1310

Functional Analysis of Graft Lamina Propria Associated Lymphocytes From a Recipient of a Human Cadaveric Small Bowel Allograft Primarily Immunosuppressed With FK 506

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TRANSPLANTATION of the intestine means transplantation of viable donor lymphocytes into the recipient. Therefore, in addition to rejection, there is the possibility of graft-vs-host disease (GVHD).[1] More potent immunosuppressants, such as FK 506, have made successful clinical small bowel transplantation a reality.[2] With improved clinical outcomes[3] it is valuable to assess the functional capabilities of the lamina propria associated lymphocytes (LPAL). Our group has studied the LPAL from serial intestinal graft biopsies taken from a recipient who primarily receives FK 506 immunosuppression.

THE PATIENT

The patient is a 27-year-old woman who, secondary to a hypercoagulable state, thrombosed her mesenteric circulation resulting in necrosis of her small intestine. Liver failure was secondary to chronic total parenteral nutrition therapy. She received a liver/small intestine allograft from an ABO compatible donor. The patient was crossmatch negative. The patient remained free of rejection until postoperative day (POD) 34. She was treated successfully and had a normal biopsy at POD 55. There were no episodes of GVHD.

MATERIALS AND METHODS

LPAL were propagated from serial proximal and distal mucosal biopsies by culturing divided biopsies for 2 weeks in RPMI 1640 plus 5% human AB serum, and 30 U/mL recombinant interleukin-2 (IL-2). Propagated LPAL from five serial biopsies of graft jejunum and ileum were tested for primed proliferative activity when challenged with irradiated donor splenocytes in a 3-day

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Table 1.

Biopsy Site and Date	Primed Lymphocyte Test†			Cell-Mediated Lympholysis‡	Histology
	Background	IL-2 Response	Donor Spleen Cells		
Jejunum POD 10	370 ± 107	17,347 ± 513	22,141 ± 19,040	4%	—
Ileum POD 10	174 ± 47	12,342 ± 45	12,651 ± 8,881	9%	—
Jejunum POD 15	177 ± 47	8273 ± 905	292 ± 395	4%	—
Ileum POD 15	1,093 ± 669	14,937 ± 3,464	3,964 ± 1,165	0%	—
Jejunum POD 25	65 ± 18	35,025 ± 2,779	16,097 ± 331	1%	—
Ileum POD 25	71 ± 59	27,457 ± 3,998	296 ± 40	4%	—
Jejunum POD 34	264 ± 66	14,409 ± 1,119	17,260 ± 1,595	51%	+
Ileum POD 34	534 ± 191	26,846 ± 3,323	37,489 ± 5,201	13%	+
Jejunum POD 55	273 ± 257	4,751 ± 744	367 ± 50	0%	—
Ileum POD 55	221 ± 208	1,771 ± 355	174 ± 67	0%	—

†Results of the PLT are expressed in counts per minute uptake of tritiated thymidine.
‡The effector to target cell ratio was 10:1 in all assays. Percentage values indicate (^{51}Cr experimental − spontaneous release)/(triton total − spontaneous release) × 100.

assay. Proliferative activity was measured as uptake of tritiated thymidine. Additional LPAL were tested for cytotoxic activity against donor splenocyte targets labelled with ^{51}Cr in a standard 4-hour cell-mediated lympholysis assay.

RESULTS

Primed proliferative alloreactive T lymphocytes were propagated from four of eight biopsies that were declared histologically "normal" (Table 1). However, none of the eight normal biopsies yielded a population of T cells that were capable of killing donor targets (>10% cytotoxicity). T lymphocytes propagated from biopsies taken during this patient's one episode of rejection (POD 34) yielded both primed proliferative *and* cytotoxic alloreactive T lymphocytes (Table 1). Follow-up biopsy at POD 55 (after solumedrol therapy) yielded neither proliferative nor cytotoxic lymphocytes.

CONCLUSIONS

Propagation of T cells demonstrating primed proliferative responses to donor antigens occurred nonspecifically throughout this patient's postoperative course. These cells likely represent a very small and adequately suppressed population of immunocompetent lymphocytes infiltrating the graft lamina propria. Their potential capabilities are suppressed by local endogenous factors and/or therapeutic FK 506 concentrations. These cells lacked cytolytic capabilities when the patient was free of rejection; however, when the patient rejected, donor-specific cytotoxic T cells were propagated. With improved clinical outcomes in human small intestine transplantation, more data can be accumulated to reveal the importance of the propagation of cytotoxic T cells from graft mucosal biopsies.

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