Transplant International

ORIGINAL ARTICLE

What happens after graft loss? A large, long-term, single-center observation

Eva Schrezenmeier[1,2], Lukas J. Lehner[1], Marina Merkel[1], Manuel Mayrdorfer[1], Wiebke Duettmann[1], Marcel G. Naik[1], Felix Fröhlich[1], Lutz Liefeldt[1], Mareen Pigorsch[3], Frank Friedersdorff[4], Danilo Schmidt[1], Matthias Niemann[4], Nils Lachmann[5,6], Klemens Budde[1] & Fabian Halleck[1]

1 Department of Nephrology and Medical Intensive Care, Charité – Universitätsmedizin Berlin, Berlin, Germany
2 Berlin Institute of Health, Berlin, Germany
3 Institute of Biometry and Clinical Epidemiology, Charité – Universitätsmedizin Berlin, Berlin, Germany
4 Department of Urology, Charité – Universitätsmedizin Berlin, Berlin, Germany
5 PIRCHE AG, Berlin, Germany
6 HLA Laboratory, Institute for Transfusion Medicine, Charité – Universitätsmedizin Berlin, Berlin, Germany

Correspondence
Dr. Eva Schrezenmeier, Department of Nephrology and Medical Intensive Care, Charité – Universitätsmedizin Berlin, Charitéplatz 1, 10117 Berlin, Germany.
Tel.: +49 30 450 614246;
fax: +49 30 450 752451;
e-mail: eva-vanessa.schrezenmeier@charite.de

ES and LJL contributed equally to this manuscript.

SUMMARY

The number of patients returning to dialysis after graft failure increases. Surprisingly, little is known about the clinical and immunological outcomes of this cohort. We retrospectively analyzed 254 patients after kidney allograft loss between 1997 and 2017 and report clinical outcomes such as mortality, relisting, retransplantations, transplant nephrectomies, and immunization status. Of the 254 patients, 49% had died 5 years after graft loss, while 27% were relisted, 14% were on dialysis and not relisted, and only 11% were retransplanted 5 years after graft loss. In the complete observational period, 111/254 (43.7%) patients were relisted. Of these, 72.1% of patients were under 55 years of age at time of graft loss and only 13.5% of patients were ≥65 years. Age at graft loss was associated with relisting in a logistic regression analysis. In the complete observational period, 42 patients (16.5%) were retransplanted. Only 4 of those (9.5%) were ≥65 years at time of graft loss. Nephrectomy had no impact on survival, relisting, or development of dnDSA. Patients after allograft loss have a high overall mortality. Immunization contributes to long waiting times. Only a very limited number of patients are retransplanted especially when ≥65 years at time of graft loss.

Transplant International 2021; 34: 732–742

Key words
dnDSA, graft loss, immunization, PIRCHE score, relisting

Received: 4 July 2020; Revision requested: 1 September 2020; Accepted: 26 January 2021;
Published online: 19 February 2021

Introduction

The number of patients who are relisted for kidney transplantation because of graft loss increases [1]. It has been shown that previous transplant failure increases mortality among waitlisted patients after relisting. This is most likely due to a collection of cumulative risks such as cardiovascular risk and the risk for cancer associated with kidney transplantation [2], but more detailed data about timing and circumstances of death after graft loss and rate of retransplantation are scarce. What exactly happens to patients after graft is largely unknown even though it has been recognized as an important late outcome parameter in kidney transplantation [3,4]. Factors that contribute to relisting of a patient are largely not described although found

doi:10.1111/tri.13834

intuitive that physically fit patients are relisted and unfit are not. Data on how nephrectomy impacts on retransplantation and relisting are inconsistent ranging from high numbers of nephrectomies in European cohorts with over 60% [5] and lower numbers around 30% in US cohorts [6]. The relation of transplant nephrectomy and short-term development of dnDSA is also inconsistent [7–9]. A longer waiting time before retransplantation is associated with worse outcomes such as early acute rejection, severe vascular and humoral rejection, all-cause mortality, and death with a functioning graft independent of HLA mismatches [10]. In this context, the precise impact of presence of de novo donor-specific antibodies (dnDSA) has not yet been investigated. Kidney transplant candidates with prior graft loss are known to be highly sensitized and have more anti-HLA antibodies, irrespective if they underwent allograft nephrectomy or only withdrawal of immunosuppression [11,12]. As a consequence, the fear comes that dnDSA lead to a higher number of unacceptable antigens, which might as a consequence lead to a longer waiting time [13]. But whether this causality between dnDSA and waiting time also considering Eurotransplant Acceptable Mismatch (AM) program in the case of retransplantation is true is not clear. The timing of occurrence of HLA immunization and the consequence of dnDSA for a potential relisting are also largely unknown.

Within the last years, more insights have been gained on human HLA biology and so-called epitopes, which are the antibody accessible binding sites of HLA alleles have been described [14]. Epitope-matching algorithms such as the PIRCHE (Predicted Indirectly ReCognizable HLA Epitopes) score likely allow a better matching then broad antigen HLA matching and thereby might improve matching algorithms, which might lead to better allograft outcomes [15]. In already performed transplants, it might be able to predict the occurrence of dnDSA better than it can be explained by a consideration of HLA antigens alone.

The aim of this study was to analyze clinical outcomes such as mortality, relisting, retransplantations, transplant nephrectomies, and immunization status in a large contemporary cohort with complete follow-up.

Methods

Study subjects

In this single-center study, all kidney graft failures of adults between 1997 and 2017 were retrospectively analyzed. The end of observation was December 31, 2017. The date of graft loss was the date when renal replacement therapy was started or the patient was retransplanted. If not otherwise indicated, only the first allograft loss within the observation period was evaluated. Patients with primary nonfunction were included in all analysis. None of the included patients received a combined transplantation. Our electronic patient data base [16] and all other available medical records were used for data collection. If needed, additional data were obtained from dialysis centers and hospitals. Missing cases for analysis are indicated in the results section. The ethics committee of Charité—Universitätsmedizin Berlin approved the study, and conformity with the Declaration of Helsinki was ensured.

Center policy

Our standard immunosuppressive protocol (except for participation in clinical trials) consisted of induction therapy (in most cases anti-IL2-R antibody), calcineurin inhibitor, mycophenolate, and steroids, aiming at a steroid-free regimen after the first year if no rejection episodes had occurred. As a center policy, we do not accept repeated mismatches in retransplantations. The listing within the Eurotransplant acceptable mismatch (AM) program has always been performed according to the current Eurotransplant rules. Since the beginning, we aimed to include all potential candidates in the AM program. Current eligibility criteria for inclusion are a cumulative waiting time of ≥2 years and a CDC complement-dependent cytotoxicity panel-reactive antibodies (cPRA) of >85% in either historic or current serum sample. After graft loss, we aim to reduce immunosuppression in a stepwise fashion. At time of graft loss, immunosuppression is reduced from a triple therapy to a double therapy and over the next 2–3 months further reduced to monotherapy, in most cases steroids (e.g., 4 mg methylprednisolone), which are then slowly tapered down to zero over the next 6–12 months in order to prevent hypocortisolism.

HLA antibody testing

HLA antibody testing has been performed routinely once yearly, when rejection was clinically suspected as well as in case of relisting for transplantation in regular quarterly screening intervals [6,17]. Until 2007, enzyme-linked immunosorbent assay (ELISA) Lambda Antigen Tray (LAT; One Lambda, Canoga Park, CA, USA) has been performed. In 2009, a complete conversion to

Luminex®-based LABScreen® mixed and SAB assay (One Lambda) has been done, from 2007 until 2009 both assays were used [18,19]. All tests were performed according to the manufacturer's instructions. For the SAB assay, a normalized mean fluorescence intensity value exceeding 1000 was defined as positive in the pre- and post-transplant setting. The HLA loci A, B, C, DRB, and DQB were considered for the definition of dnDSA. Due to missing typing, DQA, DPA, and DPB could not be considered for this analysis.

The cPRA was calculated as the percentage of positive reaction against a panel of blood donor HLA antigens based on combined HLA class I (A and B) plus II (DR) specificities according to Eurotransplant HLA point calculation requirements (https://www.etrl.org/vPRA.aspx). Epitope matching was performed to predict development of dnDSA using the PIRCHE algorithm (score 0–300). PIRCHE numbers were calculated using a web-based tool provided by PIRCHE AG (www.pirche.org) as previously described [15]. PIRCHE I and II values were computed for each patient, and the sum of PIRCHE I and II was considered as PIRCHE score.

Statistical analysis

Patient cohort characteristics and parameters were summarized as mean with standard deviation or, in case of non-normal distribution of continuous variables, as median with IQR. The time-to-event outcome data with respect to dnDSA development and death-censored allograft survival were assessed by Kaplan–Meier plots and log-rank tests. Landmark analysis was used to avoid immortal time bias. For the multivariate analyses, a cox regression and a logistic regression were conducted. Used variables are indicated in Tables S1 and S2. Analyses were conducted using SPSS 25 (IBM Corp., Armonk, NY, USA).

Results

Patient characteristics

A flowchart of the study population is shown in Fig. 1. Overall, 254 patients with 267 allograft failures were identified between 1997 and 2017. Follow-up data were available for all patients until December 31, 2017. If more than one allograft loss occurred in the observational period, only the first has been included in the subsequent analysis to avoid a dependency of data. The median follow-up of these patients after graft loss was 7.4 years (IQR: 3.9–11.4). Demographics of the population are shown in Table 1. Median age at graft loss was 57 years (IQR: 42–69). At the time of allograft failure, 39% of patients were older or equal than 65 years. In 211/254 patients, only one (first) allograft loss occurred in the observational period, whereas 39/254 patients had their second transplant failure, 3/254 patients had their third transplant failure, and one patient had a fourth transplant failure within the observational period. Patients who had their second transplant failure within the observational period were significantly younger (Table 1). The main reasons for graft loss are summarized in Table 2. Medical events were defined as severe medical conditions such as cardiovascular events or infections timely linked to a persistent and relevant decrease in allograft function. The most common reasons were rejections, medical, or multifactorial causes.

Patient outcomes

Mortality of patients after graft loss

In our cohort, 19.2% of patients died within 1 year after graft loss, 35.2% within 3 years and 76.1% 10 years after graft loss. Figure 2 illustrates the percentage of patients with status dead, transplanted, waitlisted, and on dialysis (but not waitlisted) up to 10 years after graft loss. A 10 years follow-up after graft loss was available for 62.6% of patients.

Overall survival depends on age (Fig. 3a). Furthermore, patients who were retransplanted had the best overall survival whereas patients who were not relisted had the highest mortality ($P < 0.001$; Fig. 3b). The superior overall survival of patients who were relisted was also confirmed in a landmark analysis only including patients who were relisted 1 year after graft loss (Fig. 3c).

Relisting

Only 111/254 patients (43.7%) were relisted, which was defined as registration at Eurotransplant. Relisting was mainly dependent on age. Of the relisted patients, 80 (72.1%) were younger than 55 at graft loss and only 15 (13.5%) were over 65 years of age. The majority of patients was listed within 1 year after allograft failure (77/111 patients, 69.4%; Table 3). Patients with no cPRA before relisting, low cPRA (>0, but not eligible for AM program) before relisting, or patients listed in the AM program did not differ in relisting rates (Fig. S1). A logistic regression analysis for relisting was

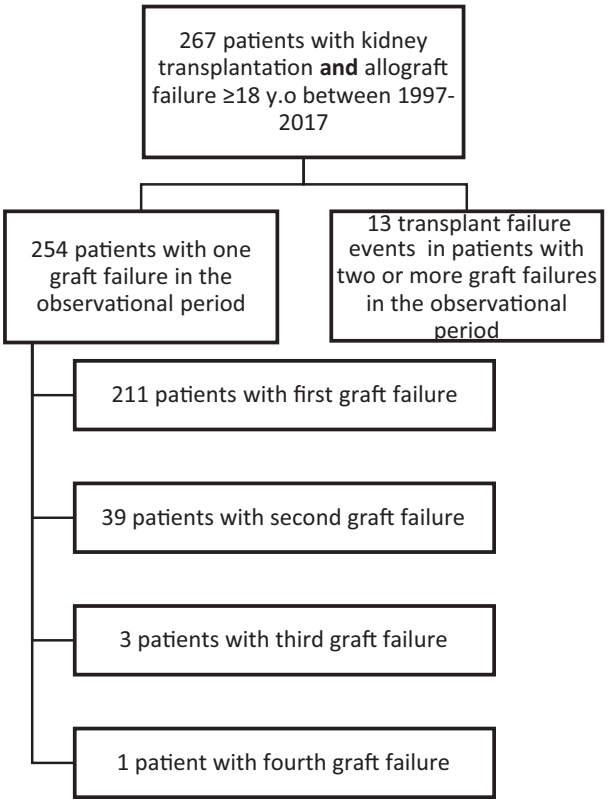

Figure 1 Study population. 267 patients received and lost an allograft in the observational period. Only the first graft loss in the observational period was included in the current analysis.

done including the explanatory variables age at transplant failure, sex, length of previous transplantation, peak cPRA after graft loss (0 or >0), coded reason for graft loss with the categories perioperative, immunologic and nonimmunologic, and the blood type was performed, and the results are reported in Table S1. The age at transplantation is associated with relisting (OR: 0.93, CI: 0.91–0.95). The younger a patient is, the more likely he is relisted. Further, the reason for graft loss is associated with relisting. Patients with a nonimmunologic cause of graft loss were more likely to be relisted compared to patients with an immunologic cause of graft loss (OR: 10.63, CI: 2.59–43.52).

Table 2. Main reasons for graft loss.

Main reasons for graft loss	% (*n*)
Antibody-mediated rejection	22.8 (58)
Medical	20.5 (52)
Multifactorial causes	18.9 (48)
T-cell-mediated rejection	14.2 (36)
Perioperative	8.3 (21)
Recurrent disease	6.3 (16)
Polyoma nephropathy	3.6 (9)
Unknown	3.1 (8)
Other	2.4 (6)

Retransplantation

During the observational period, only 42/254 patients (16.5%) with graft failure received another transplant. Of the 42 patients who received a retransplantation during the observational period, 80.9% were <55 years of

Table 1. Information about patients who lost their graft.

n	Overall cohort 254	1st graft loss 211	2nd graft loss 39
1st transplant [% (*n*)]	83 (211)		
2nd transplant [% (*n*)]	15.3 (39)		
3rd transplant [% (*n*)]	1.2 (3)		
4th transplant [% (*n*)]	0.4 (1)		
Median time to graft loss (IQR)	4.3 (1.2–8.2)	4.3 (1.1–8.2)	4.8 (1.9–7.7)
Median age at graft loss (IQR)	57 (42–69)	60 (46–70)	41 (32–51)
Age in years by category			
18–54 (%)	48.0 (122)	41.2 (87)	82.0 (32)
55–64 (%)	13.8 (35)	15.2 (32)	5.1 (2)
≥65 (%)	38.2 (97)	43.6 (92)	12.8 (5)
Male [% (*n*)]	59 (149)	57 (121)	67 (26)
Living donation [% (*n*)]	20 (51)	24 (50)	3 (1)
Median observation time after graft loss (IQR)	7.4 (3.9–11.4)	7.0 (3.8–9.9)	9.4 (6.6–13.2)
Nephrectomy [% (*n*)]	41 (104)	41 (87)	44 (17)
Relisting (%)	44 (111)	40 (84)	69 (27)
Retransplantation (%)	16.5 (42)	19.4 (41)	2.5 (1)
Median age at retransplantation (IQR)	44.6 (35.6–56.7)	50.2 (40.6–56.5)	38 (32–49.6)
Median time on dialysis in years (IQR)	4.0 (2.4–6.8)	3.9 (2.2–6.4)	6.2 (3.0–7.7)

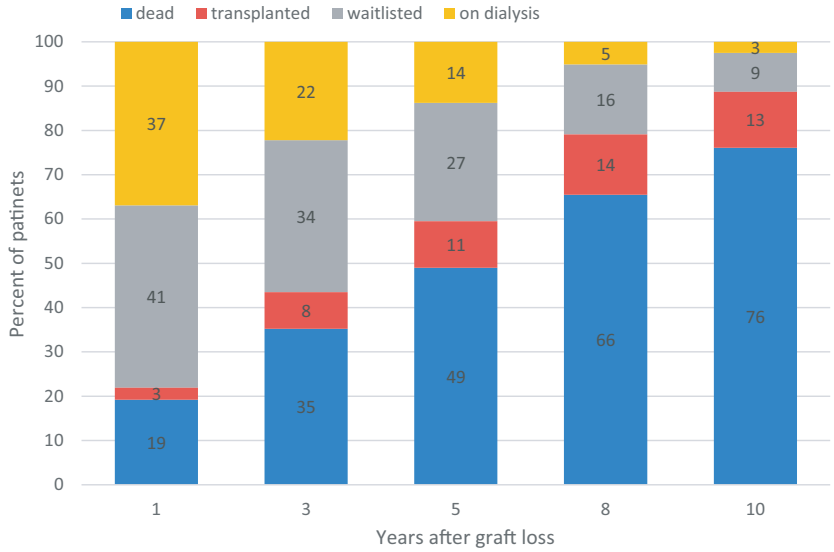

Figure 2 Percentage of patients with a particular status. Percentage of patients with status deceased, transplanted, waitlisted, and on dialysis (but not waitlisted) 1, 3, 5, 8, and 10 years after graft loss. Numbers above indicate number of patients with follow-up up the respective time point.

age at graft loss, 12% were between 55 and 64, and only 7.1% were ≥65 years at time of graft loss. Among the patients who received a retransplantation, 11/42 patients (26.1%) received a living donor kidney.

According to our centers policy, all 15 highly immunized patients were listed within the Eurotransplant AM program based on the respective Eurotransplant rules. Patients with dnDSA not listed within the AM program had to wait a significantly longer time after relisting than patients without dnDSA from the failed graft ($P = 0.024$). Waiting time in patients in the AM program was comparable to patients without dnDSA ($P = 0.447$; Fig. S2). A cox regression was conducted for retransplantation for the subset of relisted patients. The following explanatory variables were used in the model: age at transplant failure (<65 vs. ≥65 years), sex, and immunization status (positive or negative peak cPRA after graft loss or listing within the AM program). The results are reported in Table S2. The model includes 100 relisted patients, of which 33 had a retransplantation and 67 were censored. Patients with positive peak cPRA before relisting had a lower chance for retransplantation compared to patients with negative peak cPRA before relisting (HR: 0.44, CI: 0.19–0.996, $P = 0.049$).

Transplant nephrectomy

In our cohort, 90/254 (35.4%) patients had a transplant nephrectomy within the observational period. Reasons for transplant nephrectomy are summarized in Table 4. Graft intolerance syndrome and rejections were the most common reasons for graft nephrectomy. The median time of transplant nephrectomy after transplant failure was 3.1 months. One year after graft loss, 91.3% of transplant nephrectomies had been performed followed by a few additional nephrectomies up to 3 years after graft loss. Relisted patients who underwent nephrectomy had the same survival (Fig. 4a). Nephrectomy did not affect retransplantation (Fig. 4b).

Immunization

Only 84/247 (34%) patients had dnDSA before graft loss and in another 48 patients (19.4%) dnDSA were detected after allograft loss (Table 5). In 108 patients (43.7%), no dnDSA were detected in the observational period and in 7 patients DSA status after transplant loss was not available.

Of the 132 patients with positive dnDSA, 40.9% of patients had dnDSA only to class II HLA antigens, 21.7% only to class I HLA antigens, and 37.4% to both HLA classes (Table 5).

Immunization status by cPRA before transplantation and after graft loss (peak cPRA) is shown in Table 6. The number of patients with a baseline cPRA of 0 decreased from 69.7% of patients before the transplantation in the observational period to 40.2% of patients after graft loss in the observational period (Class II) whereas the number of highly immunized patients (cPRA > 90) increased from 2% to 9.1% of patients.

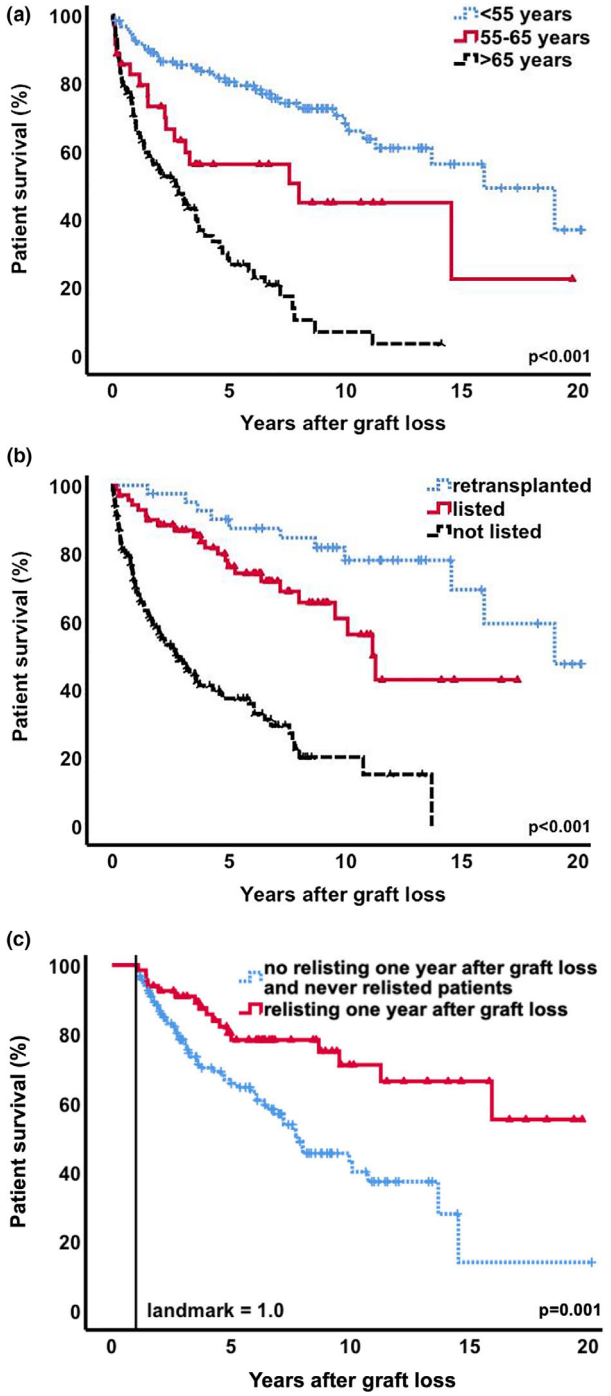

Figure 3 Death after graft loss. (a) According to age. Kaplan–Meier plots illustrating the cumulative incidence of death in patients according to age at graft loss. Patients with higher age at graft loss have a higher mortality. (b) Relisting and retransplantation. Kaplan–Meier plots illustrating the cumulative incidence of death in patients relisted and transplanted, relisted and not transplanted, or not relisted for repeat kidney transplantation showing the highest mortality among non-relisted patients. (c) Relisting. Kaplan–Meier plots with landmark at one year, illustrating the cumulative incidence of death in patients relisted or not relisted for repeat kidney transplantation showing a higher mortality among patients not relisted 1 year after transplantation.

As DSA after graft loss are only relevant for relisted patients, we next analyzed presence of dnDSA in relisted patients, which has the advantage of a complete longitudinal assessment of HLA antibodies due to regular mandatory screening of waitlisted patients. In this cohort, 38 of 111 patients (34.2%) had dnDSA already at time of graft loss, 43 of 111 (38.7%) patients developed dnDSA after graft loss, and 30 of 111 (27.0%) patients did not develop dnDSA during the observational period. Among relisted patients, the distribution of HLA immunization to class I and II was comparable to all patients with graft loss.

Interestingly, patients with nephrectomy had a comparable occurrence of dnDSA compared to patients without nephrectomy (Fig. 5). In 110 patients without dnDSA 1 year after transplant failure, 42 patients (38%) were without immunosuppression, 34% were on monotherapy, and only five were maintained on triple therapy (Table S3). As shown in this table, we could not reveal a major impact of the number of immunosuppressants on the development of dnDSA after graft loss.

PIRCHE-II score and occurrence of dnDSA

The median PIRCHE-II score in our cohort was 69.51 (IQR 37 103). For 16 patients, PIRCHE-II score could not be calculated and they were excluded in this analysis. For further analysis, rounded cohort median was used as a cutoff for classification dividing the cohort in 118 patients with a PIRCHE-II score ≤70 and 119 patients with a score >70. The occurrence of dnDSA was significantly higher in patients with a PIRCHE-II score >70 ($P = 0.001$; Fig. 6). We did not find a correlation of the PRICHE-II score and cPRA at the time point of graft loss (Fig. S3).

Discussion

The present study is to the best of our knowledge the largest study that reports detailed clinical outcomes including retransplantation rates, immunization status with regard to dnDSA, epitope data, and data on transplant nephrectomy in patients after allograft loss. In our single-center analysis, 267 allograft failures in 254 patients were identified over a 20 years period with complete follow-up of key parameters. For 159 patients, even a complete 10-year follow-up was available.

In our cohort, more than one third of patients were over 65 years old at time of graft loss. Patients with their second transplant failure were significantly

Table 3. Relisting according to age.

Age (years)	Proportion of relisted patients (%) *n* = 111	Proportion of retransplanted patients (%) *n* = 42
<55	72.1	80.9
55–64	14.4	12
≥65	13.5	7.1

Table 4. Reasons for transplant nephrectomy.

Reasons for transplant nephrectomy	% (*n*)
Rejection	24.1 (22)
Graft intolerance syndrome	20.8 (19)
Unknown	18.7 (16)
Vascular	14.2 (13)
Infection	13.1 (12)
Malignancy	4.4 (4)
Other	4.4 (4)

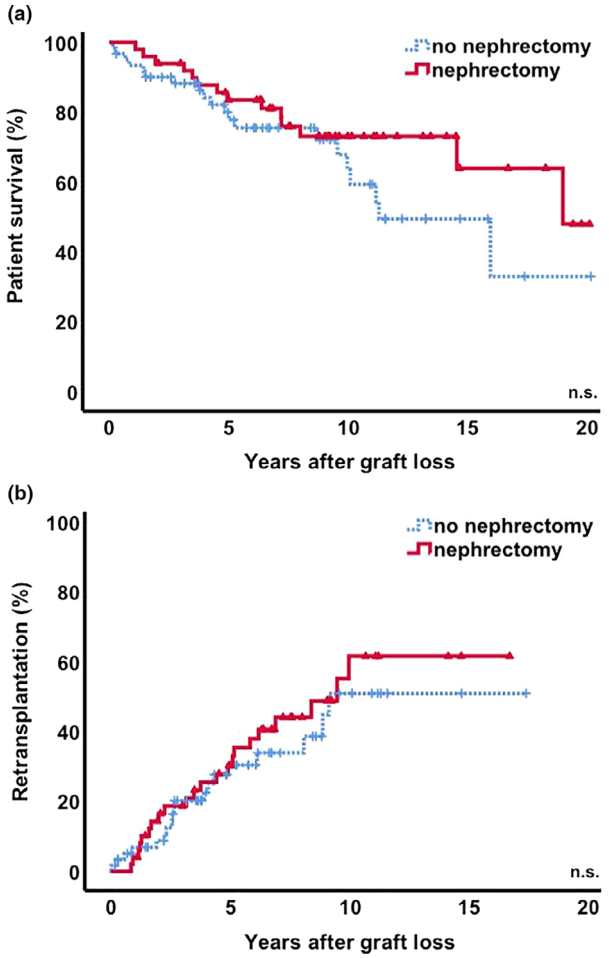

Figure 4 Allograft nephrectomy in relisted patients (*n* = 111). (a) Kaplan–Meier plots illustrating the cumulative incidence of death in the observational period depending on nephrectomy. Relisted patients with and without nephrectomy had the same survival ($P = 0.56$). (b) Kaplan–Meier plots illustrating the cumulative incidence of retransplantation. Nephrectomy did not impact relisting (data not shown) and retransplantation ($P = 0.52$).

younger than patients with their first transplant failure. This observation is in accordance with a recent publication of Assfalg *et al*. [20] who report a significantly younger age for third and fourth kidney transplant recipients and also with previous reports of UNOS/OPTN data [21,22]. Patients after repeated allograft failure represent a group of recipients that get a first transplant early in life and end up with transplant failure earlier in life again. Not surprising, mortality after graft loss mainly depends on age at graft loss. Further, retransplanted patients, followed by relisted patients, show a significantly lower mortality than patients who were not relisted for transplantation. This was an expected result that is still reassuring in terms of our center's transplant evaluation procedure. Patients with a high cumulative risk of death because of high age, cardiovascular risk, malignancies, or other medical conditions [23] are effectively separated from the collective with lower mortality likely experience advantage from retransplantation.

An important finding was the low chance of being relisted or retransplanted if patients lost their allograft aged ≥65 years. Further, in a logistic regression analysis higher age was associated with lower chances for relisting. Patients who get a first kidney transplant in older age very likely only get a kidney transplant once in their life.

Accepting organs with poor organ quality and a short organ survival might not be the right strategy in these patients. In patients ≥65 years, however, a timely transplant is essential, even for the price of lower organ quality. But as shown by our data, by far most patients will have only one chance of being transplanted. Only a small proportion of elderly patients with graft failure gets relisted and just 4% will receive another transplant. The poor chances of retransplantation in combination with high perioperative mortality after acceptance of marginal donors favor the intense search for living donors in those patients [24,25].

The waiting time for a retransplantation in our cohort did depend on the presence of dnDSA before relisting. Patients with dnDSA had a significantly longer time after relisting than patients without, except for patients, who have been listed within the Eurotransplant AM program. Patients with the AM program had a

Table 5. Occurrence of de novo donor-specific antibodies (dnDSA) in the whole cohort (n = 247, for seven individuals dnDSA follow-up was not available) and in relisted patients (n = 111).

	Overall cohort	Relisted patients
n	247	111
No dnDSA in the observational period (%)	108 (43.7)	30 (27.0)
dnDSA before graft failure (%)	84 (34.0)	38 (34.2)
dnDSA after graft failure (%)	48 (19.4)	43 (38.7)
dnDSA to class I (%)	54 (21.7)	23 (20.7)
dnDSA to class II (%)	101 (40.9)	44 (39.6)
dnDSA to class I and II (%)	37.4 (92)	44 (39.6)

waiting time comparable to those without DSA. Thus, patients in the AM program have a double profit from their listing. On the one hand, they have the same waiting time as the unimmunized controls and it has been shown that due to the excellent match, AM patients have a better 10-year graft survival compared to highly sensitized patients transplanted on the basis of avoidance of unacceptable mismatches [7]. The current allocation system leads to a discrimination of patients with a certain but not massive immunization. This has already been addressed by Ziemann *et al.* [13], who found that the waiting time is extended by 1.3 weeks if the PRA value is increased by 1% within the regular allocation and prolonged by 5 weeks within ESP. The cPRA did not impact relisting but did affect retransplantation in our cox regression model.

Currently, it is a matter of ongoing debate whether transplant nephrectomy has an impact on the development of dnDSA. The so far conducted studies are retrospective cohort studies with relatively small numbers [8,9]. Most studies report a relation between transplant nephrectomy and short-term development of dnDSA [26–28]. Nephrectomy in our cohort occurred in 35.4% of cases. A recent analysis from a Swiss cohort [5] reported a much higher rate of nephrectomies (49 nephrectomies in 77 patients with graft loss). In a large US cohort, the number of nephrectomies was described to be 31.5% 1.6 years after graft failure and was associated with a survival benefit of patients in the nephrectomy group [6]. In the present study, nephrectomy had no impact on the development of dnDSA. We focused only on the long-term effects in relisted patients since they have an assured HLA follow-up every 3 months. Almost one third of relisted patients (27%) with complete HLA antibody follow-up did not develop dnDSA at any time within the observational period, while 34.2% already had dnDSA before graft loss. As reported previously, patients without nephrectomy also may develop dnDSA over time (e.g., when immunosuppression is stopped). As a consequence, our study did not find any measurable and clinically meaningful effect of transplant nephrectomy on waiting times.

Although it is frequently reported in the literature that antibody-mediated rejection (ABMR) is a major cause of graft loss [29,30], according to our data it can maximally account for around one third of overall graft losses since the presence of DSA is prerequisite for the detection of ABMR [31]. When dnDSA are detected and a protocol biopsy is performed, ABMR is also only detected in 40% of cases [32]. As previously reported, the development of dnDSA is a risk factor for the development graft failure [15,33].

To estimate the risk for post-transplant dnDSA, simply counting the number of mismatches is an effective method to predict the development of dnDSA. However, this method can be further refined by assessing the immunogenic potential of individual HLA mismatches [34] and by considering epitopes that are present in mismatched HLA and which are involved in T-cell-mediated alloimmune responses [35]. It has been shown that it is able to predict the occurrence of dnDSA in kidney transplant recipients [15] and also after transplant nephrectomy [36]. In our cohort, the median PIRCHE score was 69.5 and patients with a PIRCHE score above this score were more likely to develop dnDSA confirming the predictive power of this tool.

The limitations of this study are its retrospective design in a single center with a limited number of patients and incomplete assessment of variables of potential interest such as blood transfusions. The retrospective designs of this study cannot show causation, but only associations. This single-center experience may not extrapolate well to other centers or other country populations.

In summary, we found within this single-center study that patients after graft loss have a high mortality when not relisted. The chance of retransplantation is very low when graft loss occurs ≥65 years of age. The presence of dnDSA does not negatively impact relisting but it does

Table 6. Baseline (before transplantation in the observational period) and peak cPRA after graft loss in the observational period.

	cPRA Class I baseline [% (*n*)]	cPRA Class II baseline [% (*n*)]	cPRA Class I peak [% (*n*)]	cPRA Class II peak [% (*n*)]
cPRA 0	63.4 (161)	69.7 (177)	42.1 (107)	40.2 (102)
cPRA 1–30	8.7 (22)	2.0 (5)	15.7 (40)	11.0 (28)
cPRA 30–60	3.1 (8)	2.4 (6)	9.1 (23)	12.6 (32)
cPRA 60–90	4.3 (11)	5.5 (14)	14.6 (37)	16.9 (43)
cPRA >90	2.0 (5)	5.0 (5)	8.3 (21)	9.1 (23)
cPRA NA	18.5 (47)	18.5 (47)	10.2 (26)	10.2 (26)

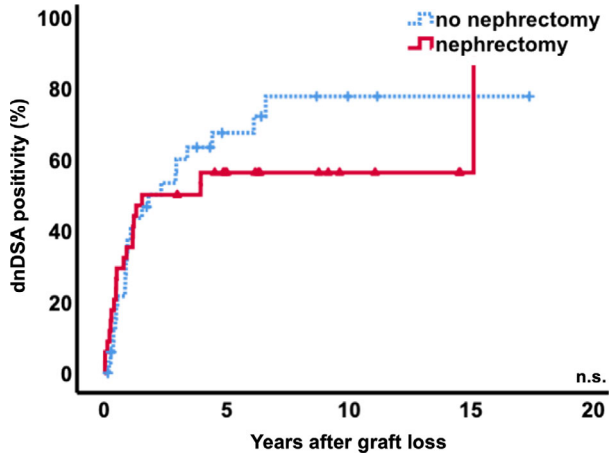

Figure 5 Transplant nephrectomy and de novo donor-specific antibodies (dnDSA) of relisted patients without preformed dnDSA at time of graft loss. Kaplan–Meier plots illustrating the cumulative incidence of dnDSA depending on nephrectomy or no nephrectomy. Nephrectomy and no nephrectomy group do not significantly differ in the development of dnDSA ($P = 0.3$).

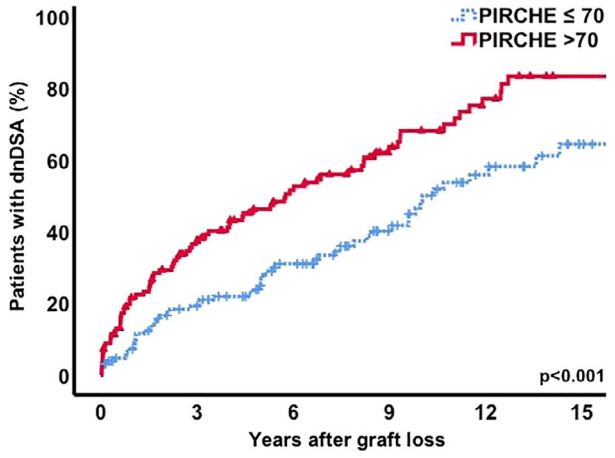

Figure 6 PIRCHE (Predicted Indirectly ReCognizable HLA Epitopes) - score. Kaplan–Meier plots illustrating the cumulative incidence of de novo donor-specific antibodies (dnDSA) stratified by a PIRCHE score ≤70 and >70.

impact waiting time until retransplantation. Transplant nephrectomy in our cohort did not impact the development of dnDSA questioning previous retrospective studies. The Eurotransplant AM program currently favors highly sensitized patients, which should lead to a close examination whether patients are eligible to be listed within AM, while generally measured by cPRA is associated with longer waiting times. In addition, in the current study, epitope matching has shown to predict alloimmunization. Therefore, this could be a strategy to improve matching algorithms as a strategy to prevent alloimmunization minimizing the group of moderately immunized patients with long waiting times after graft loss.

Authorship

ES: wrote the paper and analyzed data. LJL: wrote the paper and analyzed data. MM: collected data. MMay: collected data and performed research/study. WD: analyzed data. MGN: analyzed data. FF: designed research/study. LL: designed research/study. MP: analyzed data. FF: performed research/study. DS: contributed important reagents. NL: contributed important reagents. KB: wrote the paper, designed research/study. FH: wrote the paper, designed research/study.

Funding

The authors have declared no funding.

Conflict of interest

The authors have declared no conflicts of interest.

SUPPORTING INFORMATION

Additional supporting information may be found online in the Supporting Information section at the end of the article.

Table S1. Logistic regression for relisting.

Table S2. Cox regression for retransplantation.

Table S3. Immunosuppressive regimens of 110 patients without dnDSA at time of graft loss 1 year after graft failure.

Figure S1. Relationship of cPRA and relisting over time.

Figure S2. Relationship of dnDSA and relisting over time.

Figure S3. Correlation of cPRA Class I. cPRA Class II and PIRCHE score.

REFERENCES

1. Heaphy EL, Poggio ED, Flechner SM, *et al.* Risk factors for retransplant kidney recipients: relisting and outcomes from patients' primary transplant. *Am J Transplant* 2014; **14**: 1356.
2. Hernandez D, Muriel A, Castro de la Nuez P, *et al.* Survival in Southern European patients waitlisted for kidney transplant after graft failure: a competing risk analysis. *PLoS One* 2018; **13**: e0193091.
3. Kaplan B, Meier-Kriesche HU. Death after graft loss: an important late study endpoint in kidney transplantation. *Am J Transplant* 2002; **2**: 970.
4. Meier-Kriesche HU, Kaplan B. Death after graft loss: a novel endpoint for renal transplantation. *Transplant Proc* 2001; **33**: 3405.
5. Bonani M, Achermann R, Seeger H, *et al.* Dialysis after graft loss: the Swiss experience. *Nephrol Dial Transpl* 2018; **33**: 2182.
6. Ayus JC, Achinger SG, Lee S, Sayegh MH, Go AS. Transplant nephrectomy improves survival following a failed renal allograft. *J Am Soc Nephrol* 2010; **21**: 374.
7. Heidt S, Haasnoot GW, van Rood JJ, Witvliet MD, Claas FHJ. Kidney allocation based on proven acceptable antigens results in superior graft survival in highly sensitized patients. *Kidney Int* 2018; **93**: 491.
8. Lucisano G, Brookes P, Santos-Nunez E, *et al.* Allosensitization after transplant failure: the role of graft nephrectomy and immunosuppression – a retrospective study. *Transpl Int* 2019; **32**: 949.
9. Nimmo A, McIntyre S, Turner DM, Henderson LK, Battle RK. The impact of withdrawal of maintenance immunosuppression and graft nephrectomy on HLA sensitization and calculated chance of future transplant. *Transplant Direct* 2018; **4**: e409.
10. Wong G, Chua S, Chadban SJ, *et al.* Waiting time between failure of first graft and second kidney transplant and graft and patient survival. *Transplantation* 2016; **100**: 1767.
11. Kosmoliaptsis V, Gjorgjimajkoska O, Sharples LD, *et al.* Impact of donor mismatches at individual HLA-A, -B, -C, -DR, and -DQ loci on the development of HLA-specific antibodies in patients listed for repeat renal transplantation. *Kidney Int* 2014; **86**: 1039.
12. Lachmann N, Schonemann C, El-Awar N, *et al.* Dynamics and epitope specificity of anti-human leukocyte antibodies following renal allograft nephrectomy. *Nephrol Dial Transplant* 2016; **31**: 1351.
13. Ziemann M, Hessler N, Konig IR, *et al.* Unacceptable human leucocyte antigens for organ offers in the era of organ shortage: influence on waiting time before kidney transplantation. *Nephrol Dial Transplant* 2017; **32**: 880.
14. Larkins NG, Wong G, Taverniti A, Lim WH. Epitope matching in kidney transplantation: recent advances and current limitations. *Curr Opin Organ Transplant* 2019; **24**: 370.
15. Lachmann N, Niemann M, Reinke P, *et al.* Donor-recipient matching based on predicted indirectly recognizable HLA epitopes independently predicts the incidence of de novo donor-specific HLA antibodies following renal transplantation. *Am J Transplant* 2017; **17**: 3076.
16. Schröter K, Lindemann Trzebiatowski G, Fritsche L. TBase – An internet-based transplantation database. In: *Workshop on Concurrency, Specification and Programming, CS&P*. vol. September 28–30, 1998 (110), 1998; 1998: pp. 214.
17. Huber L, Lachmann N, Niemann M, *et al.* Pretransplant virtual PRA and long-term outcomes of kidney transplant recipients. *Transpl Int* 2015; **28**: 710.
18. Liefeldt L, Brakemeier S, Glander P, *et al.* Donor-specific HLA antibodies in a cohort comparing everolimus with cyclosporine after kidney transplantation. *Am J Transplant* 2012; **12**: 1192.
19. Lachmann N, Terasaki PI, Budde K, *et al.* Anti-human leukocyte antigen and donor-specific antibodies detected by luminex posttransplant serve as biomarkers for chronic rejection of renal allografts. *Transplantation* 2009; **87**: 1505.
20. Assfalg V, Selig K, Tolksdorf J, *et al.* Repeated kidney re-transplantation-the Eurotransplant experience: a retrospective multicenter outcome analysis. *Transpl Int* 2020; **33**: 617.
21. Redfield RR, Gupta M, Rodriguez E, Wood A, Abt PL, Levine MH. Graft and patient survival outcomes of a third kidney transplant. *Transplantation* 2015; **99**: 416.
22. Khalil AK, Slaven JE, Mujtaba MA, *et al.* Re-transplants compared to primary kidney transplants recipients: a mate kidney paired analysis of the OPTN/UNOS database. *Clin Transplant* 2016; **30**: 566.
23. Virmani S, Asch WS. The role of the general nephrologist in evaluating patients for kidney transplantation: core curriculum 2020. *Am J Kidney Dis* 2020; **76**: 567.
24. Rose C, Schaeffner E, Frei U, Gill J, Gill JS. A lifetime of allograft function with kidneys from older donors. *J Am Soc Nephrol* 2015; **26**: 2483.
25. Gill JS, Schaeffner E, Chadban S, *et al.* Quantification of the early risk of death in elderly kidney transplant recipients. *Am J Transplant* 2013; **13**: 427.
26. Billen EV, Christiaans MH, Lee J, van den Berg-Loonen EM. Donor-directed HLA antibodies before and after transplantectomy detected by the luminex single antigen assay. *Transplantation* 2009; **87**: 563.
27. Adeyi OA, Girnita AL, Howe J, *et al.* Serum analysis after transplant nephrectomy reveals restricted antibody specificity patterns against structurally defined HLA class I mismatches. *Transpl Immunol* 2005; **14**: 53.
28. Del Bello A, Congy-Jolivet N, Sallusto F, *et al.* Donor-specific antibodies after ceasing immunosuppressive therapy, with or without an allograft nephrectomy. *Clin J Am Soc Nephrol* 2012; **7**: 1310.
29. Mengel M, Sis B, Haas M, *et al.* Banff 2011 meeting report: new concepts in antibody-mediated rejection. *Am J Transplant* 2012; **12**: 563.

30. Schinstock CA, Mannon RB, Budde K, *et al*. Recommended treatment for antibody-mediated rejection after kidney transplantation: the 2019 expert consensus from the transplantion society working group. *Transplantation* 2020; **104**: 911.
31. Haas M. Evolving criteria for the diagnosis of antibody-mediated rejection in renal allografts. *Curr Opin Nephrol Hypertens* 2018; **27**: 137.
32. Bertrand D, Gatault P, Jaureguy M, *et al*. Protocol biopsies in patients with subclinical de novo DSA after kidney transplantation: a multicentric study. *Transplantation* 2020; **104**: 1726.
33. Loupy A, Hill GS, Jordan SC. The impact of donor-specific anti-HLA antibodies on late kidney allograft failure. *Nat Rev Nephrol* 2012; **8**: 348.
34. Claas FH, Dankers MK, Oudshoorn M, *et al*. Differential immunogenicity of HLA mismatches in clinical transplantation. *Transpl Immunol* 2005; **14**: 187.
35. Geneugelijk K, Thus KA, Spierings E. Predicting alloreactivity in transplantation. *J Immunol Res* 2014; **2014**: 159479.
36. Otten HG, Calis JJ, Kesmir C, van Zuilen AD, Spierings E. Predicted indirectly recognizable HLA epitopes presented by HLA-DR correlate with the de novo development of donor-specific HLA IgG antibodies after kidney transplantation. *Hum Immunol* 2013; **74**: 290.