AND METHODS

Golden Syrian hamsters were the donors and Lewis rats the recipients. Liver transplants were orthotopic and heart transplants were heterotopic.

Experimental Design

Lewis rats bearing hamster livers for 40 to 50 days under daily 1 mg/kg/d FK 506 had their immunosuppression stopped for 2 weeks on the day of skin or cardiac transplantation from third party (outbred) hamsters or from C3H mice. To study donor specificity, five Lewis rats were given two skin grafts on the day of liver transplantation under FK 506, one from the liver donor, and the other from a third party hamster.

Serum Transfer

Lewis rats were transplanted with hamster hearts and given serum from untreated or FK 506-treated liver xenograft recipients on day 6 or more than 50 days after hepatic grafting (0.5 mL/d \times 4, IV).

Adoptive Cell Transfer

Lewis rats ($n = 5$ per group) were sublethally irradiated with 7.5 Gy and transferred with lymph node cells (LNCs, 5×10^7) from naive Lewis, liver xenotransplanted Lewis rats, or a combination of both. Then, they received hamster and C3H mice skin grafts on their chest.

Mixed Lymphocyte Reaction (MLR)

LNCs from unmodified Lewis rats or those obtained from liver xenograft recipients on day 40 to 50 after transplantation were used as responders (1.75×10^5 /well), while LNCs from hamster, ACI rat, or C3H mice (2.9×10^5 /well) were used as stimulators.

RESULTS

Daily treatment of Lewis rats with FK 506 prolonged the mean survival of hamster liver xenografts from 7 to 67

days, but extended the 6-day control survival of skin grafts by only 3 days, and had no effect on the survival of heart xenografts, which underwent antibody-mediated rejection after 3 days with or without FK 506. In contrast, when hamster heart or skin grafts were transplanted at the same time as FK 506 was discontinued in rats already bearing hamster livers for 40 to 50 days, they were accepted for the ensuing 2 weeks of no treatment and thereafter with resumption of therapy (hearts: 63.0 ± 23.4 days; skin grafts: 75.7 ± 29.9 days). Under the same conditions, C3H mouse heart and skin xenografts were promptly rejected (2.7 ± 0.5 days and 10.5 ± 1.2 days, respectively). To assess the effect of residual immunosuppression, control Lewis rats without liver transplantation were pretreated for 30 days with 1 mg/kg/d FK 506 before test heart or skin xenografting, after which no treatment was given. When transplanted alone, survival of the hamster skin was prolonged an average of 3.0 days by the 30 days pretreatment but survival of the heart xenografts was the same as in the untreated controls. Mouse skin but not mouse hearts also had slight prolongation of survival after recipient pretreatment. In the experiments designed to test donor specificity both the skin grafts from the liver donor and from a third party hamster transplanted on the day of liver grafting had prolonged survival, being alive as long as the recipients lived (a mean of 73 and 69 days, respectively).

Serum transfer experiments showed no prolongation of test heart xenografts by any of the given sera, with all groups rejecting in a mean of 3 days. Adoptive transfer experiments showed that when cells from liver xenograft recipients were transferred hamster skin grafts were rejected in a mean of 15 days, which was similar as radiation controls (16 days). However, rejection of C3H mice skin grafts occurred in 9 days, as with transfer of naive rat cells. The proliferative response of the liver xenograft recipients was reduced to 50% against hamster and C3H mice LNCs, but that against ACI rat LNCs was normal.

From the Pittsburgh Transplant Institute, and the Departments of Surgery (L.A.V., J.J.F., S.C., C.F., N.M., T.E.S.) and Pathology (A.J.D.), University of Pittsburgh Health Science Center, Pittsburgh, Pennsylvania.

Address reprint requests to Thomas E. Starzl, MD, PhD, Department of Surgery, 3601 Fifth Ave, 5C Falk Clinic, University of Pittsburgh, Pittsburgh, PA 15213.

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DISCUSSION

This study has shown that the immunologic privilege of the liver in the hamster to rat xenograft model is qualitatively similar to that in hepatic allotransplantation models. Several explanations have been given to explain the liver's relative ability to withstand an antibody attack⁷ including the protection to its microvasculature provided by a double blood supply, its large microvascular surface available for antibody absorption, and secretion of new soluble class I antigens or IgG types that theoretically could neutralize preformed antigraft antibodies. We could not find evidence for the presence of enhancing serum factors or suppressor cells in our model. We found a state of unresponsiveness *in vivo* and hyporesponsiveness *in vitro*.

It is known that the Kupffer cells and macrophages lining the sinusoids are replaced in all successful hepatic grafts with recipient cells.⁸ This same kind of repopulation has been found in long surviving hamster to rat liver and cardiac xenografts.⁹ There also is evidence that the dendritic and lymphoid cells leaving the graft migrate to and nest in widely distributed host lymphoid and other tissues.¹⁰ The extent to which the peripheralization of donor cells and consequent systemic microchimerism occur depends on the immunologic substrate available in the donor organ for cell peripheralization; the liver is richly endowed with these cells. Thus, our conclusion is that the same explanation of the tolerogenicity of liver allografts applies

to xenografts. Achievement of cell repopulation requires potent immunosuppression with agents like FK 506 during the cell transition. In xenotransplant models, it also depends on the avoidance of antibody rejection which is more easily accomplished with the liver than other organs. This is the first demonstration of such hepatic tolerogenicity in a xenograft model.

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