ELSEVIER

Serum T-Lymphocyte Cytokines Cannot Predict Early Acute Rejection in Renal Transplantation

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ABSTRACT

Despite numerous studies, the precise role of Th1/Th2 cytokines in acute renal allograft rejection remains unclear. To provide insight into the role of cytokines in acute allograft rejection, we measured serum T-cell cytokine concentrations for correlation with clinical events after renal transplantation in adults. Serum Th1 (interleukin-2 [IL-2] and interferon-gamma [IFNγ] and Th2 (IL-4, IL-10) cytokine concentrations were measured in 60 consecutive living donor kidney transplant recipients namely, 40 males, overall mean age 38.82 years), on the day before as well as 7 and 14 days posttransplantation using ELISA. Patients were stratified based upon acute rejection episode (ARE) in the first month after transplantation. Immunosuppression consisted of cyclosporine, mycophenolate mofetil, and prednisolone. ARE was diagnosed based on an increased plasma creatinine of more than 50%, sonographic analysis, radioisotope scan, pathologic findings, or measured cyclosporine blood levels. Twelve ARE were diagnosed among patients (20%). There was no significant difference between the 2 groups with respect to the mean serum concentration values of IL-2, IL-10, IL-4, and IFNγ on the day before or 7 or 14 days after transplantation. This study showed that there was no correlation between the Th1/Th2 serum cytokine profiles and early ARE in living donor kidney transplantation.

AN ACUTE REJECTION EPISODE (ARE) represents a serious morbidity among renal transplant recipients, as well as possibly an important factor for the development of chronic allograft nephropathy.[1] Nineteen years ago, Mosmann and Coffman described individual CD4+ T-cell clones in mice that produced distinct profiles of cytokines.[2] Observations that mouse and human helper T-cell clones fall into 2 main groups—TH1 and TH2—that synthesize either interleukin-2 (IL-2) and interferon-gamma (IFNγ) or IL-4, respectively,[3] have suggested that cytokine products regulate immune responses.[4] TH1 cytokines include IL-2, IL-12, TNF-β, and IFNγ, while TH2 cytokines include IL-4, IL-5, IL-6, IL-10, and IL-13.[5] In general, TH1-type cytokines favor the development of cellular immune responses and IgG2a antibody production, while TH2 cytokines favor humoral responses, especially IgE and IgG1 production.[5,6] The ability of TH1 and TH2 cells to stimulate different immune response is reinforced by the ability of some TH1 and TH2 cytokines to cross-regulate each other's development and function.[7,8] IFNγ and IL-12 decrease the levels of TH2 cytokines, while IL-4 and IL-10 decrease the levels of TH1 cytokines. Several groups have presented data that link the Th1 cytokine pattern to allograft rejection, and the Th2 cytokine pattern to transplant tolerance.[9–11] Cytokine profiles in recipient blood prior to transplantation have been suggested to be associated with transplant outcomes. However, various studies of cytokines in peripheral blood and urine from kidney transplant patients have yielded variable results.[12,13] In this study, we assessed pre- and posttransplantation serum cytokine levels among renal transplant patients who did versus did not develop ARE.

MATERIALS AND METHODS

The 60 candidates for living unrelated kidney transplantation between April 2003 and December 2003 included 40 males and an

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Table 1. Cytokine Concentrations on the Day Before Transplantation (Mean ± SD)

	Group 1	Group 2	*P*
IL-2 (pg/mL)	12.64 ± 4.11	15.86 ± 4.57	.382
IL-4 (pg/mL)	20.23 ± 6.63	19.35 ± 5.44	.431
IL-10 (pg/mL)	9.82 ± 3.32	14.64 ± 4.46	.664
IFNγ (pg/mL)	1.66 ± 0.87	2.14 ± 0.94	.722

overall mean age of 38.82 years. Serum samples (5 mL blood without anticoagulant) were collected 24 hours before as well as 7 and 14 days after transplantation. Each sample was tested for serum concentrations of IL-2, IFNγ, IL-4, and IL-10, using enzyme-linked immunosorbent assays (Bender Med kits, Germany). The immunosuppressive regimen for all 60 recipients included cyclosporine (6 mg/kg), prednisolone (1 mg/kg/d), and mycophenolate mofetil (1000 mg twice daily). Before transplantation, all patients were negative for panel-reactive antibodies with negative lymphocytotoxic crossmatches. HLA matching was not considered in this study. Twelve (20%) of the 60 recipients experienced an ARE within 20 days after transplantation. Each ARE was diagnosed based on an increased plasma creatinine by more than 50%, sonographic criteria, radioisotope scan, pathologic findings, or measured cyclosporine blood levels. Data were analyzed using the paired Student *t* test.

RESULTS

Tables 1, 2, and 3 and Figures 1 and 2 show serum levels of Th1 and Th2 cytokines in the 2 patient groups before and after renal transplantation. The mean concentration levels for the groups with (G1) and without (G2) ARE were: IL-2 pretransplantation 12.64 ± 4.11 vs 15.86 ± 4.57 pg/mL, respectively (P = .382); IL-2 at 7 days posttransplantation 11.23 ± 3.24 vs 14.66 ± 4.23 pg/mL, respectively (P = .651); IL-2 at 14 days posttransplantation 9.56 ± 2.32 vs 10.73 ± 3.14 pg/mL, respectively (P = .443); IL-4 pretransplanation 20.23 ± 6.63 vs 19.35 ± 5.44 pg/mL, respectively (P = .431); IL-4 at 7 days posttransplantation 24.54 ± 7.24 vs 27.74 ± 10.43 pg/mL, respectively (P = .462); IL-4 at 14 days posttransplantation 18.45 ± 5.13 vs 17.32 ± 4.32 pg/mL, respectively (P = .564); IL-10 pretransplantation 9.82 ± 3.32 vs 14.64 ± 4.46 pg/mL, respectively (P = .664); IL-10 at 7 days posttransplantation 7.55 ± 3.51 vs 9.61 ± 3.22 pg/mL, respectively (P = .654); IL-10 at 14 days postransplantation 10.23 ± 4.12 vs 12.21 ± 3.65 pg/mL, respectively (P = .436); IFNγ pretransplantation 1.66 ± 0.87 vs 2.14 ± 0.94 pg/mL, respectively (P = .722); IFNγ at 7 days posttransplantation 1.44 ± 0.53 vs 1.45 ± 0.24 pg/mL, respectively (P = .413); IFNγ at 14 days posttransplantation 1.27 ± 0.36 vs 1.63 ± 0.43 pg/mL, respectively (P = .322). These data showed that there was no correlation between Th1/Th2 serum cytokine profiles and early ARE among living donor kidney transplantations.

Table 2. Cytokine Concentrations at 7 Days Posttransplantation (Mean ± SD)

	Group 1	Group 2	*P*
IL-2 (pg/mL)	11.23 ± 3.24	14.66 ± 4.23	.651
IL-4 (pg/mL)	24.54 ± 7.24	27.74 ± 10.43	.462
IL-10 (pg/mL)	7.55 ± 3.51	9.61 ± 3.22	.654
IFNγ (pg/mL)	1.44 ± 0.53	1.45 ± 0.24	.413

Table 3. Cytokine Concentrations at 14 Days Posttransplantation (Mean ± SD)

	Group 1	Group 2	*P*
IL-2 (pg/mL)	9.56 ± 2.32	10.73 ± 3.14	.443
IL-4 (pg/mL)	18.45 ± 5.13	17.32 ± 4.32	.564
IL-10 (pg/mL)	10.23 ± 4.12	12.21 ± 3.65	.436
IFNγ (pg/mL)	1.27 ± 0.36	1.63 ± 0.43	.322

DISCUSSION

Polarized Th1 cells produce IL-2, IFN-γ, and lymphotoxin, which promote the development of cytotoxic T cells and support delayed-type hypersensitivity reactions (DTH) and facilitate antibody-dependent cellular cytotoxicity (ADCC), serving as effector mechanisms against intracellular pathogens and allotransplants. It has been suggested that tolerance to an allograft may be achieved by immune deviation of CD4 T cells to a polarized Th2 phenotype. If one assumes that a Th1-dominated immune response is responsible for allograft rejection, a pattern of Th1 cytokines should be detectable in the rejecting graft in the absence of typical Th2-associated cytokines. In this investigation, we evaluated serum levels of certain cytokines as known predictors of ARE. We categorized our 60 patients as those who had at least 1 ARE in the first month after transplantation (n = 12) versus no evidence of AR during this period (n = 48). Our data did not show a correlation between serum Th1/Th2 cytokine concentrations and ARE. This finding differs from other results. Sadeghi and coworkers[14] observed that the 38 patients who experienced biopsy-proven ARE displayed elevated IFNγ prior to transplantation. Another investigation among 14 kidney transplant patients by Rostaing and colleagues compared cytokine-expressing T-cell frequencies pretransplantation as well as at 3 and 6 months afterward. They observed that the group that developed ARE displayed a higher frequency of both IL-2 and IFNγ positive cells.[15] Amirzargar and coworkers[11] found that Th2 cytokines at 2 weeks posttransplantation were higher among the group without versus with ARE.

Other investigators did not observe an exact correlation between Th1/Th2 cytokines and ARE.[19] Krams et al[16] detected IL-4 in both rejecting and rejected renal allografts. Tai et al[17] found the up-regulation of both Th1 (IL-2, IFNγ) and Th2 (IL-4 and IL-10) cytokines in spontaneously accepted mouse liver allografts. Dallman et al[18] reported expression of IL-2, IL-4, and lymphotoxin in rejecting allografts. Our data suggested that the Th1/Th2 cytokine pattern in the early days following renal transplantation did not predict that the recipient showed stable renal function after transplantation or was prone to an ARE. So it seems likely that we cannot use them to predict ARE in renal transplantation.

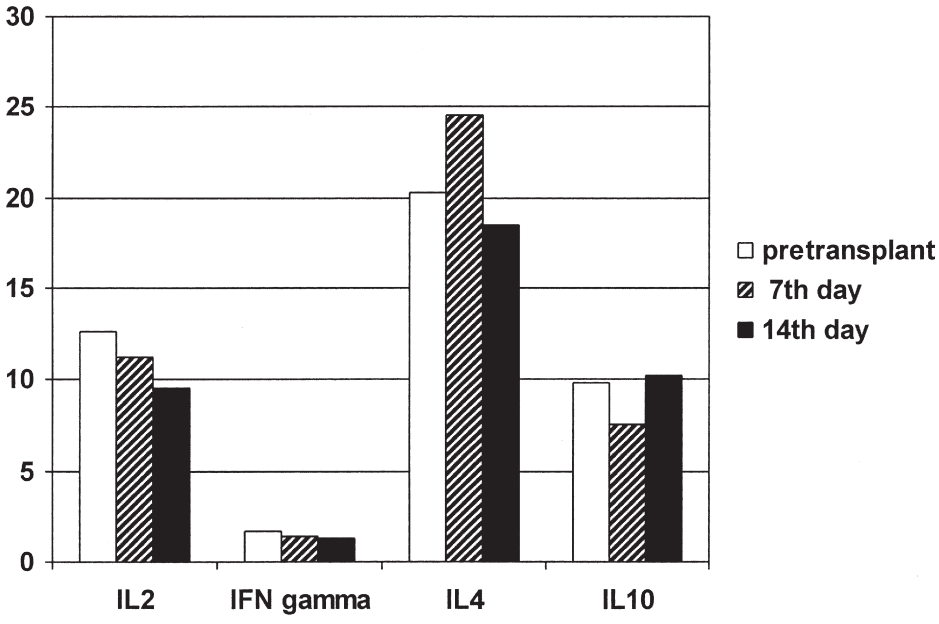

Fig 1. Serum cytokine levels (pg/mL) in patients with acute rejection.

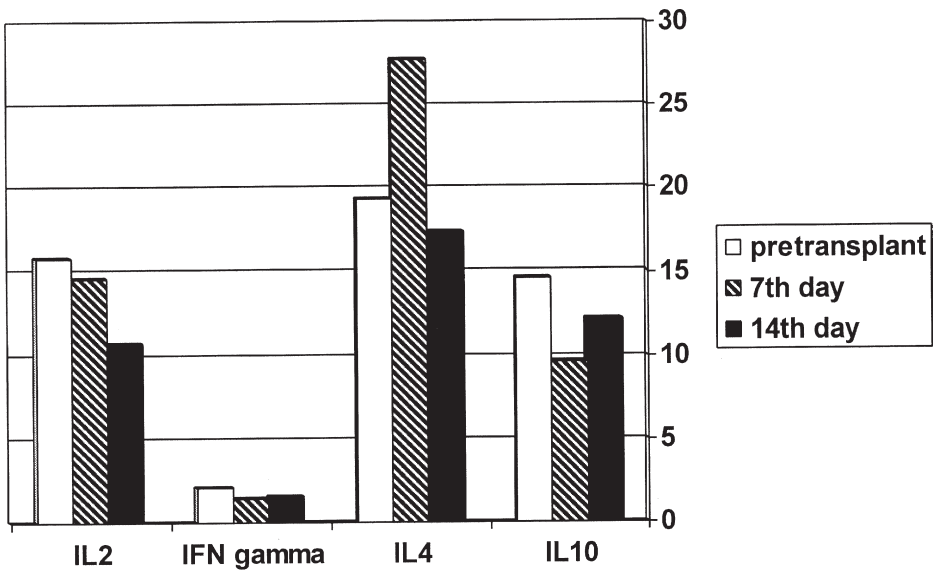

Fig. 2. Serum cytokine levels (pg/mL) in patients without acute rejection.

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