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Long-Term Function and Morphology of Intestinal Allografts in Outbred Canine Transplantation Model

K. Iwanami, T. Ishikawa, T. Okuda, Y. Zhu, A. Tuerler, B.A. Moore, S. Zhang, A.J. Bauer, R. Venkataramanan, K. Abu-Elmagd, T.E. Starzl, M.A. Nalesnik, and N. Murase

THE OUTCOME OF small bowel transplantation (SBTx) has improved during the last 10 years[1]; however, information on function and morphology of intestinal allografts long term after SBTx is limited.[2] Using canine allo-SBTx model, this study investigated the absorption, motility, and histopathology of long-surviving intestinal allografts.

MATERIALS AND METHODS

Fourteen outbred hound dogs of both sexes weighing 17 to 24 kg with the mean age of 22.6 ± 3.1 months underwent orthotopic transplantation of the entire small bowel as previously described.[3] Four recipients received autologous grafts (Group 1). Ten recipients of allogeneic SBTx were treated with cyclosporine (CsA, 20 to 30 mg/kg/day) and retrospectively divided into two groups according to the absence (Group 2, $n = 6$) or presence (Group 3, $n = 4$) of clinical symptoms, including continuous diarrhea, anorexia, malnutrition, and severe body weight loss. Between 110 and 210 days after auto- and allo-SBTx, intestinal graft function was evaluated by pharmacokinetics of oral CsA (10 mg/kg) and D-xylose absorption test (0.5 g/kg). Intestinal transit time was also determined by a barium X-ray study. In addition, intestinal graft tissues were obtained by open biopsy in recipients of Groups 1 and 2 and by sacrifice in Group 3. Samples were analyzed for intestinal brush border glycosidase[4] activity, in vitro circular muscle mechanical activity assay,[5] and histopathology. Histopathological changes were graded with a blinded fashion.

RESULTS

Recipients in Groups 1 (auto-SBTx) and 2 (allo-SBTx without symptoms) had comparable body weight gain by the time of this study (105% of pretransplant weight) with satisfactory serum total protein and cholesterol levels (Table 1). In contrast, recipients in Group 3 developed severe diarrhea 30 to 40 days after SBTx and suffered from malnutrition and significant body weight loss (76.5% of pretransplant weight) in spite of their voracious appetite. Intestinal transit time of recipients in Groups 1 and 2 at 4 to 7 months after SBTx was 185 minutes, slightly shorter than that of normal unoperated dogs; but it was extremely shortened to 60 ± 30 minutes in Group 3. Pharmacokinetics of CsA correlated well with overall recipient nutritional condition: peak blood concentration (Cmax, ng/mL) and area under the curve (AUC, ng × h/mL) of CsA in Groups 1 and 2 were comparable to those seen in normal unoperated dogs. In contrast, they were significantly decreased in Group 3. D-xylose absorption was inferior in all groups compared to normal animals; especially, recipients in Group 3 showed 65.3% reduction. These results indicated comparable long-term function of intestinal allografts in Group 2 and autografts in Group 1, and impaired intestinal function in Group 3 allografts.

From the Departments of Surgery (K.I., T.I., Y.Z., S.Z., K.A.E., T.E.S., N.M.),Pathology (R.V., M.A.N.), Thomas E. Starzl Transplantation Institute, University of Pittsburgh Medical Center, Pittsburgh, Pennsylvania and Medicine/Gastroenterology (A.T., B.A.M., A.J.B., R.V.), Thomas E. Starzl Transplantation Institute, University of Pittsburgh Medical Center, Pittsburgh, Pennsylvania, USA.

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Address reprint requests to K. Iwanami, MD, 200 Lothrop St, E1555 Biomedical Science Tower, Pittsburgh, PA 15261.

Table 1. Intestinal Function Long Term After Auto- and Allo-Small Bowel Transplantation

	Percent BW	Defecation/Day	Total Protein (g/dL)	Total Cholesterol (ng/dL)	CsA/Cmax (ng/mL)	CsA/AUC (ng × hr/mL)	D-xylose/2 hr (mg/dL)	Maltase (nmol/min · mg tissue)	Intestinal Transit (min)
Normal		2.5 ± 0.2	6.3 ± 0.1	221 ± 41	1592 ± 221	8352 ± 298	59.5 ± 6.9	14.9 ± 3.2	220 ± 10
Group 1	5.6 ± 0.3	3.5 ± 0.5	6.2 ± 0.2	194 ± 27	1086 ± 30	9366 ± 925	30.0 ± 7.4*	16.3 ± 2.9	185 ± 20
Group 2	5.3 ± 3.0	5.3 ± 0.5*	6.2 ± 0.7	174 ± 8	1879 ± 180	11505 ± 1646	43.2 ± 6.8	16.0 ± 5.8	189 ± 24
Group 3	−23.5 ± 1.6	7.3 ± 0.1*	4.1 ± 0.5*	107 ± 13*	732 ± 45	2445 ± 248*	23.8 ± 6.8*	13.8 ± 3.6	60 ± 30*

Values are expressed as mean ± SE.
*$P < .05$ compared with normal controls (ANOVA).

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In vitro circular muscle contractile activity with and without bethanechol stimulation was nearly normal in both Groups 1 and 2, but allografts in Group 3 showed significantly decreased contraction after bethanechol stimulation. Mucosal maltase activity was not significantly different among normal control and three transplant groups. Histological evidence of mucosal changes, including villous atrophy, inflammation, and erosion, were only seen in Group 3, and allografts in Group 2 showed normal slender villi without any inflammation or increase of crypt apoptosis. Interestingly, however, mild to moderate inflammation in the submucosa and muscle layers, especially near the Auerbach's plexus, was seen in all allografts in Groups 2 and 3, regardless of clinical symptoms. Furthermore, arterial changes, such as arteritis, focal intimal inflammation, and intimal thickening, were noticed in 25% of Group 2 and 75% of Group 3 allografts. These latter changes in the submucosa and muscle layers of the intestine, as well as graft vascular changes, were not correlated with any clinical symptom or nutritional status of recipients. Autografts in Group 1 were intact and totally free from these histopathological changes.

DISCUSSION

These results indicate that denervated intestinal allografts are able to efficiently digest and absorb nutrients to maintain and support lives. Allograft rejection appears to remain a major obstacle for long-term adequate intestinal graft function. Clinical symptoms in this study were only associated with histopathological changes in mucosal layer, indicating that the histopathological changes in the submucosa and muscle layers of the intestinal allografts do not correlate with any clinical symptom, and the ultimate significance of these changes remains a topic of study. Further efforts will be required to diagnose rejection of intestinal allografts that are functioning long term after SBTx.

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