Influence of a low-dose tacrolimus protocol on the appearance of de novo donor-specific antibodies during 7 years of follow-up after renal transplantation

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ABSTRACT

Background. Tacrolimus (TAC) is a key immunosuppressant drug for kidney transplantation (KTx). However, the optimal serum trough level of TAC for good long-term outcomes remains unclear. This study aimed to investigate the relationship between the maintenance TAC trough level and the appearance of de novo donor-specific anti-human leukocyte antigen (HLA) antibodies (dnDSAs).

Methods. A total of 584 KTx recipients were enrolled in this study, of whom 164 developed dnDSAs during the follow-up period and 420 did not.

Results. We found no significant relationship between TAC trough level during the follow-up period and dnDSA incidence. Patients who developed dnDSAs had a significantly greater number of HLA-A/B/DR mismatches (3.4 ± 1.3 versus 2.8 ± 1.5; P < 0.001), were more likely to have preformed DSAs (48.2% versus 27.1%; P < 0.001) and showed poor allograft outcome.

Conclusions. There was no clear relationship between TAC trough level and dnDSA incidence for KTx recipients whose TAC trough levels were kept within the narrow range of 4–6 ng/mL during the immunosuppression maintenance period.

Keywords: clinical research/practice, de novo donor-specific anti-HLA antibodies, kidney transplantation/nephrology, rejection, tacrolimus

INTRODUCTION

With advances in immunosuppressive therapy and complication management, long-term graft survival after kidney transplantation (KTx) has improved in recent years. Therefore, lifelong immunosuppressive management is required to prevent allograft rejection and nonimmunological complications such as chronic allograft dysfunction, cardiovascular diseases, infectious diseases, malignancy, hypertension, dyslipidemia and diabetes mellitus [1, 2]. These nonimmunological complications are deeply related to long-term administered immunosuppressive medications such as tacrolimus (TAC) and steroids.

TAC is a current key drug mainly used as an immunosuppressant for KTx recipients [3]. In general, lower TAC concentration due to patient nonadherence or inadequate immunosuppression is one of the causes of acute rejection [4–6] through the development of de novo donor-specific anti-human leukocyte antigen (HLA) antibodies (dnDSAs) [7], especially during the early phase after transplantation, leading to deterioration of graft function.

On the other hand, high TAC trough levels during the maintenance period may cause progressing arteriolar hyalinosis, arteriosclerosis and interstitial fibrosis and tubular atrophy, likewise leading to the deterioration of graft function. Therefore the appropriate concentration of TAC for long-term use remains a subject of controversy.

At our institution, a TAC protocol using a lower trough concentration has been in use since the 2000s. The aim of this study is to investigate the relationship between maintenance trough levels of TAC and the rate of dnDSA appearance during long-term follow-up after KTx. We conducted a retrospective review and statistical analysis assessing the relationship between TAC trough concentration and dnDSA incidence over an average period of 7 years.

MATERIALS AND METHODS

Patient population

We evaluated a total of 994 patients who received a transplant at the Department of Urology of Tokyo Women’s Medical
University between 2000 and 2015 (Figure 1). Of these patients, 410 were excluded: pediatric recipients, deceased transplant recipients, extremely highly sensitized recipients such as complement-dependent cytotoxicity T-test positive and/or desensitization treatment-resistant (highly sensitized patients such as desensitization treatment–reactive patients were included [8]), recipients who did not undergo regular follow-up [regular renal biopsies and/or regular monitoring for DSAs by single-antigen bead assay (SABA)] and recipients who underwent follow-up at unknown hospitals. Thus 584 patients were included in this study.

Data were extracted from the Japan Academic Consortium of Kidney Transplantation study II [University Hospital Medical Information Network (UMIN) Clinical Trials Registry number: UMIN 000033449]. The study protocol was approved by the institutional research ethics committee (approval number 4460) and was consistent with the guidelines of the Declaration of Helsinki. All patients provided written informed consent.

Immunosuppressive regimen

Beginning a week before transplantation, all patients were treated following a triple immunosuppressive protocol including TAC (0.1 mg/kg/day), mycophenolate mofetil (MMF; 1500 mg/day if body weight was <50 kg, 2000 mg/day if ≥50 kg) and methylprednisolone (MP; 20 mg/day). Recipients whose transplants took place after 2002 also received basiliximab as an induction immunosuppression therapy, administered on the day of transplantation and on postoperative Day 4 [9]. Beginning in 2005, a single dose of rituximab (200 mg) was administered within 3–4 days before the operation if the transplant was ABO incompatible, the patient was highly sensitized or the etiology of end-stage renal disease (ESRD) was immunoglobulin A (IgA) nephropathy.

Plasma exchange (PE) was performed according to a previously reported protocol used for ABO-incompatible transplantation or before transplantation in patients with a history of sensitization [10]. This protocol involves a total of two or three sessions before surgery to reduce the anti-A/B antibody titer to ≥1:32 [11]. Some patients also received preoperative PE therapy to prevent recurrence of their original kidney disease, such as focal segmental glomerulosclerosis (FSGS). Intravenous immunoglobulin (IVIG) therapy was also performed for highly sensitized cases. Antithymocyte globulin was not used for induction in this study.

Patients received either a twice-daily dose of TAC (Prograf; Astellas Fujisawa, Osaka, Japan) or a once-daily dose of a prolonged-release formulation (Graceptor; Astellas Fujisawa). The dose was adjusted to maintain a trough level of TAC in whole blood of 8–12 ng/mL during the first month after KTx, 7–9 ng/mL during the second and third months and 4–6 ng/mL thereafter. The trough concentration of TAC was measured and analyzed before KTx; 2 weeks after KTx; 1, 3 and 6 months after and yearly from 1 to 10 years after. Routine TAC level measurements were performed in our institution by chemiluminescence immunoassay. When the results were 2 standard deviations (SDs) above or below normal, the assay was repeated and the average was taken as the value to be recorded. TAC levels given in this study are the average values for 3 months around each observation time.
The dose of MMF (Cellcept; Roche, Nutley, NJ, USA) was decreased to 1000–1500 mg/day ~2 weeks after KTx, then to 1000 mg/day ~1 month after KTx. The dose was also reduced in some patients who experienced side effects, such as diarrhea and viral infection. MP was administered intravenously at doses of 500 mg on the day of transplantation, 250 mg the day after the operation and 125 mg 2 days after the operation. MP was orally started 5 days after the operation at a dose of 20 mg/day; the dose was then tapered to 4 mg/day within 1 month after transplantation.

Graft function was evaluated according to estimated glomerular filtration rate (eGFR), calculated using the Modification of Diet in Renal Disease equation, which was measured and analyzed at our hospital before KTx; 2 weeks after KTx; 1, 3 and 6 months after and yearly from 1 to 10 years after.

Assessment of preformed DSAs and dnDSAs

Serum collected within 6 months before transplantation was analyzed by SABA (FlowPRA; One Lambda, Canoga Park, CA, USA) as previously reported [10]. Luminescence was read on a LABScan 100 Lumix system (One Lambda) and a mean fluorescence intensity >1000 was considered a positive result.

Polymerase chain reaction sequence-specific oligonucleotide technology (LABType XR; One Lambda) was used for HLA typing of recipients and donors (HLA-A, -B, -C, -DQ and -DR). Until 2016, routine HLA typing was performed only for HLA-A, HLA-B and HLA-DR; HLA-C and HLA-DQ were typed only if antibodies against them were detected by SABA, whereupon typing was performed to determine whether the antibodies were DSAs or non-DSAs. Patients with anti-HLA antibodies that recognized and cross-reacted with epitope groups specific to donor-mismatched HLAs were considered to have DSAs.

Serum analysis by SABA was again performed 6 months after KTx and then followed up annually. SABA was also performed when a patient underwent for-cause biopsy or when rejection was suspected on clinical evaluation.

During the follow-up period, dnDSAs sometimes disappeared and reappeared. Patients who tested positive for dnDSAs ever once during this period were included in the dnDSA group.

Diagnosis and treatment of rejection

Protocol allograft biopsies were performed within 6 months and around 1-year post-KTx and, if possible, annually thereafter. When rejection was suspected, an episode biopsy was performed. The type of rejection was classified as T-cell-mediated rejection (TCMR) or antibody-mediated rejection (ABMR) according to the Banff 2015 criteria. All biopsy specimens were evaluated using light microscopy and the specimens obtained were evaluated for C4d using immunofluorescence staining. Two or three core biopsy samples were obtained using a spring-loaded 16-gauge biopsy gun under ultrasound guidance. The diagnosis of rejection was made by pathologists from our institution's pathology department.

All patients showing TCMR or ABMR received bolus MP (500 mg) intravenously for 2 days. Patients diagnosed as having acute or chronic active ABMR were treated with PE and/or IVIG and administered rituximab. Patients diagnosed with acute or chronic active TCMR were treated with antithymocyte globulin.

Statistical analyses

Statistical analyses were performed with SAS version 9.4 (TSIM5; SAS Institute, Cary, NC, USA). One-way analysis of variance was used to compare normally distributed continuous variables, while the chi-square test was used to compare nominal variables; a two-tailed P-value <0.05 was considered statistically significant. A logistic regression analysis was also performed, with P-values <0.05 considered statistically significant.

RESULTS

Demographics and baseline characteristics

A total of 584 transplant recipients were enrolled in this study, of whom 164 developed dnDSAs during the follow-up period and 420 did not. Thereafter we divided the enrolled patients into two groups (Group 1, no dnDSA, n = 420; Group 2, dnDSAs, n = 164). Patient demographics are given in Table 1. Patients were followed up for an average of 7.4 ± 2.6 years, without significant differences between the two groups.

In Group 2, dnDSAs appeared at 812 ± 102 days on average after KTx (Figure 2). Class II dnDSAs are more common than Class I (140 cases [85.4%] and 50 cases [30.5%], respectively) (Table 1). There were no significant differences between the two groups with regard to sex, dialysis duration or renal function before KTx. With regard to the etiology of ESRD, the incidence of IgA nephropathy was significantly higher in Group 1 than in Group 2. There were no significant differences between the groups with regard to patients' medical history of KTx, pregnancy or blood transfusion.

Donor and transplant details are given in Table 2. There were no significant differences between the groups with regard to donor profiles (age, sex and blood relationship with recipients). The number of mismatches in HLA-A/B/DR was more significant in Group 2 than in Group 1 (3.4 ± 1.3 in Group 2 versus 2.8 ± 1.5 in Group 1; P < 0.001). Patients with preformed DSA also showed a higher incidence of dnDSA production (48.2% in Group 2 versus 27.1% in Group 1; P < 0.001). Details regarding preformed DSAs are given in Table 3; some patients had several types. There were no significant differences between the groups with regard to the immunosuppressant regimen used.

The

TAC dosage and trough level

There were no significant differences between the two groups with regard to mean TAC trough concentration during the follow-up period (Figure 3). Furthermore, there were no significant differences in the mean TAC dosage per kilogram during the follow-up period, except at 6 months and 1 year after KTx (Figure 4). For each group as a whole, the mean trough...
The concentration of TAC was within set target ranges throughout the study period. At every follow-up appointment in the outpatient clinic we monitored the TAC trough level and adjusted the dose of TAC to be administered until the next appointment. However, intrapatient variability in TAC exposure may occur as a result of nonadherence to treatment, abnormal absorption after gastrointestinal tract surgery or pharmacological interaction with other drugs, and certain genetic polymorphisms may also have an effect [12, 13]; accordingly, some patients showed TAC trough levels outside the target ranges [14]. The percentage of patients with trough levels outside the target ranges at each time point (2 weeks; 1, 3 and 6 months; 1, 2, 3, 4, 5, 6, 7, 8, 9 and 10 years after KTx) were 29.0, 37.8, 45.4, 27.7, 23.9, 24.1, 25.3, 28.1, 26.2, 28.1, 25.3, 27.3, 29.3 and 25.1% in Group 1 and 31.2, 37.4, 42.5, 29.6, 24.5, 29.5, 23.2, 19.2, 23.8, 24.5, 22.4, 21.2, 25.6 and 15.1% in Group 2, respectively. Thus TAC levels in the majority of patients were maintained within our specified target ranges.

Graft outcomes

Biopsy-proven rejection was observed in 158 patients (Table 4). The incidence of ABMR was more significant in Group 2 than in Group 1 [acute ABMR, 35/420 (8.3%) in Group 1 versus 48/164 (29.3%) in Group 2, P < 0.001; chronic ABMR, 22/420 (5.2%) in Group 1 versus 58/164 (35.4%) in Group 2, P < 0.001].

Regarding graft function, eGFR 1–4 years and 6–7 years after KTx was significantly worse in Group 2 than in Group 1 (Figure 5). There were no significant differences at 2 weeks, 1, 3 or 6 months or 5, 8, 9 or 10 years after KTx. The rate of death-censored graft failure was also significantly worse in Group 2 than Group 1 [18/420 (4.3%) in Group 1 versus 28/164 (17.1%) in Group 2; P < 0.001]. Patients in Group 2 also had a higher incidence of proteinuria when compared with patients in Group 1 (data not shown). There was no significant difference between groups in the rate of death with a functioning graft.

Relationship between dnDSA incidence and TAC trough level

The 584 patients were divided into three groups according to their average TAC trough concentrations during the study period.
maintenance period, calculated based on 11 time points (6 months after KTx and yearly from 1 to 10 years after KTx) as
follows: low, \(\leq 4\) ng/mL (\(n = 39\)); medium, 4–6 ng/mL (\(n = 488\)); and high, >6 ng/mL (\(n = 57\); Table 5). There were no significant differences among these groups with regard to dnDSA incidence, eGFR (Figure 6), donor sex and age, HLA-A/B/DR mismatches, preformed DSA, medical history or the incidence of biopsy-proven rejection. In all three groups, patients’ TAC trough concentrations stayed within the group range throughout the follow-up period (Figure 7).

Risk factors for dnDSA development
We performed a logistic regression analysis to determine the expected risk factors for development of dnDSAs. The average TAC trough concentration of each patient during the maintenance period was calculated based on 11 time points, as previously described. Preformed DSA (\(P = 0.0001\)) and HLA-A/B/DR mismatches (\(P = 0.0005\)) were found to be risk factors for the development of dnDSAs, but the average TAC trough concentration was not (\(P = 0.328\); Figure 8).

**DISCUSSION**
TAC as a calcineurin inhibitor is a key drug for mainstream immunosuppression following KTx [3], along with antimetabolite medicine and steroids. TAC contributes to the prevention of allograft rejection through suppression of T-cell activity. Thus the management of TAC concentration is important for successful immunosuppressive therapy both before and after KTx.

In general, patients who develop dnDSAs after KTx have a higher rate of rejection [15], decreased graft function and poorer graft survival rates than those who do not [16–22]. In some recent studies, lower TAC trough levels were associated with the production of dnDSAs [7], leading to ABMR and graft function deterioration [5]. However, these findings apply only to the early phases after KTx, e.g. within 5 years, and there are few reports dealing with the relationship between TAC trough concentration and long-term graft outcomes.

Higher TAC immunosuppression may be associated with complications such as chronic calcineurin inhibitor toxicity, including arteriosclerosis, hyalinosis and ischemic changes, leading to deterioration, infection, malignancy, hypertension, dyslipidemia, diabetes mellitus and other problems. These factors may affect graft and patient survival in the long term. Thus post-KTx management using a lower TAC level may be acceptable and

| Table 2. Donor and transplantation information |
|-----------------------------------------------|
| Variable | Total | Group 1, no dnDSA | Group 2, dnDSA | P-value |
| Donor information | | | | |
| Age at transplant (years), mean ± SD | 57.7 ± 9.4 | 57.8 ± 9.5 | 57.3 ± 9.4 | 0.592 |
| Donor sex, n (%) | | | | |
| Male | 185 (31.7) | 134 (31.9) | 51 (31.1) | 0.851 |
| Female | 399 (68.3) | 286 (68.1) | 113 (68.9) | |
| Donor type, n (%) | | | | |
| Father | 73 (12.5) | 54 (12.9) | 19 (11.6) | 0.177 |
| Mother | 193 (33.0) | 148 (35.2) | 45 (27.4) | |
| Sibling | 78 (13.4) | 58 (13.8) | 20 (12.2) | |
| Child | 5 (0.9) | 2 (0.5) | 3 (1.8) | |
| Spouse | 219 (37.5) | 147 (35.0) | 72 (43.9) | |
| Other blood relative | 8 (1.4) | 6 (1.4) | 2 (1.2) | |
| Non-blood relative | 8 (1.4) | 5 (1.2) | 3 (1.8) | |
| Transplantation information | | | | |
| Histocompatibility | | | | |
| ABO incompatibility, n (%) | 186 (31.8) | 134 (31.9) | 52 (31.7) | 0.963 |
| HLA-A/B/DR mismatches, mean ± SD | 2.9 ± 1.5 | 2.8 ± 1.5 | 3.4 ± 1.3 | <0.001 |
| Preformed DSA, n (%) | 193 (33.0) | 114 (27.1) | 79 (48.2) | <0.001 |
| Immunosuppressive therapy, n (%) | | | | |
| TAC | 584 (100) | 420 (100) | 164 (100) | |
| MMF | 553 (94.7) | 399 (95.0) | 154 (93.9) | 0.595 |
| MP | 584 (100) | 420 (100) | 164 (100) | |
| Basiliximab | 458 (78.4) | 324 (77.1) | 134 (81.7) | 0.228 |
| Rituximab | 350 (59.9) | 250 (59.5) | 100 (61.0) | 0.772 |
| DFPP/PE | 290 (49.7) | 205 (48.8) | 85 (51.8) | 0.528 |
| IVIG | 14 (2.4) | 8 (1.9) | 6 (3.7) | 0.247 |

Bold values are statistically significant at \(P < 0.05\).
For the donor type, ‘other blood relative’ represents, for example, aunts, cousins, nephews and nieces (no uncles in this study) and ‘non-blood relative’ represents, for example, fathers, mothers, brothers, sisters and sons-in-law. DFPP, double filtration plasmapheresis.

| Table 3. Preformed DSA information |
|-----------------------------------|
| Variable | Class 1 | Class 2 |
| Preformed DSA | 93 | 123 |
| (n = 193) | | |
| Type of HLA locus | A | B | Cw | DP | DQ | DR |
| 52 | 45 | 5 | 2 | 35 | 86 |
FIGURE 3: Mean TAC concentrations of each group in the different follow-up durations (mean ± SD).

FIGURE 4: Mean TAC doses of each group in the different follow-up durations (mean ± SD). *P < 0.05 calculated with Student’s t-test.

Table 4. Incidence rates of rejection and outcomes

| Variable                          | Total | Group 1, no dnDSA | Group 2, dnDSA | P-value |
|-----------------------------------|-------|-------------------|----------------|---------|
| Follow-up duration (years), mean ± SD | 584   | 420               | 164            | 0.569   |
| Biopsy-proven rejection, n (%)    |       |                   |                |         |
| Acute rejection                   | 158 (27.1) | 84 (20.0)       | 74 (45.1)     | <0.001  |
| A-TCMR                            | 104 (17.8) | 63 (15.0)       | 41 (25.0)     | 0.005   |
| A-ABMR                            | 83 (14.2)  | 35 (8.3)         | 48 (29.3)     | <0.001  |
| Chronic rejection                 | 89 (15.2)  | 30 (7.1)         | 59 (36.0)     | <0.001  |
| C-TCMR                            | 14 (2.4)   | 9 (2.1)          | 5 (3.0)       | 0.52    |
| C-ABMR                            | 80 (13.7)  | 22 (5.2)         | 58 (35.4)     | <0.001  |
| Outcomes, n (%)                   |       |                   |                |         |
| Death-censored graft failure      | 46 (7.9)   | 18 (4.3)         | 28 (17.1)     | <0.001  |
| Death with functioning graft      | 12 (2.1)   | 11 (2.6)         | 1 (0.6)       | 0.124   |

Bold values are statistically significant at P < 0.05.
A-TCMR, acute T-cell-mediated rejection; A-ABMR, acute antibody-mediated rejection; C-TCMR, chronic T-cell-mediated rejection; C-ABMR, chronic antibody-mediated rejection.
there may be many long-term advantages for post-KTx management as long as the patient does not develop dnDSAs.

At our institution, a protocol using a lower trough concentration of TAC has been in use since the 2000s. We investigated whether dnDSA incidence differs according to the recipient’s immunological sensitization, the immunosuppressive regimen used or the TAC trough level during maintenance. All the patients in this study received TAC according to the aforementioned protocol and their TAC levels were well managed within the set target ranges.

Previous studies have reported the prevalence of dnDSA development to be approximately 2–10% at 1 year post-KTx, reaching 10–40% by 4–5 years post-KTx [23–27]. In our study, dnDSAs were seen in 164 recipients (28.1%) over an average observation period of 7 years. Reports of the median onset of dnDSAs vary from 3.8 to 68 months after KTx [23, 27, 28]; in our study, dnDSAs appeared at 27.1 ± 2.27 months after KTx. In a previous study we reported the dnDSA incidence for ABO-compatible KTx recipients as 13% at 5 years after KTx and the time of onset

Table 5. Classifications according to average TAC concentration

| Variable                                      | Total       | ≤4          | 4–6         | >6          | P-value |
|-----------------------------------------------|-------------|-------------|-------------|-------------|---------|
| TAC concentration during the follow-up duration (ng/mL), mean ± SD | 584 ± 0.83  | 3.67 ± 0.29 | 4.97 ± 0.50 | 6.68 ± 0.91 | < 0.001 |
| dnDSA, n (%)                                  | 164 (28.1)  | 10 (25.6)   | 138 (28.3)  | 16 (28.1)   | 0.940   |
| Age at transplant (years), mean ± SD         | 45.2 ± 13.4 | 45.2 ± 14.0 | 45.2 ± 13.4 | 45.1 ± 13.6 | 0.982   |
| Recipient sex, n (%)                          | 379 (64.9)  | 26 (66.7)   | 311 (63.7)  | 42 (73.6)   | 0.321   |
| Male                                          | 205 (35.1)  | 13 (33.3)   | 177 (36.3)  | 15 (26.3)   |         |
| Female                                        | 193 (33.0)  | 14 (35.9)   | 159 (32.6)  | 20 (35.1)   | 0.862   |
| HLA-A/B/DR mismatches, mean ± SD             | 2.9 ± 1.5   | 2.7 ± 1.5   | 2.9 ± 1.5   | 2.8 ± 1.5   | 0.436   |
| Preformed DSA, n (%)                          | 107 (18.3)  | 7 (17.9)    | 93 (19.1)   | 7 (12.3)    | 0.458   |
| Age at transplant (years), mean ± SD         | 57.7 ± 9.4  | 58.5 ± 9.1  | 57.6 ± 9.5  | 57.6 ± 9.4  | 0.866   |
| Donor sex, n (%)                              | 185 (31.7)  | 13 (33.3)   | 155 (31.8)  | 17 (29.8)   | 0.818   |
| Male                                          | 399 (68.3)  | 26 (66.7)   | 333 (68.2)  | 40 (70.0)   |         |
| Female                                        | 158 (27.1)  | 11 (28.2)   | 129 (26.4)  | 18 (31.6)   | 0.701   |
| Acute rejection                               | 104 (17.8)  | 8 (20.5)    | 83 (17.0)   | 13 (22.8)   | 0.503   |
| A-TCMR                                        | 83 (14.2)   | 5 (12.8)    | 70 (14.3)   | 8 (14.0)    | 0.966   |
| A-ABMR                                        | 8 (15.2)    | 8 (15.4)    | 74 (15.2)   | 7 (12.3)    | 0.543   |
| Chronic rejection                             | 14 (2.4)    | 1 (2.6)     | 11 (22.5)   | 2 (3.5)     | 0.841   |
| C-TCMR                                        | 80 (13.2)   | 8 (15.4)    | 66 (13.5)   | 6 (10.5)    | 0.364   |
| C-ABMR                                        |               |             |             |             |         |

FIGURE 5: Mean eGFR of each group in the different follow-up durations (mean ± SD).

*P < 0.05, **P < 0.01 calculated with Student’s t-test.
as 46.5 months after KTx [29]. The reason for this discrepancy could be that in the previous study, antibodies against HLA-C and HLA-DQ were not counted as DSAs, thus dnDSA incidence could have been underestimated.

In our study, patients with dnDSAs had a much higher rate of ABMR (29.3% had acute ABMR, 35.4% had chronic ABMR) than those without. The prevalence of chronic ABMR was particularly high. A reason for this could be the long follow-up period, averaging 7 years. The presence of dnDSAs over a prolonged period is associated with chronic ABMR and deteriorated allograft function.

We found the number of mismatches in HLA-A/B/DR and the presence of preformed DSAs to be risk factors for dnDSA
formed DSAs are suggested as main risk factors for dn
Thus poor HLA histocompatibility and the existence of pre-
these cells react to the allograft, readily producing dn
somewhat efficacious in preventing
expression in order to prevent recurrence. This may have been
we usually administered rituximab as induction immunosup-
nificantly higher in Group 1 than in Group 2. For these patients,
the etiology of ESRD, the incidence of IgA nephropathy was sig-
deviation and/or progression to ABMR. Further analysis
KTx recipients still depends on preventing dnDSA develop-
through immunosuppressive therapy. Managing KTx recipients using lower TAC concentrations is thought to im-
and the existence of preformed DSAs. Thus poor HLA histocompatibility and the existence of pre-
formed DSAs are suggested as main risk factors for dnDSA produ-
duction that may lead to allograft deterioration. With regard to
the etiology of ESRD, the incidence of IgA nephropathy was sig-
nificantly higher in Group 1 than in Group 2. For these patients,
we usually administered rituximab as induction immunosuppres-
In conclusion, our institution has used an immunosuppres-
protocol with lower target TAC concentrations since the
2000s, with no significant differences in dnDSA incidence com-
pared with other institutions. There were no clear relationships
between patients who developed dnDSAs and those who did not.
Therefore management of KTx recipients at a lower TAC concentration appears not to be a main risk factor for develop-
ing dnDSAs and its advantages should be taken into account
with regard to long-term outcomes for patients and allografts.
However, as development of dnDSAs is closely associated with
allograft deterioration, immunosuppressive therapy that pre-
vents dnDSA development is still required. Thus it is important
to use appropriate immunosuppressive therapies that reduce
the risk of complications, including those caused by the immu-
nological sensitization of individual recipients.

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K.U., H.I., T.S. and M.F. participated in the research design,
writing the article, performance of the research and data analysis. K.K., T.H., Y.K., M.O., K.T. and K.N. contributed to
the design and editing of the article.

CONFLICT OF INTEREST STATEMENT
The authors of this manuscript have no conflicts of interest to disclose as described by the Nephrology Dialysis Transplantation.

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