Dysregulation of Interleukin 23 Receptor Expression in Kidney Allografts Associated with Composite Outcome

Kuang-Chin Hsiao1,2, Wan-Ru Chao2,3, Jen-Pi Tsai4, Mei-Chin Wen5, Jong-Da Lian5, Wen-Chin Lee1, Jong-Yu Huang1, Shun-Chi Chang1 and Horng-Rong Chang2,6

1Division of Nephrology, Department of Medicine, Show Chwan Memorial Hospital, Changhua, Taiwan
2Institute of Medicine, Chung Shan Medical University, Taichung, Taiwan
3Department of Pathology, Chung Shan Medical University, Taichung, Taiwan
4Department of Nephrology, Buddhist Dalin Tzu Chi General Hospital, Chiayi, Taiwan
5Department of Pathology, Taichung Veterans General Hospital, Taichung, Taiwan
6Division of Nephrology, Department of Medicine, Chung Shan Medical University Hospital, Taichung, Taiwan

Abstract

**Background:** Interleukin 23 (IL-23) and interleukin 23 receptor (IL-23R) play a role in the pathogenesis of multiple autoimmune processes and renal inflammation, but research has yet to clarify the histological association of IL-23/IL-23R and transplant kidney allografts.

**Methods:** Between July 2009 and August 2011, 31 renal transplant recipients who received sonography-guided kidney allograft biopsy were enrolled in this retrospective study. The patients were divided into two groups including group A (patients reaching composite outcome) and group B (patients not reaching composite outcome). The composite outcome was defined as serum creatinine (Scr) doubling and lower estimated glomerular filtration rate (eGFR). Specimens of 31 patients were examined by the immunohistochemical stain of IL-23 and IL-23R in allograft kidneys, and clinico-pathological associations were evaluated.

**Results:** Of the 31 patients, group A had 15 patients (48.3%) and group B had 16 patients (52.7%). Group A had significantly higher Scr, lower eGFR, and low serum albumin (p=0.024). Univariate analysis showed that group A was negatively associated with atrophic glomerular mesangial cell cytoplasmic IL-23R expression (p=0.044). The decreased expression of IL-23R could be due to higher acute antibody-mediated rejection with heavy proteinuria in our study. In other words, the more the glomerular damage due to antibody-mediated rejection, the less the expression of IL-23R in atrophic glomerular mesangial cell cytoplasm.

**Conclusions:** The patients with composite outcome may have decreased expression of IL-23R in atrophic glomerular mesangial cell cytoplasm.

Keywords: IL-23; IL-23 receptor; Immunohistochemistry stain; Renal transplantation

Introduction

A kidney transplant is the best choice for the treatment of end stage kidney disease. Rejection of transplanted tissues involves the interplay between mechanisms that maintain tolerance to the graft and factors that promote rejection. Acute rejection continues to be one of the most important causes of graft loss and involves the cellular and/or humoral immune response [1]. Cellular rejection is characterized morphologically by the presence of mononuclear cells in the interstitial, tubular, and glomerular compartments [1-3]. Moreover, humoral rejection is associated with vascular involvement (vasculitis), deposition of immunoglobulins (C4d deposition in peritubular capillaries), and activation of the complement cascade [4-6].

Patients with acute allograft rejection present with an acute rise in the Scr and lower eGFR. A rising Scr level, however, is a relatively late development in the course of a rejection episode and usually indicates the presence of significant histological damage [1]. New or worsening proteinuria may also be present with/without decreased serum albumin level, especially in the case of acute humoral rejection [6].

T lymphocytes not only play an essential role in the initiation of the cascade of mechanisms underlying rejection but also participate in mechanisms that maintain graft tolerance [7,8]. Naïve CD4+ helper T cells have been shown to develop into at least 4 types of committed helper T cells, namely, T helper (Th) 1, Th2, Th17, and regulatory T cells [3,5,9]. Interleukin 23 (IL-23) affects interferon-γ production (IFN-γ) by T and natural killer cells, activates memory T cells, stimulates Th1 cell responses, and enhances inflammation by stimulating the production of proinflammatory cytokines [10,11].

IL-23 mediates these effects through binding a receptor composed of IL-12Rβ1 and interleukin 23 receptor (IL-23R), with the latter being located on chromosome 1p31.3. IL-23R, which is the initial sensor of the IL-23 signal, also determines Th17 cell expansion and in turn serves
as an important gate for Th17 cell mediated autoimmune responses [12]. An accumulating body of literature reports that the presence of IL-23R gene polymorphism is associated with various autoimmune diseases such as rheumatoid arthritis, Crohn’s disease, Grave’s ophthalmopathy and graft-versus-host [13-16]. Recently, Tsai et al. [17] revealed an association between interleukin 23 receptor polymorphism and kidney transplant outcomes.

Based on the aforementioned background research, we hypothesize that IL-23 and IL-23R are related to the immuno-modulatory functions of transplanted kidneys. The aim of this study is thus to examine the clieno-pathological correlation of expressions of IL-23/IL-23R in kidney transplant allografts.

Materials and Methods

Study design and patients

From July 2009 to August 2011, pathological specimens from 31 renal transplantation recipients who received sonography-guided kidney allograft biopsy were retrospectively recruited. Institutional review board approval of Chung Shan Medical University Hospital was obtained for the review of patients’ medical records, data analysis and pathological specimens staining, and the need for informed consent was waived.

Patient age, gender, body mass index, status of cigarette smoking, hypertension, diabetic mellitus, hepatitis B, hepatitis C, blood pressure, dialysis mode before transplantation, immusupressant drug (tacrolimus and cyclosporin) and drug trough level were recorded. Labotory data including SCr, eGFR, hemoglobin, hemoglobin A1c (HBA1C), lipid titer, albumin, uric acid and dipstick urine protein. eGFR was calculated by the abbreviated Modification of Diet in Renal Disease formula (aMDRD): eGFR=186 x (serum Creatinine)-1:154 x (age)-0:203 x (0.742 if female). Chronic kidney disease was defined by K/DOQI guidelines [18].

Primary composite outcome

The primary outcome measured in this study was a composite endpoint of time to first event with a doubling of SCr or declining of eGFR of more than 30%. The composite group (group A) was defined as reaching the doubling value of baseline SCr or declining of baseline eGFR of more than 30%. The group B was defined as not reach composite outcome. A baseline SCr/eGFR was defined as the best stable level of SCr/eGFR within six months between allograft biopsy, and terminal SCr/eGFR was defined as the following of SCr/eGFR to decline in eGFR as an endpoint for kidney failure [21].

Clinical pathological diagnosis

The clinical pathological diagnosis was acute tubular injury (n=5), mild acute cellular rejection (n=8), severe acute cellular rejection (n=13) and antibody mediated rejection (ABMR) (n=5). The severe acute cellular rejection was defined as Banff type acute cellular rejection IA, IB and IIA [22]. If there were more than two pathological diagnoses of ex calcineurin inhibitor nephrotoxicity, chronic fibrosis change, de novo glomerulonephritis, diabetic nephropathy or polyomavirus nephropathy in one renal specimen, we chose the major pathological result as the pathological diagnosis in this study.

Tissue processing

Pathologic material was processed by conventional histological procedures. The specimens were collected by sonography-guided kidney allograft biopsy. Each section was at least 2×0.5 cm2. The formalin-fixed, paraffin-embedded tissues were cut into 4-mm hematoxylin- and eosin-stained (H&E staining) sections and examined to evaluate the glomerular, renal tubular, and interstitial conditions. The scoring of fibrosis was based on Banff scoring for chronic lesions [22] with interstitial fibrosis score and glomerular fibrosis score.

Each patient who received kidney allograft biopsy was received medical therapy according to the pathological diagnosis. When the renal insufficiency recovered, the patient was discharged with an outpatient department follow up. The remaining biopsy tissue was stained by innumohistochemical (IHC) of IL-23 and IL-23R. The study protocol was approved by the local Ethics Committee.

Immunohistochemical staining

Paraffin embedded kidney tissue sections (4-mm) on poly-l-lysine-coated slides were deparaffinized. After treatment with 3% H2O2 in methanol, the sections were hydrated with gradient alcohol and PBS, incubated in 10 mM citrate buffer, and finally heated at 100 uC for 20 min in PBS. Slides were incubated with the anti-IL-23 and IL-23R antibody (Santa Cruz, CA, USA) for 20 min at room temperature, and then with a horseradish peroxidase (HRP)/Fab polymer conjugate for another 30 min. Then, slides were thoroughly washed three times with PBS, and the sites of peroxidase activity were visualized using 3, 3-diamino-benzidine tetrahydrochloride as a substrate and hematoxylin as the counter stain.

Semi-quantitative grading

All IHC stain data were independently scored by two blinded pathologists using the following scale: 0=no staining, 1=mild staining, 2=moderate staining, 3=high staining (Figure 1). Every slide was examined entirely for nuclear and cytoplasmatic IL-23/IL-23R stains in the normal and atrophic renal tubules, in the normal and atrophic glomeruli, and in the renal interstitium. Each 2×0.5 cm2 section contained at least 10 glomerular areas, and the actual number of examined glomeruli was based on the sectioned tissue size. The results of nuclear and cytoplasmatic staining were recorded separately. The intensity of IL-23 and IL-23R staining was classified as with staining (1, 2 and 3) or without staining (0).

Statistical analysis

Continuous and categorical data were expressed as median (25%-75%) and as proportions, respectively. Categorical variables were analyzed by the chi-square test or Fisher’s exact test. Spearman’s rank correlation coefficient was used for association between clinical variables when reaching composite outcome. A p-value less than 0.05 was considered statistically significant. All data were analyzed using SPSS version 14.0 statistical software.
Results

We classified the 31 patients divided into group A (n=15) and group B (n=16). There were significant differences in the eGFR (terminal), SCR (terminal) and albumin level between those two groups. Our results indicate that group A had low terminal eGFR (15.6 mL/min for group A, 58.5 mL/min for group B, p<0.001), high terminal SCR (3.3 mg/dl for group A, 1.2 mg/dl for group B, p<0.001) and lower albumin levels (3.5 g/dl for group A, 3.8 g/dl for group B, p=0.025) (Table 1).

| Patients reaching composite outcome (group A) | Patients not reaching composite outcome (group B) | P value |
|-----------------------------------------------|-----------------------------------------------|---------|
| median                                       | median                                       | 25%-75% |
| Patient number (n)                           | 15                                           | 16      |
the intensity of IL-23/IL-23R expression in each component of the
and interstitium.

We compared the association between group A and group B with
the intensity of IL-23/IL-23R expression in each component of
the specimen in Table 2. However, there was no relationship found
for the different regions of renal tissues including the glomerular, tubular
and interstitial.

### Table 1: Demographic and clinical characteristics of patients divided
by patients reaching composite outcome (left) and patients not
reaching composite outcome (right).

| Score          | Patients reaching composite outcome | Patients not reaching composite outcome | P value |
|---------------|------------------------------------|----------------------------------------|--------|
| IFS           | Without                            | 5 (33.3)                               | 9 (60) | 0.143 |
|               | With                               | 10 (66.7)                              | 6 (40) |        |
| GFS           | Without                            | 12 (80)                                | 15 (100)| 0.224 |
|               | With                               | 3 (20)                                 | 0      |        |
| IL23 (Gn)n    | Without                            | 6 (54.5)                               | 9 (81.8)| 0.361 |
|               | With                               | 5 (45.5)                               | 2 (18.2)|        |
| IL23 (Gn)c    | Without                            | 9 (81.8)                               | 5 (45.5)| 0.183 |
|               | With                               | 2 (18.2)                               | 6 (54.5)|        |
| IL23 (Ga)n    | Without                            | 12 (82.3)                              | 6 (100)| 1      |
|               | With                               | 1 (7.7)                                | 0      |        |
| IL23 (Ga)c    | Without                            | 10 (76.9)                              | 6 (100)| 0.517 |
|               | With                               | 3 (23.1)                               | 0      |        |
| IL23 (ATn)n   | Without                            | 15 (100)                               | 15 (93.8)| 1    |

Composite outcome: first event with doubling of SCr or declining eGFR more
than 30%. HTN, hypertension; DM, diabetes mellitus; HBV, hepatitis B; HCV,
hepatitis C; BMI, body mass index; HD, hemodialysis; PD, peritoneal dialysis;
SBP, systolic blood pressure; DBP, diastolic blood pressure; eGFR, estimated
glomerular filtration; SCr: serum creatinine; HbA1C: hemoglobin A1c.

* p<0.05 indicates significance.

### Table 2: Intensity of IL-23/IL-23R receptor expression in different
regions of renal tissues divided by patient reach composite outcome
(left) and patient did not reach composite outcome (right).

| Patients reaching composite outcome | Patients not reaching composite outcome | P value |
|------------------------------------|----------------------------------------|--------|
| With                              | Without                                | 0      | 1 (6.3) | |
| With                              | Without                                | 15 (100) | 15 (93.8)| 1 |
| With                              | Without                                | 7 (100)  | 2 (66.7) | 0.172 |
| With                              | Without                                | 14 (93.3)| 11 (68.8)| 0.172 |
| With                              | Without                                | 1 (6.7)  | 5 (31.3) | |
| With                              | Without                                | 4 (44.4)| 3 (30)  | 0.65 |
| With                              | Without                                | 5 (55.6)| 7 (70)  | |
| With                              | Without                                | 8 (88.9) | 8 (80)  | 1 |
| With                              | Without                                | 9 (69.2)| 5 (71.4)| 1 |
| With                              | Without                                | 4 (30.8)| 2 (28.6)| |
| With                              | Without                                | 13 (100)| 5 (71.4)| 0.111 |
| With                              | Without                                | 0       | 2 (28.6)| |
| With                              | Without                                | 9 (60)  | 8 (50)  | 0.576 |
| With                              | Without                                | 6 (40)  | 8 (50)  | |
| With                              | Without                                | 2 (13.3)| 3 (18.8)| 1 |
| With                              | Without                                | 13 (86.7)| 13 (81.3)| |
| With                              | Without                                | 3 (50)  | 3 (75)  | 0.571 |
| With                              | Without                                | 3 (50)  | 1 (25)  | |
| With                              | Without                                | 1 (16.7)| 1 (25)  | 1 |
| With                              | Without                                | 5 (83.3)| 3 (75)  | |
| With                              | Without                                | 10 (86.7)| 13 (81.3)| 0.433 |
| With                              | Without                                | 5 (33.3)| 3 (18.8)| |
| With                              | Without                                | 5 (33.3)| 7 (43.8)| |

Composite outcome: first event with doubling of SCr or declining eGFR more
than 30%. IFS, interstitial fibrosis score; GFS, glomerular fibrosis score; (Gn)n,
nuclear staining intensity of normal glomerulus mesangial cell; (Gn)c,
cytoplasmic staining intensity of normal glomerulus mesangial cell; (Ga)n,
nuclear staining intensity of atrophy glomerulus mesangial cell; (Ga)c,
cytoplasmic staining intensity of atrophy glomerulus mesangial cell; (ATn)n,
nuclear staining intensity of normal renal tubule; (ATn)c, cytoplasmic staining
intensity of normal renal tubule; (ATc)n, nuclear staining intensity of
interstitium; (IT)c, cytoplasmic staining intensity of interstitium.

p<0.05 indicates significance.
associated with eGFR (terminal) \((r=0.78, p<0.001)\), positively associated with SCr (terminal) \((r=0.784, p<0.001)\), and negatively associated with albumin level \((r=0.427; p=0.024)\) (Table 3).

| R     | P value |
|-------|---------|
| Patient number (n) | 0.032 | 0.862 |
| Age (year) | -0.222 | 0.905 |
| Gender F/M (n) | 0.128 | 0.491 |
| HTN (n, %) | 0.081 | 0.666 |
| DM (n, %) | -0.099 | 0.598 |
| HBV (n, %) | 0.314 | 0.085 |
| HCV (n, %) | -0.38 | 0.839 |
| Smoke (n, %) | 0.177 | 0.341 |
| BMI (kg/m2) | 0.016 | 0.933 |
| HD/PD (n) | -0.23 | 0.213 |
| Pathology diagnosis | 0.281 | 0.125 |
| Drug | 0.18 | 0.332 |
| SBP (mmHg) | -0.166 | 0.371 |
| DBP (mmHg) | -0.296 | 0.106 |
| eGFR (biopsy) (ml/min) | -0.238 | 0.196 |
| eGFR (baseline) (ml/min) | -0.307 | 0.093 |
| eGRF (terminal) (ml/min) | -0.78 | <0.001* |
| SCr (biopsy) (mg/dl) | 0.232 | 0.21 |
| SCr (baseline) (mg/dl) | 0.342 | 0.06 |
| SCr (terminal) (mg/dl) | 0.784 | <0.001* |
| Hemoglobin (g/dl) | 0.029 | 0.877 |
| HbA1C (%) | 0.025 | 0.898 |
| Low-density lipoprotein (mg/dl) | 0.078 | 0.686 |
| Total cholesterol (mg/dl) | -0.079 | 0.671 |
| Triglyceride (mg/dl) | -0.184 | 0.322 |
| Albumin (mg/dl) | -0.427 | 0.024 |
| Uric acid (mg/dl) | 0.091 | 0.638 |
| Urine dipstick protein (mg/dL) | 0.309 | 0.09 |

Composite outcome: first event with doubling of SCr or declining eGFR more than 30%. HTN, hypertension; DM, diabetes mellitus; HBV, hepatitis B; HCV, hepatitis C; BMI, body mass index; HD, hemodialysis; PD, peritoneal dialysis; SBP, systolic blood pressure; DBP, diastolic blood pressure; eGFR, estimated glomerular filtration; SCr, serum creatinine; HbA1C, hemoglobin A1c.

Table 4 shows the Spearman's correlation between pathological variables and primary composite outcome. Univariate analysis indicated that IHC IL-23R expression intensity stain was negatively associated with atrophic glomerular mesangial cell cytoplasm \((r=0.454, p=0.044)\).

| R     | P value |
|-------|---------|
| IFS | 0.267 | 0.153 |
| GFS | 0.333 | 0.072 |
| IL23 (Gn)n | 0.293 | 0.186 |
| IL23 (Gn)c | -0.378 | 0.083 |
| IL23 (Ga)n | 0.16 | 0.513 |
| IL23 (Ga)c | 0.294 | 0.222 |
| IL23 (ATn)n | -0.177 | 0.341 |
| IL23 (ATn)c | 0.177 | 0.341 |
| IL23 (ATa)c | 0.509 | 0.133 |
| IL23 (IT)c | -0.311 | 0.089 |
| IL23R (Gn)n | -0.15 | 0.541 |
| IL23R (Gn)c | -0.122 | 0.62 |
| IL23R (Ga)n | 0.023 | 0.924 |
| IL23R (Ga)c | -0.454 | 0.044* |
| IL23R (ATn)c | 0.074 | 0.694 |
| IL23R (ATa)n | 0.25 | 0.486 |
| IL23R (ATa)c | 0.102 | 0.779 |
| IL23R (IT)n | 0.167 | 0.371 |
| IL23R (IT)c | -0.107 | 0.567 |

Composite outcome: first event with doubling of SCr or declining eGFR more than 30%. IFS, interstitial fibrosis score; GFS, glomerular fibrosis score; (Gn)n, nuclear staining intensity of normal glomerulus mesangial cell; (Gn)c, cytoplasmic staining intensity of normal glomerulus mesangial cell; (Ga)n, nuclear staining intensity of atrophy glomerulus mesangial cell; (Ga)c, cytoplasmic staining intensity of atrophy glomerulus mesangial cell; (ATn)n, nuclear staining intensity of normal renal tubule; (ATn)c, cytoplasmic staining intensity of normal renal tubule; (ATa)n, nuclear staining intensity of atrophy renal tubule; (ATa)c, cytoplasmic staining intensity of atrophy renal tubule; (IT)n, nuclear staining intensity of interstitium; (IT)c, cytoplasmic staining intensity of interstitium.

*: p<0.05 indicates significance

Table 4: Spearman's correlation association about pathological variables when reaching composite outcome.

Figure 2 illustrates the different expression intensities of H&E stain (panel A, C, E) and IL-23 IHC stain (panel B, D, F) in patients reaching composite outcome; H&E and IL-23 IHC stain in patients not reaching composite outcome is shown in Figure 3. Finally, a decreased intensity of IL-23R expression in atrophic glomerular mesangial cell cytoplasm in group A (panel A) can be seen in Figure 4.
Discussion

Our results demonstrate that patients reaching the composite outcome were negatively associated with atrophic glomerular mesangial cell cytoplasmic IL-23R expression (p=0.044). However, IL-23 expression was not associated with the outcome in any of the renal tissue, including the glomerular, tubule and interstitium.

In the clinical variable analysis, the group A was negatively associated with terminal eGFR (r=0.78, p<0.001), positively associated with terminal SCR (r=0.784, p<0.001), and negatively associated with albumin level (r=0.427; p=0.024). Because the definition of composite outcome was doubling volume of SCR or a declining baseline eGFR of more than 30% in our study, the results of terminal Scr and eGFR could be due to the definition. Furthermore, compared with group B, group A had more ABMR (26.7% vs. 6.3%) in the pathological diagnosis and higher urine dipstick protein (>100mg/dL, 20% vs. 6.25%). Given that AMBR usually presents with transplant glomerulopathy and proteinuria [22,23], the lower albumin level in group A (3.5 mg/dl vs. 3.8 md/dl, p=0.025) could be caused by a higher ABMR with transplant glomerulopathy and high urine dipstick protein in our study.

There have been several reports about interlukin marker expression in acute allograft rejection. Byung at al. revealed that higher infiltration by Th17 cells is associated with severe acute T-cell-mediated graft rejection. Higher infiltration of Th17 is significantly associated with the severity of allograft dysfunction and tissue injury [24]. IL-17 expression by tubular epithelial cells in renal transplant recipients with acute antibody-mediated rejection has also been observed, with IL-17 tubular expression being directly and significantly correlated with the extension of C4d deposits [25].
Although interleukin expression was found to be approximately expressed in the renal tubule and interstitium in previous research, several studies have revealed the correction of IL-23 and glomerular disease. Paust et al. reported that the IL-23/Th17 axis contributes to renal injury in experimental glomerulonephritis, with IL-17 enhancing the production of the proinflammatory chemokines CCL2/MCP-1, CCL3/MIP-1, and CCL20/LARC in mouse mesangial cells. They further found that IL-23 p19/- mice developed less severe nephritis as measured by renal function, albuminuria, and frequency of glomerular crescent formation [26]. IL-23 receptor expression has also been shown to be up-regulated in lupus nephritis; the greater the increase in serum level of IL-23R, the greater the glomerular damage of lupus nephritis in mice [27].

In our study, the patients reaching the composite outcome had a decline of IL-23R expression in atrophic glomerular mesangial cell cytoplasm. ABMR was more evident with transplant glomerulopathy. In other words, the more the serum level of IL-23R, the greater the glomerular damage of lupus nephritis in mice [27].

**Figure 4**: Representative panels showing a decreased intensity of IHC IL-23R expression in atrophic glomerular mesangial cell cytoplasm in a patient reaching composite outcome (group A) (IHC stain, x400). (A) IHC IL-23R stain in a patient reaching composite outcome (group A), (B) IHC IL-23R stain in a patient not reaching composite outcome (group B), (C) positive control of IL-23R IHC stain, (D) negative control of IL-23R IHC stain. The arrow indicates the IL-23R IHC stain in an atrophic glomerular mesangial cell.

In summary, our analysis of the expression of IL-23R in allograft specimens indicates a possible role of IL-23R over mesangial cytoplasm of atrophic glomerular cells. However, because most of the pathologic changes of acute allograft rejection were involved in the renal interstitium, the finding of IL-23R expression over glomeruli needs further study for clarification.

**References**

1. Meier-Kriesche HU, Schold JD, Srinivas TR, Kaplan B (2004) Lack of improvement in renal allograft survival despite a marked decrease in acute rejection rates over the most recent era. Am J Transplant 4: 378-383.
2. Trpikov K, Campbell P, Pazderka F, Cockfeld S, Srole K, et al. (1996) Pathologic features of acute renal allograft rejection associated with donor-specific antibody. Analysis using the Banff grading schema. Transplantation 61: 1586-1592.
3. Afzali B, Lombardi G, Lechler RI, Lord GM (2007) The role of T helper 17 (Th17) and regulatory T cells (Treg) in human organ transplantation and autoimmune disease. Clin Exp Immunol 148: 32-46.
4. Colvin RB, Cohen AH, Saisonitz C, Bonsib S, Buick M, et al. (1997) Evaluation of pathologic criteria for acute renal allograft rejection: reproducibility, sensitivity, and clinical correlation. Journal of the American Society of Nephrology 8: 1930-1941.
5. Aki A, Luo S, Wood KJ (2005) Induction of transplantation tolerance-the potential of regulatory T cells. Transpl Immunol 14: 225-230.
6. Mauiyed S, Crespo M, Collins AB, Schneeberger EE, Pascual MA, et al. (2002) Acute humoral rejection in kidney transplantation: IL-12 family of cytokines and autoimmune disease. J Am Soc Nephrol 13: 779-787.
7. Feucht HE, Schneeberger H, Hillebrand G, Burkhardt K, Weiss M, et al. (1993) Capillary deposition of C4d complement fragment and early renal graft loss. Kidney international 45: 1333-1338.
8. Collins AB, Schneeberger EE, Pascual MA, Saidman SL, Williams WW, et al. (1999) Complement activation in acute humoral renal allograft rejection: diagnostic significance of C4d deposits in peritubular capillaries. J Am Soc Nephrol 10: 2208-2214.
9. Mosmann TR, Cherwinski H, Bond MW, Giedlin MA, Coffman RL (1986) Two types of murine helper T cell clone. I. Definition according to profiles of lymphokine activities and secreted proteins. Journal of immunology 175: 5-14.
10. Trinchieri G, Pflanz S, Kastelein RA (2003) The IL-12 family of heterodimeric cytokines: new players in the regulation of T cell responses. Immunology 19: 641-644.
11. Markey KA, MacDonald KP, Hill GR (2008) Impact of cytokine gene polymorphisms on graft-vs-host disease. Tissue Antigens 72: 507-516.
12. Chu CQ, Wittmer S, Dalton DK (2000) Failure to suppress the expansion of the activated CD4 T cell population in interferon gamma-deficient mice leads to exacerbation of experimental autoimmune encephalomyelitis. J Exp Med 192: 123-128.
13. Elmaagacli AH, Koldehoff M, Landt O, Beelen DW (2008) Relation of an interleukin-23 receptor gene polymorphism to graft-versus-host disease after hematopoietic-cell transplantation. Bone Marrow Transplant 41: 821-826.
14. Duerr RH, Taylor KD, Brant SR, Rioux JD, Silverberg MS, et al. (2006) A genome-wide association study identifies IL23R as an inflammatory bowel disease gene. Science 314: 1461-1463.
15. Huber AK, Jacobson EM, Jazdzewski K, Concepcion ES, Tomer Y (2008) Interleukin (IL)-23 receptor is a major susceptibility gene for Graves' ophthalmopathy: the IL-23/T-helper 17 axis extends to thyroid autoimmunity. J Clin Endocrinol Metab 93: 1077-1081.
16. Faragó B, Magyari L, Sáfrány E, Cööngei V, Járomi L, et al. (2008) Functional variants of interleukin-23 receptor gene confer risk for rheumatoid arthritis but not for systemic sclerosis. Ann Rheum Dis 67: 248-250.

17. Tsai JP, Yang SF, Wu SW, Hung TW, Tsai HC, et al. (2011) Association between interleukin 23 receptor polymorphism and kidney transplant outcomes: a 10-year Taiwan cohort study. Clin Chim Acta 412: 958-962.

18. National Kidney Foundation (2002) K/DOQI clinical practice guidelines for chronic kidney disease: evaluation, classification, and stratification. Am J Kidney Dis 39: S1-266.

19. Knoll GA, Cantarovitch M, Cole E, Gill J, Gourishankar S, et al. (2008) The Canadian ACE-inhibitor trial to improve renal outcomes and patient survival in kidney transplantation-study design. Nephrology Dialysis Transplantation 23: 354-358.

20. Fellström B, Holdaas H, Jardine AG, Nyberg G, Grönhagen-Riska C, et al. (2005) Risk factors for reaching renal endpoints in the assessment of Lescol in renal transplantation (ALERT) trial. Transplantation 79: 205-212.

21. National Kidney Foundation (2014) GFR decline as an endpoint in clinical trials for CKD.

22. Haas M, Sir B, Racusen LC, Soley K, Glotz D, et al. (2014) Banff 2013 meeting report: inclusion of cld-negative antibody-mediated rejection and antibody-associated arterial lesions. Am J Transplant 14: 272-283.

23. Djamali A, Kaufman DB, Ellis TM, Zhong W, Matas A, et al. (2014) Diagnosis and management of antibody-mediated rejection: current status and novel approaches. Am J Transplant 14: 255-271.

24. Chung BH, Oh HJ, Piao SG, Sun IO, Kang SH, et al. (2011) Higher infiltration by Th17 cells compared with regulatory T cells is associated with severe acute T-cell-mediated graft rejection. Exp Mol Med 43: 630-637.

25. Loverre A, Tataranni T, Castellano G, Divella C, Battaglia M, et al. (2011) IL-17 expression by tubular epithelial cells in renal transplant recipients with acute antibody-mediated rejection. Am J Transplant 11: 1248-1259.

26. Paust HJ, Turner JE, Steinmetz OM, Peters A, Heymann F, et al. (2009) The IL-23/Th17 axis contributes to renal injury in experimental glomerulonephritis. J Am Soc Nephrol 20: 969-979.

27. Zhang Z, Kyttaris VC, Tsokos GC (2009) The role of IL-23/IL-17 axis in lupus nephritis. J Immunol 183: 3160-3169.