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Influence of Donor-Recipient Strain Combinations on Immunologic Responses after Allogeneic Rat Small Bowel Transplantation

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GENETIC FACTORS are known to control alloreactivity and can affect the outcome after orthotopic small bowel transplantation (SBTx). The present study was undertaken to determine the effect of donor and recipient strains on long-term graft viability and the incidence of graft versus host disease (GVHD) after rat SBTx.

MATERIALS AND METHODS

Operation and Experimental Design

Orthotopic SBTx with portocaval drainage[1] was performed with or without immunosuppression in 12 fully allogeneic rat strain combinations between LEW (RT^l), BN (RT^n), PVG ($RT1^c$), and ACI ($RT1^a$) rats (purchased from Harlan Sprague-Dawley Inc, Indianapolis, IN). For immunosuppression, FK 506 dissolved in HCO-60 and D-mannitol carrier solvent (gift from Fujisawa Pharm Co Ltd, Osaka, Japan) was given intramuscularly (0.64 mg/kg/d on days 0 to 13).

Immunologic Testing of Long-Term Survivors after SBTx

Animals surviving > 150 days without signs of rejection were further evaluated for their immunologic status by skin grafting, mixed lymphocyte reaction (MLR), and cell-mediated lympholysis (CML). Recipient chimerism was also studied by immunohistochemistry.

RESULTS

Animal Survival after SBTx

In all 12 combinations, untreated recipients died of rejection with median survivals of 5 to 14 days. There were no statistically significant differences in survival when the results were examined from the perspective of either the donor or the recipient strain.

Treatment with FK 506 significantly prolonged overall survival ($P < .01$); however, the degree of prolongation and incidence of GVHD was dependent on the recipient strain. Survival of BN recipients was less than that of any other recipients regardless of the donor strain. However, within the BN group, the specific cause for the poor outcome differed. BN recipients of ACI grafts died of rejection with a median survival of 91 days. When PVG or LEW donors were used, the BN recipients invariably succumbeded to GVHD between 32 and 42 days. No other recipients had any signs of GVHD when treated with FK 506. Survival of the LEW recipients showed some dependence on the donor strain. ACI grafts were most promptly rejected, with a median survival of 133 days. Although most of the LEW recipients of BN (6/10) or PVG (3/4) grafts survived for more than 150 days, 70% of the BN grafts and 50% of the PVG grafts in LEW recipients were associated with diarrhea and more than 10% body weight loss. In contrast, all PVG and ACI recipients survived for > 150 days with a steady weight gain, except for three rats that died early because of obstructive ileus.

Pathologic Observations (HE Staining)

Results of the examination of specimens from treated animals were congruent with the clinical impression in different strain combinations. The skin of BN recipients of LEW or PVG bowel grafts showed changes typical of acute GVHD.[2] The bowel graft was normal in these animals. In contrast, findings of the ACI grafts in BN recipients and all strain grafts in LEW recipients are diagnostic of chronic rejection.[3] Although both ACI and PVG recipients appeared healthy after SBTx, the grafts in PVG recipients showed minimal changes of chronic rejection, whereas ACI recipients appeared to be completely spared from rejection. Because signs of rejection were minimal to rare in treated ACI and PVG recipients, they were further evaluated for their immunologic status.

Immunologic Testing of Long-Term Survivors after SBTx

Skin Graft Challenge. Long-surviving PVG and ACI recipients of bowel grafts rejected third-party skin graft in the same tempo as naive animals (median = 10.5 and 10 days, respectively). In contrast, the survival of skin grafts from the bowel donor strain was more than double in PVG recipients (median = 23.5 days) and indefinite (median > 100 days) in ACI recipients.

MLR. Long-surviving PVG recipients normally responded to bowel donor and third-party stimulators. In contrast, all long-surviving ACI recipients had reduced reactivity to donor strain stimulators but responded normally to third-party stimulators.

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CML. The CML responses of long-surviving ACI recipients to donor strain and third-party targets were essentially identical to naive ACI responses.

Assessment for Microchimerism. Immunopathologic study of lymphoid tissues obtained from long-surviving ACI recipients of PVG grafts revealed some $OX27^+$ cells (PVG donor origin). However, the presence of ACI donor cells ($MN4\text{-}91\text{-}6^+$) was less obvious in the long-surviving PVG recipients of ACI grafts.

DISCUSSION

Our results show that a short course of immunosuppression unmasks strong recipient effects on the outcome of isolated small bowel transplantation. Both ACI and PVG recipients demonstrated consistent long-term survival, whereas LEW recipients showed substantial chronic rejection regardless of donor strain. On the other hand, BN recipients were relatively unique in experiencing fatal GVHD with PVG or LEW grafts, whereas they aggressively rejected ACI grafts. The unique susceptibility of BN recipients to GVHD might have some relationship to the relative ease with which autoimmune disease was induced by injecting heavy metals to this strain.[4]

Immunohistochemical staining showed that the ACI recipients, which were tolerant of subsequent donor skin grafts, were microchimerics. Persistence of donor cells in the recipients has been thought to be a key for graft acceptance.[5] Although a correlation was noted between the clinical and pathologic condition of the bowel grafts and subsequent skin graft acceptance, the same concordance was not reflected in the in vitro immunologic testing. Perhaps isolation of the recipient cells from naturally occuring microenvironmental factors and network reactions, which have been altered by transplantation, is responsible for this enigma.

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