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Case Report

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Immune Tolerance in Transplant Kidney

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Abstract

Immune tolerance in transplantation is the successful preservation of the allograft throughout the recipient's lifetime. Cases with immunotolerance are of great importance in understanding the immune tolerance process and for possible innovations in the diagnosis and treatment processes. We aimed to present a patient who underwent renal transplantation from a cadaveric donor and revealed immunosuppressive withdrawal after two years of the transplantation. Cadaveric renal transplantation was performed on the 65-year-old patient after an 18-hour cold ischemia period. A 3/6 haplotype match and negative panel reactive antibody results were detected for both class I and class II antigens. The patient was discharged on post-operative day 13. Non-compliance was detected in the post-operative 29th month, and a renal biopsy was performed in the 56th month. Histological findings were nonspecific. The patient still lives with normal allograft functions. The absence of a clinical rejection in the follow-up of our case, which cannot be counted in the low-risk group in terms of rejection, reveals the importance of immune tolerance in maintaining allograft functions.

Keywords: Immune tolerance, renal transplantation, immunity

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INTRODUCTION

Immune tolerance in transplantation is the successful preservation of the allograft throughout the lifetime of the recipient. Achieving immune tolerance depends on the ability to control humoral responses in the recipient. This is mostly achieved by using immunosuppressive drugs lifelong. Non-adherence to treatment via discontinuation of immunosuppressive drugs in renal transplant recipients is an important cause of graft dysfunction. It had been detected that allograft functions can be maintained without immunosuppressive drugs in some of the renal transplant recipients (1-3). Many studies have been performed on the peripheral blood of immune tolerant cases in order to detect a non-invasive marker of immune tolerance. A biomarker that reflects immune tolerance can be used to evaluate the efficacy of immunosuppressive drugs, to highlight the mechanisms of immune tolerance in transplantation, and to detect the patients who can be followed up with or without reduced doses of immunosuppressive drugs (4). Although immune tolerance seemed to be a complicated process that involves both humoral and cellular immunity, the dominance of B lymphocytes was observed in several studies (1, 2). Immunosuppressive interleukins (IL) such as IL-10 and various mechanisms that interrupt the differentiation of B lymphocytes might play a simultaneous role in immune tolerance. In our case, we aimed to present a renal transplant recipient with immune tolerance after seven years of hemodialysis.

CASE PRESENTATION

A 65-year-old male patient was hospitalized in our transplantation clinic for cadaveric renal transplantation. The written informed consent was obtained from the patient before using the medical records in case report. It was learned from the patient's medical history that nephrec-

tomy was performed 10 years ago because of recurrent attacks of pyelonephritis due to urinary system stone disease. He was learned to be on hemodialysis three times per week for seven years before the renal transplantation. It was learned from the medical records that the patient was diagnosed with type-2 diabetes mellitus and essential hypertension 20 years ago. The patient was of 59 years of age at the time of transplantation while the donor was a 42-year-old male. Cadaveric renal transplantation was performed after an 18-hour cold ischemia period. The blood type of both the recipient and the donor was A Rh^+. 2 units of erythrocyte suspension were transfused into the recipient before the surgery. Body mass indexes were 24,3 and 29,7 for the recipient and the donor, respectively. A 3/6 haplotype match ($A_{24,}$ B_{18}, $DRB1_{11}$) and negative panel reactive antibody results for both class I and class II antigens were detected at the evaluation before the transplantation. Cadaveric renal transplantation was performed simultaneously with induction therapy with anti-thymocyte globulin (ATG). Prednisolone and mycophenolate mofetil were administered as well. Intra-operative renal biopsy revealed arteriolar hyalinosis (ah):1 and arterial intimal fibrosis (cv):0; but interstitial fibrosis and/or tubular atrophy (IFTA) was not detected. 2,700 cc urine output was reached at post-operative day one. In total, 5 dosages of ATG were given intermittently. Tacrolimus was added to the therapy on post-operative day eight. The patient was discharged with triple immunosuppressive therapy consisting of 20 mg daily prednisolone, 750 mg twice daily mycophenolate mofetil, and 3 mg daily tacrolimus on post-operative day 13. Plasma urea, creatinine, and tacrolimus levels were 88 mg/dL, 1.49 mg/dL, and 3.12, respectively, at discharge. The patient came to the control visits regularly for two years. Plasma tacrolimus levels were checked regularly at the control visits. Patient's non-compliance was detected at the post-operative 29th month. Serum tacrolimus level was found to be zero at the confirmation test. Patient did not come to the control visits for a while. At the post-operative 56th month, renal biopsy was performed. Plasma creatinine was 1.3 g/dL at the time of biopsy. 15 glomeruli with normal morphology were observed in the renal biopsy. In these glomeruli, inflammatory infiltration (g1); peritubular capillaries close to the cortex, medulla, interstitial inflammatory infiltration (i1); and a small number of tubular inflammatory infiltrate (t1) were detected. There was cv1 intimal fibrosis, but no IFTA was detected. Leukocyte common antigen (LCA) staining was positive, SV40 staining was negative, and C4d staining was positive in approximately 10% of glomerular capillaries and peritubular capillaries. Although histological findings were nonspecific, based on the features observed during immunohistochemistry tests, the possibility of subclinical rejection could not be ruled out and clinical follow-up was suggested. Panel reactive antibody (PRA) testing with Luminex method, which was performed simultaneously with biopsy, revealed negative results for class I and positive results for class II (MFI: 5000-10.000) (DQ_{5}, $DQ_{6,}$ DQ_{2}) antibodies. During the follow-up period, the patient's non-compliance persisted. Plasma levels of urea and creatinine were 19 mg/dL and 1.03 mg/dL, respectively, at post-operative 80th month, i.e., 24 months after the biopsy. The laboratory results at discharge (post-operative day 13), 29th month (time of drug withdrawal), 56th month (time of protocol biopsy), and 80th month (time of last control visit) are presented in Table 1. Another renal biopsy was planned in order to compare the findings with the ones from the biopsy performed at the 56th month, but the patient refused the procedure. As far as we could establish, the patient who did not use any immunosuppressive drugs and did not come to the control visits regularly is still living with normal allograft functions.

Main Points

- Immune tolerance in transplantation is the successful preservation of the allograft throughout the lifetime of the recipient. This is mostly achieved by using immunosuppressive drugs lifelong.
- Although non-adherence to treatment is an important cause of graft dysfunction, in immune tolerant cases graft survival can be obtained with unclear mechanisms despite the lack of immunosuppressive drugs.
- Immune tolerant cases must be followed up closely and protocol biopsies must be performed in order to contribute to the identification of immune tolerance mechanisms.

DISCUSSION

Spontaneous tolerance following renal transplantation was first established in 1975 (5). In the first case, it was considered that there was no need to restart immunosuppressive treatment unless rejection developed following discontinuation of immunosuppression. However, subsequent studies that evaluated greater number of cases emphasized the high rates of acute rejection and graft function losses (6). In our case, immunosuppressive drugs were used for two years after transplantation. The results of renal biopsy performed two years after the discontinuation of immunosuppressive drugs were interpreted as nonspecific but also compatible with subclinical rejection in the light of immunohistochemical findings. The absence of a clinical rejection in the follow-up of our patient, who could not be counted in the low-risk group in terms of rejection, reveals the effectivity of immune tolerance in terms of preservation of allograft functions. Currently, many immunosuppressive drugs are used simultaneously to preserve allograft function from recipients' immunity after renal transplantation. In order to achieve sufficient immunosuppression, plasma levels of immunosuppressive drugs must be checked and, if necessary, the dosages of immunosuppressive drugs must be arranged at control visits. Checking plasma levels of immunosuppressive drugs confirmed non-compliance to the treatment in our case. At the control visit, we found the plasma tacrolimus level to be zero. The absence of an approved measurement or biomarkers that can be used to establish donor-specific immune tolerance are major barriers to clinical trials. Two clinical trials, which evaluated the effects of discontinuation of calcineurin inhibitors in patients with low rejection risk were terminated early because of rejection attacks and/or donor-specific antibody (DSA) devel-

Table 1. Laboratory results of the recipient at discharge, post-operative 29th month, 56th month, and 80th month

Laboratory results	Discharge (Post-operative 13th day)	29th Month (Time of drug withdrawal)	56th Month (Time of protocol biopsy)	80th Month (Time of the last control visit)
Glucose (mg/dL)	101	184	235	125
Urea (mg/dL)	88	30	40	19
Creatinine (mg/dL)	1.49	1.05	1.1	1.03
Sodium (mmol/L)	139	136	136.8	129.2
Potassium (mmol/L)	5.4	3.5	4.5	4.4
Chloride (mmol/L)	101	108	110	93.5
Calcium (mg/dL)	8.7	9.5	9.9	9.1
Phosphorus (mg/dL)	4.4	3.7	3.8	2.93
Hemoglobin (g/dL)	10.8	15.1	13.8	12.8
Hematocrit (%)	33.3	42.2	39.6	35.3
Leukocyte (x10^3/mL)	11.6	6.8	7.7	6.21
Platelet (x10^3/mL)	195	147	213	143
Albumin (g/dL)	3.52	4.21	4.5	3.9
Spot urine protein/creatinine	0.242	0.152	0.151	0.16

opment after drug discontinuation (7, 8). Therefore, cases with immune tolerance are of great importance for understanding the immune tolerance process and for possible innovations in the diagnosis and treatment processes. Therefore, we thought that the immune tolerance of our case, which cannot be counted in the low-risk group in terms of rejection risk, is clinically important.

Although the mechanisms of immune tolerance have not been identified clearly, some clinical and gene expression differences have been identified in clinical studies. For this purpose, patients who were receiving immunosuppressive drugs and were clinically stable were compared with patients who were immune tolerant, and it was found that there is an increase in the expression of genes associated with B lymphocyte activation and differentiation in patients with immune tolerance (1, 2).

The roles of B lymphocytes in immune tolerance were obtained from the studies that compared patients receiving immunosuppressive drugs with patients with immune tolerance (9). It is, therefore, considered that these data may represent the effects of immunosuppression on B lymphocytes rather than being a specific marker of tolerance. Also, there was no consistent difference in gene profiles or cell frequencies in patients with immune tolerance compared with healthy control groups. Besides, immunosuppressive drugs were shown to be affective on gene profiles and cell subgroups in patients with immune tolerance. In a study, calcineurin inhibitors were found to reduce B cell count in kidney transplant recipients (10). In our case, patient received tacrolimus for two years after the transplantation. This may have contributed to the development of immune tolerance.

It is known that DSA can cause rejection at renal transplantation (11). Although class II antibodies were detected positive at the PRA examination, which was performed simultaneously with renal biopsy in our case, it was not clear whether the antibodies detected were donor specific, and the lack of rejection may be because of the non-specificity of the antibodies to donor.

In a case report, while immunosuppressive acting IL-10 levels were found to be increased; IFN-γ levels were found to be decreased for a patient with immune tolerance after kidney transplantation. B lymphocytes and natural killer (NK) cells are both thought to contribute to the maintenance of long-term graft function (12). In our case report, although we aimed to evaluate the interleukin levels simultaneously with biopsy parameters, we could not perform another biopsy or blood sampling because of the patient's refusal.

CONCLUSION

Although there is some evidence of immune tolerance at the cellular and gene expression levels, there is still no clear marker for routine clinical use. Large-scale clinical studies are needed to compare patients with immune tolerance and healthy control groups and, thereby, eliminate the effects of immune suppression. For this purpose, biopsy data of immune tolerant cases may provide valuable information. Immune tolerant cases

must be followed up closely and protocol biopsies must be performed in order to contribute to the identification of immune tolerance mechanisms.

Informed Consent: Written informed consent was obtained from the patient.

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