Treatment of Chronic Active Antibody-mediated Rejection in Renal Transplant Recipients – A single center retrospective study

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Hsien-Fu Chiu  hsienfuchiu@gmail.com
Taichung Veterans General Hospital
Corresponding Author
ORCiD: 0000-0001-9565-002X

Mei-Chin Wen
Taichung Veterans General Hospital

Ming-Ju Wu
Taichung Veterans General Hospital

Cheng-Hsu Chen
Taichung Veterans General Hospital

Tung-Min Yu
Taichung Veterans General Hospital

Ya-Wen Chuang
Taichung Veterans General Hospital

Shih-Ting Huang
Taichung Veterans General Hospital

Shang-Feng Tsai
Taichung Armed Forces General Hospital

Ying-Chih Lo
Taichung Veterans General Hospital

Hao-Chung Ho
Taichung Veterans General Hospital

Kuo-Hsiung Shu
Lin Shin Medical Corporation Lin Shin Hospital

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Abstract

Background: A plethora of evidences suggest that the most important cause of late graft loss in renal transplant recipients is chronic active antibody-mediated rejection. However, there are no consensus on treatment strategies. Methods: We retrospectively analyzed clinical and pathological data of renal transplant recipients who received kidney graft biopsy with confirmed diagnosis of chronic active antibody-mediated rejection in the past 7 years. The patients were divided into two groups according to treatment strategy: Group 1: aggressive treatment (double filtration plasmapheresis and one of the followings: rituximab, intravenous immunoglobulin, antithymogycte globulin, bortezomib, or methylprednisolone pulse therapy); and group 2: supportive treatment. Results: From February 2009 to December 2017, a total of 82 graft biopsies with diagnosis of chronic active antibody mediated rejection were identified. Kaplan-Meier analysis of death-censored graft survival showed a worse survival in group 2 (P = 0.015 by log-rank test). Adverse event-free survival was lower in group 1, whereas patient survival was no significant different. Proteinuria and supportive treatment were independent risk factors for graft loss in multivariate analysis. Conclusions: Aggressive treatment was associated with better graft outcome. However, higher incidence of adverse events merit personalized treatment, especially for those with higher risk of infection. Appropriate prophylactic antibiotics are recommended for aggressive treatment patients.

Introduction

Chronic active antibody mediated rejection (CAMR) had become one of the most frequent etiology associated with graft failure. Diagnosis is based on the detection of donor-specific antibodies (DSAs) and specific morphologic lesions, most importantly, microvascular inflammation/injury with or without capillary C4d deposition (1). A variety of treatment
strategies have been proven effective for acute antibody mediated rejection (2). On the contrary, treatment for CAMR has remained a major challenge.

Current therapeutic approaches for CAMR are based on retrospective studies and pilot trials, including intravenous immunoglobulin (IVIG) plus rituximab (3, 4), proteasome inhibitor-bortezomib (5), complement inhibitor-eculizumab (6), and IL-6 receptor blocker (7). However, the result in these studies not always turned out to be effective (4, 5), and the adverse events derived from these immunosuppressants are of great concern. Therefore, our study aimed to present the outcome of CAMR in our center and to compare graft survival between different treatment strategies.

Materials And Methods

Patients and sample collection

We retrospectively analyzed clinical and pathological data of renal transplant recipients (RTx) who received kidney graft biopsy with confirmed diagnosis of CAMR in the past 7 years in Taichung Veterans General Hospital. The clinicians arranged a graft biopsy whenever there was renal dysfunction or appearance/elevation of proteinuria. If the patient had multiple biopsies, the data of the first confirmation biopsy were used for statistical analysis. To avoid confounding factors, patients with coexisted DM nephropathy, recurrent glomerulonephritis (GN) or de novo GN were excluded. All of the patients were transplanted with a negative cytotoxicity cross-match assay performed on T and B lymphocytes with the sera of the day of transplantation. Induction therapy included thymoglobulin or basiliximab. Maintenance immunosuppression included calcineurin inhibitors (CNIs) tacrolimus or cyclosporine A, mycophenolate, and prednisone. mTOR inhibitor, either sirolimus or everolimus, was prescribed in few patients depending on the discretion of the physician.

One or more of the following treatment strategies were selected for CAMR treatment
according to the patient’s clinical condition and decision of the individual practitioners: no
treatment, methylprednisolone (MP) pulse therapy (usually 500mg of MP for 3 days),
double filtration plasmapheresis (DFPP), rituximab intravenous bolus (375 mg/m^2),
intravenous immunoglobulin (IVIG) (2 g/kg), rabbit antithymocyte globulin (ATG)
(Thymoglobulin 1-1.5mg/kg for 3–5 days). In a few patients, bortezomib (1.3 mg/m^2) were
also used. Multiple treatments, usually yearly, were performed if follow-up graft biopsy
revealed persistent lesions. The patients were divided into two groups according to
treatment strategy. Group 1: aggressive treatment (DFPP and one of the followings:
rituximab, IVIG, ATG, bortezomib, or MP pulse therapy); and group 2: supportive
treatment. In group 1, patients were usually treated annually with DFPP plus one of the 5
drugs, but different in each year in order to accomplish a wide blockade of the
alloimmunity.
Lab data including serum creatinine, estimated glomerular filtration rate (eGFR),
proteinuria (measured by either albumin to creatinine ratio or protein to creatinine ratio),
panel reactive antibody (PRA) titer were retrieved from medical charts. The Banff scores
of each item were retrieved from the formal pathologic report made by a well-trained
pathologist familiar with kidney transplant pathology.

**End points**
The patients were followed up until graft loss or death or the end of 2017. Primary end
point was graft survival after treatment in the 2 groups. Secondary outcome included
patient survival and the occurrence of major adverse events. Major adverse event was
declared by any event that was associated with death, admission to hospital, prolongation
of a hospital stay, persistent or significant disability or incapacity, or was otherwise life-
threatening in connection with specific treatment, according to World Health Organization
Histopathology and diagnosis of AMR

Graft biopsy specimen was fixed in alcoholic Bouin’s fixative and paraffin-embedded for routine light microscopy with hematoxylin-and eosin, periodic acid-Schiff, Masson’s trichrome, and Jones silver methenamine stains. Immunofluorescence studies for IgG, IgA, IgM, C3, C4d, C1q, kappa, and lambda light chains were done. Electron microscopy was routinely performed for graft biopsy.

The graft biopsies were reviewed by a renal pathologist and graded according to Banff 2017 criteria (8). The biopsies were graded from 0 to 3 according to the Banff histologic parameters: glomerulitis (g), interstitial inflammation (i), tubulitis (t), endarteritis (v), peritubular capillary inflammation (ptc), transplant glomerulopathy (cg), mesangial matrix increase (mm), interstitial fibrosis (ci), tubular atrophy (ct), arterial fibrous intimal thickening (cv), and arteriolar hyaline thickening (ah) (1, 9). C4d staining was graded from 0 to 3 by the percentage of peritubular capillaries with linear staining. For CAMR, all 3 criteria in the following were met for diagnosis according to Banff 2017 criteria: (1) morphologic evidence of chronic tissue injury, (2) evidence of current/recent antibody interaction with vascular endothelium, (3) serologic evidence of donor-specific antibodies (DSA, to HLA or other antigens). C4d staining in the biopsy tissue or expression of validated transcripts/classifiers may substitute for DSA (8). Determination of HLA antibody by Luminex method is expensive in Taiwan and is not affordable to every patient. Gene expression is not performed routinely. For those who didn’t perform DSA, C4d staining should be positive for the definite diagnosis of CAMR.

Data analysis

Continuous variables were described using means and standard deviations (SD) or medians and interquartile ranges (IQR). Categorical variables were shown as frequency
Continuous variables were examined for normality by Kolmogorov-Smirnov test. The $\chi^2$ test or Fisher exact test were performed to compare categorical variables. One-way analysis of variance (ANOVA) testing was performed to compare continuous variables normally distributed. The Kruskal-Wallis test was performed to compare continuous variables not normally distributed.

Freedom from graft loss, adverse events and patient survival were estimated by the Kaplan-Meier method, and the log-rank test was used to assess group differences. Cox proportional hazards regression analyses were conducted to determine the association of graft loss with key variables including age, creatinine at the time of biopsy, transplant duration, donor type, PRA class I, PRA class II, proteinuria, Banff scores, and aggressive treatment. The multivariate model was adjusted for the following parameters: PRA class II, proteinuria and creatinine at the time of biopsy, ci+ct, transplant duration and aggressive treatment. All statistics were analyzed using SPSS software (version 21.0, Chicago, IL, USA). Significance was defined as $P < 0.05$.

Results

Comparison of patient demographics

From February 2009 to December 2017, a total of 85 graft biopsies with diagnosis of CAMR were identified. 3 cases were excluded for analysis owing to short follow up duration (less than 6 months). Group 1 comprised of 59 cases, whereas group 2 comprised of 23 cases.

In group 1, besides DFPP, 40 patients had received Rituximab, 10 patients had received IVIG, 10 patients had received bortezomib, whereas 4 patients had received antithymocyte globulin and 17 patients had received MP pulse therapy only.

There was no statistical significance difference between group 1 and group 2 in terms of age, donor type, transplant duration, follow up duration, percentages of diabetes mellitus, hepatitis B or C, panel reactive antibody (PRA) class I and II titer, percentages of patients
who received induction treatment, immunosuppressive regimen (cyclosporine based or tacrolimus based), serum creatinine, proteinuria, and Banff scores (including cg, ci + ct, mm, g+ptc) (Table 1).

Survival analysis

Patients were followed for a median of 32.59 (IQR 24.01–49.89) months after the diagnosis of CAMR. A total of 22 (26.82%) patients lost their allograft, including 11/59 patients (18.64%) in group 1 and 11/23 (47.83%) patients in group 2. Median graft survival was 6.45 and 3.68 years for group 1 and group 2, respectively. Overall median graft survival was 5.6 years. Kaplan-Meier analysis of death-censored graft survival showed a worse survival in group 2 ($P = 0.015$ by log-rank test) (Figure 1).

A total of 9 (10.97%) patients died after the diagnosis of CAMR, including 6/59 (10.16%) patients in group 1, and 3/23 (13.04%) in group 2. All the mortality cases in group 1 died of sepsis. On the other hand, 2 of those in group 2 died of sepsis, and 1 case died of hemorrhagic shock due to hemothorax. Patient survival at the end of this study was not significant different between these groups ($P = 0.567$ by log-rank test) (Figure 2).

Predictors of graft loss

By univariate analysis, the significant predictors of graft loss for CAMR were creatinine, proteinuria, PRA class II, cg ≥ 1, ci + ct ≥ 3, and supportive treatment. We constructed a multivariate regression model for graft loss analysis by adjustment of proteinuria, creatinine, cg score and aggressive treatment. Supportive treatment (HR 2.86, 95%CI [1.05–7.77]) and proteinuria (HR 1.39, 95% CI [1.06–1.83]) were independently associated with graft loss. (Table 2)

Adverse events

Major adverse events were demonstrated in table 3. Total number of adverse events were 54 in group 1 and 7 in group 2. Mean number of adverse events per patient was higher in
group 1 ($P < 0.001$). Adverse event free survival was significantly better in group 2 ($P = 0.002$ by log-rank test) (Figure 3). The most frequent adverse events in aggressive treatment group were CMV disease, leucopenia, urinary tract infection, pneumonia, infectious diarrhea, and \textit{Pneumocystis carinii} pneumonia (PCP). Median adverse event free survival was 6.0 (95% CI: 3–24) months in the aggressive treatment group.

Discussion

In this retrospective study, we indicated that aggressive treatment for CAMR patients was associated with better graft survival. However, aggressive treatment group also had higher incidence of adverse events and a reduced adverse event free survival. The factors independently associated with graft loss were proteinuria and supportive treatment. Currently, there are no approved treatments for CAMR. Billing et al. reported a prospective pilot study on an antihumoral therapy consisting of IVIG and rituximab in 20 paediatric renal transplant recipients with CAMR. Over an observation period of 2 years, IVIG and rituximab significantly reduced or stabilized the progressive loss of transplant function (3, 10) However, the subgroup with transplant glomerulopathy (TG) was associated with a poorer response. Another study conducted by Bachelet et al. showed IVIG with rituximab treatment for severe TG in CAMR did not change the natural history of TG (4). Recently, a multicenter, prospective, randomized double-blind clinical trial for evaluation the efficacy and safety of IVIG with rituximab also revealed no difference between the treatment and placebo groups in eGFR decline, increase of proteinuria, and MFI of the immunodominant DSA. The author considered the presence of TG as an inclusion criteria (mean cg score in the treatment group: 2.3 ± 0.8), and this may be the reason of a poor response in this study (11). In fact, there was evidence that the combination of IVIG and rituximab appeared to be beneficial in patients with high levels of microvascular injury, for example biopsies with $g \geq 2$ and/or $(g + ptc) \geq 4$ (12). On the
contrary, patients with low microvascular injury scores appeared less likely to benefit from antihumoral therapy.

Bortezomib had also been evaluated in patients with CAMR. Clinical experience of bortezomib in transplantation had shown variable results among patients with different disease states and populations. Recently, a randomized, placebo-controlled trial (the Bortezomib in Late Antibody-Mediated Kidney Transplant Rejection [BORTEJECT] Trial) investigated whether two cycles of bortezomib (each cycle: 1.3 mg/m² intravenously on days 1, 4, 8, and 11) prevent GFR decline by halting the progression of late DSA-positive ABMR. However, no significant differences between bortezomib- and placebo-treated groups in median measured GFR at 24 months, 2-year graft survival, urinary protein concentration, and DSA levels (5). Advanced tissue injury and higher proportion of preformed DSAs (19 of 21 patients) might be a possible explanation. On the other hand, HLA antibodies produced by long-lived plasma cells (LLPCs) are more refractory to proteasome inhibitor therapy. LLPC resistance and immunologic compensatory mechanisms may also play a role for treatment failure (13).

In our study, CAMR were diagnosed at relatively early stage (median cg score: 1.0, ci+ct: 2.0) with compared to previous studies (mean cg score: 2.0; ci+ct: 3.5 in a recent clinical trial (11) and mean cg score: 2.2; ci+ct score: 2.8 in a previous retrospective study (4)). Furthermore, the microvascular injury was prominent (median [g + ptc] score: 3.5). The above characteristics made our patients more likely to respond to antihumoral therapy. The graft survival was significantly better in the aggressive treatment group with compared to supportive treatment group. Supportive treatment was a predictor of graft loss in univariate analysis (HR 2.77, 95% CI [1.19–6.41], P = 0.017). After adjustment of proteinuria, creatinine, and cg score, supportive treatment was still an independent risk factor of graft loss (HR 2.86, 95%CI [1.05–7.77], P = 0.038). Our study highlight the
importance of aggressive treatment in CAMR under the circumstance of earlier stage and higher degree of microvascular injury.

Rituximab, IVIG, and bortezomib treatment are not reimbursed by national insurance in Taiwan and should be self-paid. Therefore, some patients received methylprednisolone pulse therapy with plasmapheresis only. These patients had graft survival between aggressive treatment and supportive treatment group (supplement Fig 1). Previously, Redfield et al. had reported a retrospective study for outcome of CAMR (14). The author divided their patients into three groups: steroid/IVIG with rituximab or antithymocyte globulin, steroid/IVIG alone or in combination, and no treatment. The most aggressive treatment group had the best graft survival, which was in line with our study. However, the graft biopsies of CAMR in Redfield’s series had relatively advanced disease (median cg of 2 and proteinuria > 1g). Therefore, the graft survival in our study was better (overall median graft survival 5.4 years vs. 1.9 years).

Following aggressive treatment of CAMR, adverse event is an important issue. The most frequently prescribed antihumoral agent for our patients was rituximab, followed by IVIG, bortezomib, and ATG. A retrospective study published by Kamar et al. revealed that, following rituximab treatment, 9.1% of kidney transplant patients died due to infectious disease (15). This result was similar to that in our aggressive treatment group (10.16%). The most common adverse events in our patients were CMV disease, urinary tract infection, bacterial / PCP pneumonia, and infectious diarrhea.

CMV infection had been reported to be associated with Rituximab, ATG, and bortezomib treatment in renal transplant and myeloma patients (16–18). ABO-incompatible kidney transplant recipients who received rituximab had higher incidence of CMV disease (16). Studies revealed an increased frequency of CMV disease associated with ATG treatment, probably due to the release of TNF-α after ATG administration, which may stimulate
cellular nuclear factor κB and viral replication via binding to the promoter region of the CMV immediate-early antigen gene (17). Furthermore, several studies indicated that bortezomib treatment is associated with higher risk of viral infection, including CMV (18–20). Basler M et al. demonstrated reduced cytotoxic T cell response and impaired viral clearance in bortezomib treated mice (19).

PCP is a major cause of morbidity and mortality in patients receiving immunosuppressant therapies. The risk factors for the development of PCP in kidney transplant patients include: overall load of immunosuppressive therapy, higher donor age, higher recipient age, lymphopenia, previous CMV infection, or treatment used for episodes of graft rejection (21).

Focusing on the mortality cases in our aggressive treatment group, 3 of 6 mortality cases died of CMV disease, including 1 CMV pneumonia and 2 CMV colitis. Adequate valganciclovir prophylaxis may reduce mortality rate by 50 percent in our patients who received aggressive treatment.

Kamar et al. demonstrated that the median duration between last rituximab and first infection episode in kidney transplant recipients was about 5 months (15), which was in accordance with our study (6 months). They found the independent predictive factors for infection-induced death were the combined use of rituximab and ATG given for induction or anti-rejection therapy, recipient age, and bacterial and fungal infections. Our previous policy about prophylactic antibiotics (valganciclovir and trimethoprim-sulfamethoxazole) was to give these 2 agents for just one month after aggressive treatment. In this regards, it is reasonable to recommend that valganciclovir and trimethoprim-sulfamethoxazole prophylaxis should be given for at least 5–6 months after aggressive anti-rejection therapy.

Despite the significantly higher rate of adverse events in aggressive treatment group,
there was no significant difference in patient survival (Figure 2), implying that the patients still could have a reasonable chance of survival if these complications can be treated judiciously.

There are limitations in our study. Firstly, this is a retrospective study. There were no rules for treatment of CAMR in our cohort, and the need for treatment was determined by each clinical physician. Second, DSA was performed in limited patients, thus the role of DSA can not be obtained exactly from this study. This is because determination of HLA antibody by Luminex method is expensive in Taiwan and is not affordable to many patients. To compensate this shortcoming, annual PRA are usually performed, which is reimbursed by national health insurance.

In conclusion, although there have been no consensus on treatment strategies on CAMR, aggressive treatment before advanced tissue injury is still associated with better graft outcome in our series. However, higher incidence of adverse events cannot be overlooked. To mitigate potential life-threatening infections, longer duration of trimethoprim-sulfamethoxazole and valganciclovir prophylaxis should be considered after aggressive treatment for rejection.

**Abbreviations**

CAMR, chronic antibody-mediated rejection; RTx, renal transplant recipients; GN, glomerulonephritis; PRA, panel-reactive antibody; DSA, donor-specific antibody; eGFR, estimated glomerular filtration rate; IVIG, intravenous immunoglobulin; ATG, antithymocyte globulin; PCP, *Pneumocystis carinii* pneumonia; MP, methylprednisolone; DFPP, double filtration plasmapheresis.

**Declarations**

*Ethics approval and consent to participate*
The study and protocol were reviewed and approved by the Medical Ethics Committee of Taichung Veterans General Hospital. Written informed consent for participation was waived because of the retrospective nature of the study.

The study complies with the Declaration of Helsinki.

Consent for publication

Not applicable.

Competing interests

The author declare that they have no competing interests.

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Availability of data and materials.

The individual patient-level data was not made publically available due to containing potentially identifying patient data; however, the study data may be made available from the authors upon reasonable request.

Authors’ contributions

HFC: drafted the manuscript and involved with data analysis and interpretation;

MCW: pathological diagnosis of graft biopsy, revision of the manuscript. MJW, CHC, TMY, YWC, STH: revised the manuscript and involved with patient care. SFT, YCL, HCH: drafted the manuscript, final approval of the manuscript. KHS(corresponding author): drafted, revised, and final approval of the manuscript.

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Tables
Table 1. Comparison of patient demographics between different treatment groups

|                                | Group 1, Aggressive treatment (N = 59) | Group 2, Supportive treatment (N = 23) | P value |
|--------------------------------|----------------------------------------|----------------------------------------|---------|
| Age at biopsy                  | 50.47 [42.76-58.79]                    | 55.64 [47.06-65.35]                    | 0.052   |
| Donor type                     |                                        |                                        | 0.245   |
| Deceased                       | 43 (74.1%)                             | 19 (86.4%)                             |         |
| Living                         | 15 (25.9%)                             | 3 (13.6%)                              |         |
| Transplant duration (mo)       | 95.61 [62.22-161.63]                   | 123.22 [68.91-209.42]                  | 0.394   |
| Follow up duration (mo)        | 34.75 [27.62-50.97]                    | 30.88 [14.79-44.82]                    | 0.163   |
| DM                             | 9 (16.7%)                              | 3 (13.0%)                              | 0.690   |
| HBV                            | 5 (9.1%)                               | 1 (4.3%)                               | 0.476   |
| HCV                            | 8 (14.5%)                              | 4 (17.4%)                              | 0.752   |
| HLA mismatches                 | 2.0 [0.0-3.5]                          | 0.0 [0.0-1.0]                          | 0.279   |
| PRA                            |                                        |                                        |         |
| Class I                        | 0.00 [0.00-15.65]                      | 0.00 [0.00-0.00]                       | 0.293   |
| Class II                       | 19.00 [0.00-58.50]                     | 32.00 [0.00-81.75]                     | 0.911   |
| Induction                      |                                        |                                        | 0.657   |
| ATG                            | 1 (1.8%)                               | 0 (0.0%)                               | 0.388   |
| Anti-CD 25                     | 12 (28.6%)                             | 3 (13.0%)                              |         |
| No induction                   | 30 (71.4%)                             | 20 (87.0%)                             |         |
| Regimen                        |                                        |                                        | 0.498   |
| CsA based                      | 16 (30.2%)                             | 7 (38.9%)                              |         |
| FK-506 based                   | 37 (69.8%)                             | 11 (61.1%)                             |         |
| Creatinine                     | 1.80 [1.40-2.42]                       | 1.84 [1.50-2.94]                       | 0.635   |
| eGFR                           | 31.35 [23.25-45.83]                    | 24.70 [19.90-40.25]                    | 0.148   |
| Proteinuria (g/d)              | 0.51 [0.22-1.74]                       | 1.49 [0.40-2.03]                       | 0.094   |
| Banff score                    |                                        |                                        |         |
Table 2. Predictors of death-censored graft loss in cAMR patients

A. Univariate analysis

| Predictor                        | Hazard Ratio (95% CI) | P Value |
|----------------------------------|-----------------------|---------|
| Age                              | 1.02 (0.98-1.07)      | 0.518   |
| Creatinine (mg/dl)               | 1.31 (1.12-1.52)      | 0.002   |
| PRA class I                      | 1.01 (0.99-1.02)      | 0.186   |
| PRA class II                     | 1.03 (1.01-1.04)      | 0.002   |
| Proteinuria, g/d                 | 1.37 (1.15-1.64)      | 0.0004  |
| cg score ≥ 1                     | 4.97 (1.47-16.65)     | 0.009   |
| (ci + ct) ≥ 3                    | 6.32 (2.01-19.85)     | 0.002   |
| C4d score ≥ 1                    | 1.36 (0.58-3.19)      | 0.476   |
| mm ≥ 1                           | 1.82 (0.48-6.84)      | 0.374   |
| Transplant duration (mo)         | 1.00 (0.99-1.01)      | 0.68    |
| No treatment                     | 2.77 (1.19-6.41)      | 0.017   |

B. Multivariable analysis

| Predictor                        | Hazard ratio (95% CI) | P value |
|----------------------------------|-----------------------|---------|
| Supportive treatment             | 2.86 (1.05-7.77)      | 0.038   |
| Proteinuria (g/d)                | 1.39 (1.06-1.83)      | 0.016   |
| Creatinine (mg/dl)               | 1.11 (0.73-1.68)      | 0.621   |
| cg score ≥ 1                     | 3.00 (0.81-11.22)     | 0.102   |

* The multivariate model was adjusted for the following parameters: proteinuria, creatinine, cg score, and treatment strategy.

Table 3. Major Complications. (Definition: admission, organ failure or mortality)
| Infection                          | Group 1, Aggressive treatment (N = 59) | Group 2, Supportive treatment (N = 23) | P-Value |
|-----------------------------------|---------------------------------------|---------------------------------------|---------|
| CMV disease                       | 10                                    | 3                                     | 0.663   |
| Bacterial pneumonia               | 9                                     | 2                                     | 0.433   |
| PCP pneumonia                     | 4                                     | 0                                     | 0.650   |
| Cryptococcal pneumonia            | 1                                     | 0                                     | 0.505   |
| Penicillium marneffei pneumonia   | 1                                     | 0                                     | 0.505   |
| Infectious diarrhea               | 6                                     | 1                                     | 0.372   |
| Urinary tract infection           | 8                                     | 0                                     | 0.212   |
| Epididymitis                      | 1                                     | 0                                     | 0.505   |
| Cellulitis                        | 3                                     | 0                                     | 0.889   |
| Abscess, retroperitoneal          | 1                                     | 0                                     | 0.505   |
| Abscess, lung                     | 1                                     | 0                                     | 0.505   |
| Peritonitis                       | 1                                     | 0                                     | 0.505   |
| Fungemia                          | 1                                     | 0                                     | 0.505   |
| Leukopenia                        | 8                                     | 1                                     | 0.231   |
| Mortality                         | 6                                     | 3                                     | 0.708   |
| Total number of AEs               | 54                                    | 7                                     |         |
| Mean number AEs per patient       | 1.0 [0.0-2.2]                         | 0.0 [0.0-0.7]                         | < 0.001 |

* All the mortality cases in the aggressive treatment group died of sepsis, whereas two of those in no treatment group died of sepsis, another case died of hemorrhagic shock. PCP, *Pneumocystis carinii* pneumonia. AE, adverse events.

**Figures**
Figure 1

Kaplan-Meier graft survival analysis. Graft survival was constructed for aggressive treatment group and supportive treatment group. Kaplan-Meier analysis of death-censored graft survival showed a significantly worse survival in supportive treatment group (P = 0.015 by log-rank test).
Figure 2

Kaplan-Meier analysis of patient survival. Study groups did not significantly differ in Kaplan-Meier patient survival ($P = 0.567$ by log-rank test).
Kaplan-Meir analysis of the occurrence of major adverse events. Survival without adverse events was significantly reduced in the aggressive treatment group ($P = 0.002$ by log-rank test).

**Supplementary Files**

This is a list of supplementary files associated with the primary manuscript. Click to download.

Supplement Fig.tif