Transplant International

ORIGINAL ARTICLE

Pre-transplant donor-specific HLA antibodies and risk for poor first-year renal transplant outcomes: results from the Swiss Transplant Cohort Study

Caroline Wehmeier[1], Patrizia Amico[1], Daniel Sidler[2], Urs Wirthmüller[3], Karine Hadaya[4], Sylvie Ferrari-Lacraz[5], Déla Golshayan[6], Vincent Aubert[7], Aurelia Schnyder[8], Kata Sunic[9], Thomas Schachtner[10], Jakob Nilsson[11], Stefan Schaub[1] & the Swiss Transplant Cohort Study*

1 Clinic for Transplantation Immunology and Nephrology, University Hospital Basel, Basel, Switzerland
2 Department of Nephrology and Hypertension, Inselspital, Berne University Hospital and University of Berne, Berne, Switzerland
3 Department of Laboratory Medicine, Inselspital, Berne University Hospital and University of Berne, Berne, Switzerland
4 Divisions of Nephrology and Transplantation, Geneva University Hospitals, Geneva, Switzerland
5 Transplantation Immunology Unit, Service of Immunology and Allergy and Service of Laboratory Medicine, Geneva University Hospital and Medical School, Geneva, Switzerland
6 Transplantation Center, Lausanne University Hospital, Lausanne, Switzerland
7 Division of Immunology and Allergy, Lausanne University Hospital, Lausanne, Switzerland
8 Department of Nephrology and Transplantation Medicine, Kantonsspital St. Gallen, St. Gallen, Switzerland
9 HLA Laboratory, Blutspende Schweizerisches Rotkreuz Ostschweiz, St. Gallen, Switzerland
10 Division of Nephrology, University Hospital Zurich, Zurich, Switzerland
11 Department of Immunology, University Hospital Zurich, Zurich, Switzerland

SUMMARY

The aim of this study was to analyze first year renal outcomes in a nationwide prospective multicenter cohort comprising 2215 renal transplants, with a special emphasis on the presence of pre-transplant donor-specific HLA antibodies (DSA). All transplants had a complete virtual crossmatch and DSA were detected in 19% (411/2215). The investigated composite endpoint was a poor first-year outcome defined as (i) allograft failure or (ii) death or (iii) poor allograft function (eGFR $\leq$25 ml/min/1.73 m^2) at one year. Two hundred and twenty-one (221/2215; 10%) transplants showed a poor first-year outcome. Rejection (24/70; 34%) was the most common reason for graft failure. First-year patient's death was rare (48/2215; 2%). There were no statistically significant differences between DSA-positive and DSA-negative transplants regarding composite and each individual endpoint, as well as reasons for graft failure and death. DSA-positive transplants experienced more frequently rejection episodes, mainly antibody-mediated rejection (both $P < 0.0001$). The combination of DSA and any first year rejection was associated with the overall poorest death-censored allograft survival ($P < 0.0001$). In conclusion, presence of pre-transplant DSA per se does not affect first year outcomes. However, DSA-positive transplants experiencing first year rejection are a high-risk population for poor allograft survival and may benefit from intense clinical surveillance.

Transplant International 2021; 34: 2755–2768

Key words
allograft failure, allograft loss, donor-specific HLA antibodies, DSA, rejection, renal transplantation

Received: 22 July 2021; Revision requested: 11 September 2021; Accepted: 21 September 2021; Published online: 13 October 2021

doi:10.1111/tri.14119

Correspondence
Caroline Wehmeier MD, Clinic for Transplantation Immunology and Nephrology, University Hospital Basel, Petersgraben 4, 4031 Basel, Switzerland.
Tel.: 0041 61 328 6730;
fax: 0041 61 2652410;
e-mail: caroline.wehmeier@usb.ch

*The members of the STCS are indicated in the Acknowledgments.

Introduction

Over the last decades, better pre- and post-transplant management of renal allograft recipients translated into substantially improved renal transplant outcomes [1]. This progress is most clearly reflected by the short-term transplant success with long-term outcomes currently stagnating [2–4], which is generally shifting the research focus beyond the first year post-transplant. However, the first year is the most critical and complications during this period can imprint the subsequent post-transplant course by impairing the functional reserve of the transplanted organ [5–7]. Therefore, it is still necessary to analyze outcomes of this period in more detail. Interestingly, one recent study by Coemans *et al.* reported a deceleration of short-term improvement in allograft survival over the past 20 years in Europe, even after accounting for changing recipient and donor characteristics [5]. This is an interesting finding in light of concurrent advances in immunological risk assessment by the introduction of single-antigen bead technology that enabled highly sensitive detection and assignment of pre-transplant donor-specific antibodies (DSA). While these antibodies have clearly shown to be a risk factor for antibody-mediated rejection and are associated with a detrimental effect on long-term allograft survival [8–10], their short-term impact has been less well characterized.

Only few studies have so far analyzed first-year renal transplant outcomes in more detail. Most of these studies either focused on the very early post-transplant period up to three months or did not take the presence of pre-transplant DSA into account [11–14]. In a recent cohort study from Finland, an increased level of panel-reactive antibodies was identified as an independent risk factor for allograft failure within the first year [13]. However, it is a matter of debate whether the level of panel-reactive antibodies confers an immunological risk in the absence of DSA, which pinpoints that the influence of DSA should be investigated independently [10,15–18].

For this study, we analyzed the data from the Swiss Transplant Cohort Study, a nationwide cohort project including >90% of all Swiss renal transplants performed since 2008 and providing detailed data on pre-transplant DSA. Our aim was to investigate first-year renal outcomes in this contemporary cohort of renal allograft recipients with a special emphasis on the presence of pre-transplant DSA.

Material and methods

Study design and data collection

This study (project number FUP142) was conducted within the framework of the Swiss Transplant Cohort Study (STCS). The STCS is a nationwide multicenter observational cohort of solid organ recipients that has been approved by the ethics committees of all Swiss transplant centers. All patients gave their written informed consent for participation. Since this study is a nested project of the STCS, it was separately approved by the ethics committee of Northwestern and Central Switzerland (www.eknz.ch; project-ID 2019-02122).

Since 2008, the STCS prospectively collects detailed patient- and transplant-specific data at the time of transplantation as well as at month 6, at month 12 and yearly thereafter by using standardized case report forms.

Patient population

Between May 2008 and December 2017, 2874 kidney transplants were performed in Switzerland. Of those, 2647 kidney transplants participating in the STCS and having complete data sets were eligible for study inclusion (Fig. 1). In total, 432 transplants (16%) were excluded for the reasons as detailed in Fig. 1. ABO incompatible transplants were not considered due to the distinct immunological risk as well as possible misclassification of rejection resulting from almost universal C4d positivity in peritubular capillaries of allograft biopsies. The final study population consisted of 2215 kidney transplants in 2179 patients. Only 35 patients (2%) received more than one transplant within the study period. The median follow-up of the study population was 4.4 (2.1–7.0) years.

Transplant International 2021; 34: 2755–2768

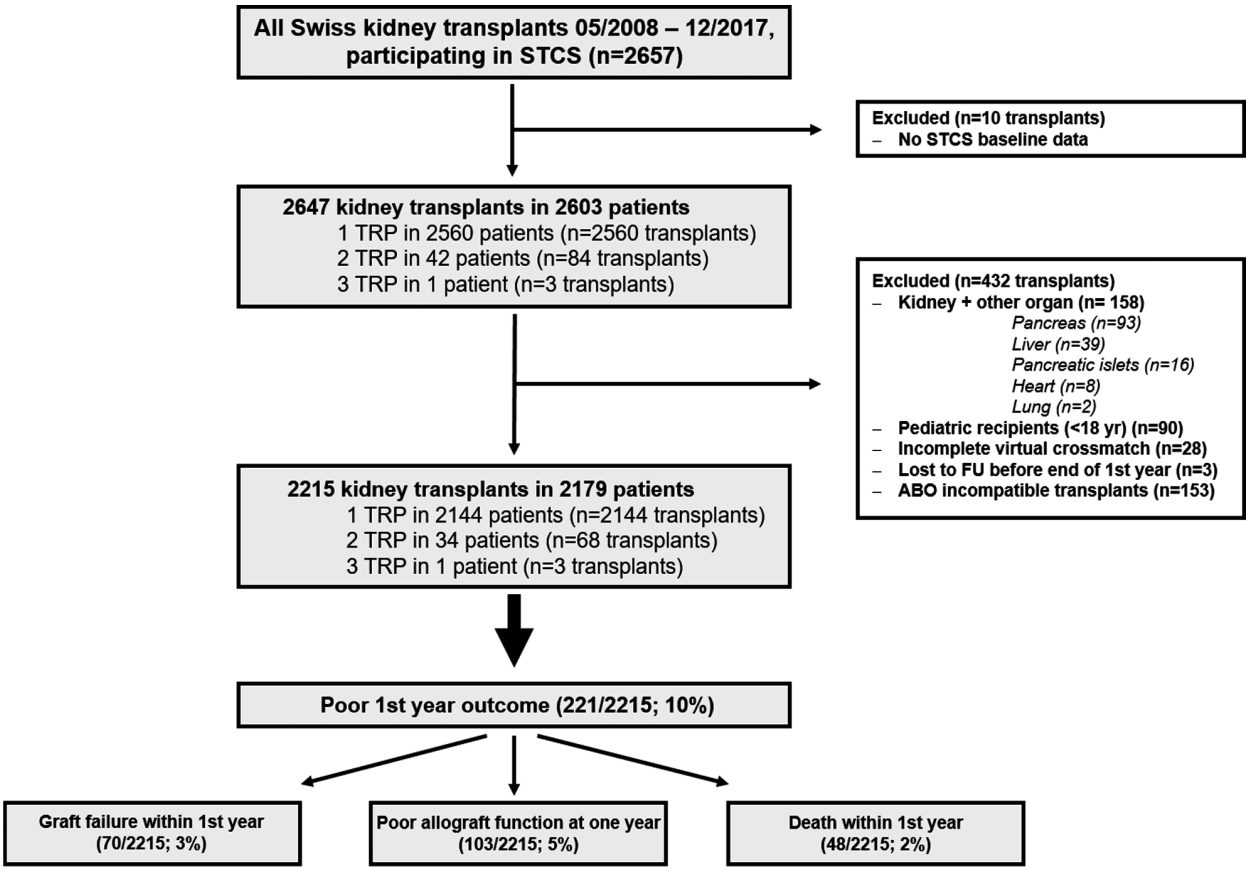

Figure 1 Study flowchart. Abbreviations: FU, follow-up; STCS, Swiss Transplant Cohort Study; TRP, transplantation.

Investigated outcomes and definitions

The investigated composite endpoint was a poor first year outcome, which was defined as follows:

i Allograft failure within the first year post-transplant or

ii Poor allograft function defined as eGFR ≤25 ml/min/1.73 m^2 at the end of the first year or

iii Death of the patient within the first year post-transplant

For definition of poor allograft function at the end of the first year, we used the creatinine that was recorded at the time of the follow-up visit at month 12. The estimated glomerular filtration rate (eGFR) was calculated by using the Chronic Kidney Disease Epidemiology Collaboration (CKD-EPI) equation. The eGFR cutoff for poor allograft function was arbitrarily defined based on our clinical experience.

If a patient reached more than one of the defined endpoints by the end of the first year post-transplant, namely experienced graft loss and died later on, only the first event was considered for analysis. Long-term outcomes were followed until December 2019 (censoring).

Assignment of reasons for graft loss and patient's death

Graft loss was defined as return to dialysis or preemptive re-transplantation before dialysis was needed. All allograft losses, as well as all patient deaths, were attributed to specific causative groups. In case of multiple suspected causes or ambiguity, all available medical records as well as histology results were taken into consideration and the most likely dominant cause was assigned in accordance with the treating physician's opinion at the respective transplant center.

Allograft biopsies and definition of rejection

Only biopsy-proven rejection was considered. Allograft biopsies (both surveillance and indication) were performed according to the local protocol and the judgement of the treating physician at the six Swiss

transplant centers. In the database, biopsy results were either recorded by the individual Banff scores (e.g. t, i, ptc, g, etc.) or stored as text, which we translated into Banff scores (e.g. moderate capillaritis translated into ptc2) to obtain a Banff-defined diagnosis. Rejection was defined according to the 2015 Banff criteria [19]. The Banff phenotypes "Borderline changes" and "C4d staining without evidence of rejection" were not considered as rejection in our analysis.

DSA assignment by virtual crossmatch

Pre-transplant DSA were assigned by virtual crossmatching (i.e. comparison of the donor's HLA typing with the recipient's HLA antibody specificities). Both historical and current HLA antibodies with a mean fluorescence intensity (MFI) > 500–1000 (depending on the center-specific cutoff) were included. All transplants had negative complement-dependent cytotoxicity crossmatches.

Presence of HLA antibodies was in 99.5% determined by Luminex bead-based multiplexing technology from one vendor. While some HLA laboratories directly used single-antigen bead (SAB) technology (LABScreen Single Antigen; OneLambda), other first screened by using mixed beads (LABScreen Mixed, OneLambda) and, if positive, subsequently performed SAB testing.

HLA typing was performed by using sequence-specific oligonucleotide (SSO) and sequence-specific primer (SSP) technologies. In order to enable a complete virtual crossmatch, HLA typing of all loci against which HLA antibody specificities in the recipient's serum were detected was required (e.g. a Cw6 antibody in the recipient's serum required typing of the donor's HLA C locus). If not already available, such data were retrospectively added to the database in 2018.

Statistical analysis

Data were analyzed using JMP Version 14 software (SAS institute Inc., Cary, NC, USA). For visualization of data, we used GraphPad Prism Version 8 (GraphPad Software, San Diego, CA, USA). Categorical data are presented as counts and/or percentages and were analyzed by Pearson's chi-square test. Continuous data are shown as median and interquartile ranges (IQR) and compared by Wilcoxon rank sum tests. Survival curves were generated by the Kaplan-Meier method, and the groups compared using the log-rank test. Multivariable logistic regression was used to investigate independent predictors for rejection among pre-transplant DSA-positive transplants. Receiver operating characteristic (ROC) statistics were applied to describe the relationship of both cumulative and dominant DSA MFI with occurrence of any first-year rejection. For all tests, a (two-tailed) P-value < 0.05 was considered to indicate statistical significance.

Results

Study population characteristics

Among the study population consisting of 2215 kidney transplants, 411 (19%) had pre-transplant DSA. Table 1 summarizes the baseline characteristics of the study population, stratified by the presence of pre-transplant DSA. As expected, DSA-positive transplants were more often females and had more frequently previous transplants. In addition, these patients underwent more often hemodialysis before transplantation and primarily received a deceased donor transplant from a slightly younger donor. While HLA class I mismatches were equally distributed among both groups, pre-transplant DSA-positive transplants had more DRB1 mismatches. However, the median number of HLA A/B/DRB1 mismatches did not differ.

In this contemporary transplant cohort, the majority of transplant recipients received a maintenance immunosuppression consisting of a calcineurin inhibitor, mycophenolic acid, and prednisone (97% and 95%, respectively). In DSA-positive transplants, tacrolimus was more frequently applied compared with cyclosporine than in DSA-negative transplants. Two-third of transplants with DSA were treated with antithymocyte globulin or thymoglobuline +/− intravenous immunoglobulin as an induction therapy.

Frequency of investigated outcomes in the first year post-transplant

In total, 221 transplants (10%) had a poor first-year outcome (Fig. 1). Allograft failure occurred in 70 transplants (3%) and 103 transplants (5%) showed a poor allograft function at one year. Death with a functioning graft within the first year was overall rare and occurred in only 48 patients (2%). Interestingly, the frequency of neither the composite endpoint nor any individual outcome was significantly different between transplants with and without pre-transplant DSA (Table 2). This did not change when we considered transplants having historical but no current DSA as DSA-negative (data not shown).

Transplant International 2021; 34: 2755–2768

Table 1. Recipient, donor, and transplant characteristics in transplants with and without pre-transplant donor-specific HLA antibodies (DSA).

	DSA (n = 411)	No DSA (n = 1804)	P-value
Age at transplantation, years	54 (44–61)	55 (44–63)	0.04
Female gender	197 (48%)	609 (34%)	<0.0001
Underlying renal disease			
Glomerulonephritis	92 (22%)	449 (25%)	<0.0001
ADPKD	75 (18%)	341 (19%)	
Diabetic nephropathy	28 (7%)	161 (9%)	
Vascular nephropathy	30 (7%)	222 (12%)	
Interstitial nephropathy	10 (3%)	63 (4%)	
Other	100 (24%)	403 (22%)	
Not specified	58 (14%)	211 (12%)	
Reflux/Pyelonephritis	18 (4%)	93 (5%)	
Hereditary (not ADPKD)	11 (3%)	57 (3%)	
Congenital	13 (3%)	42 (2%)	
Unknown	76 (19%)	165 (9%)	
Dialysis modality			
Hemodialysis	318 (77%)	1245 (69%)	0.006
Peritoneal dialysis	46 (12%)	246 (14%)	
Preemptive	47 (11%)	310 (17%)	
Unknown	0	3 (0%)	
Previous transplants			
Any organ	181 (44%)	221 (12%)	<0.0001
Previous kidney transplants	176 (43%)	186 (10%)	<0.0001
HLA mismatches			
A, % with 0/1/2	14/47/39	16/46/38	0.80
B, % with 0/1/2	6/41/53	9/40/51	0.18
DRB1, % with 0/1/2	10/56/34	18/53/29	0.002
A/B/DRB1 total	4 (3–5)	4 (3–5)	0.08
Cytomegalovirus status			
Low risk (D−/R−)	64 (16%)	346 (19%)	0.06
Intermediate risk (R+)	277 (67%)	1091 (61%)	
High risk (D+/R−)	65 (16%)	349 (19%)	
Unknown	5 (1%)	18 (1%)	
Epstein-Barr-Virus status			
Low risk (D−/R−)	3 (1%)	16 (1%)	0.049
Intermediate risk (D+/R+)	402 (98%)	1712 (95%)	
High risk (D+/R−)	6 (1%)	62 (3%)	
Unknown	0	14 (1%)	
Deceased donor (DD)	294 (72%)	1118 (62%)	0.0003
Donor age, years	53 (41–62)	55 (45–64)	0.016
Detailed deceased donor type			
Donation after brain death	271 (66%)	1008 (56%)	0.43
Donation after circulatory death	23 (6%)	107 (6%)	
Unknown	0	3 (0%)	
Double kidney transplants	3 (1%)	35 (2%)	0.09
Cold ischemia time (DD), hours	9.0 (6.9–12.1)	9.4 (7.3–12.3)	0.10
Maintenance therapy			
FK-MPA-Pred	354 (86%)	1364 (75%)	<0.0001
CyA-MPA-Pred	44 (11%)	357 (20%)	
CNI-based other	10 (2%)	19 (1%)	
mTOR-containing	2 (1%)	48 (3%)	
Other	1 (0%)	14 (1%)	
Unknown	0	0	

Table 1. Continued.

	DSA (n = 411)	No DSA (n = 1804)	P-value
Induction therapy			
ATG/Thymo +/− IvIg	275 (67%)	274 (15%)	<0.0001
Basiliximab	135 (33%)	1473 (82%)	
None	1 (0%)	57 (3%)	

ADPKD, autosomal-dominant polycystic kidney disease; ATG, anti-thymocyte globulin; CNI, calcineurin inhibitor; CyA, cyclosporine; D, donor; DD, deceased donor, FK, tacrolimus; DSA, donor-specific HLA antibodies; IvIg, intravenous immunoglobulinMPA, mycophenolic acid, Pred, prednisone; mTOR, mammalian target of rapamycin inhibitors (sirolimus or everolimus); R, recipient; Thymo, thymoglobuline.

Table 2. Frequency of investigated outcomes in transplants with and without pre-transplant donor-specific HLA antibodies (DSA).

	DSA (n = 411)	No DSA (n = 1804)	P-value
Investigated outcomes			
Composite outcome	45 (11%)	176 (10%)	0.47
Allograft failure within first year	18 (4%)	52 (3%)	0.12
Poor allograft function at one year	14 (3%)	89 (5%)	0.18
Death within first year	13 (3%)	35 (2%)	0.12
Reasons for allograft failure within first year	n = 18	n = 52	
Rejection	7 (39%)	17 (33%)	0.77
Vascular or surgical	5 (27%)	18 (34%)	
Ischemia-reperfusion injury/graft quality	1 (6%)	8 (15%)	
Recurrence of underlying disease	2 (11%)	3 (6%)	
BKV nephropathy	1 (6%)	1 (2%)	
Other	2 (11%)	5 (10%)	
Reasons for death within first year	n = 13	n = 35	
Infection	5 (38%)	13 (37%)	0.79
Unobserved, no autopsy	4 (30%)	5 (14%)	
Cardiovascular	0	5 (14%)	
Gastrointestinal	1 (8%)	4 (11%)	
Malignancy	1 (8%)	3 (9%)	
Suicide	1 (8%)	2 (6%)	
Trauma	0	1 (3%)	
Cerebrovascular	0	1 (3%)	
Unknown	1 (8%)	1 (3%)	

BKV, BK polyomavirus.

In order to confirm that our arbitrarily defined cut-off for poor allograft function at one year indeed affected the longevity of the transplanted organ, we investigated long-term outcomes in these transplants. As illustrated in Figure S1, allografts with an eGFR ≤25 ml/min/1.73 m^2 at one year had a significantly inferior graft survival compared with allografts that did not reach this endpoint (5-year graft survival 48% versus 85%; $P < 0.0001$). This was mainly driven by a significantly worse death-censored graft survival (5-year death-censored graft survival 59% versus 93%; $P < 0.0001$). However, poor allograft function at one year was also associated with a negative impact on patient survival (5-year patient survival 81% versus 91%; $P = 0.005$).

Reasons for allograft failure within the first year

The reasons for allograft failure over the course of the first year post-transplant in pre-transplant DSA-positive

Transplant International 2021; 34: 2755–2768

and DSA-negative transplants are summarized in Fig. 2a. Overall, rejection processes (34%), vascular or surgical problems (33%), and ischemia-reperfusion injury/graft quality (13%) were the most frequently identified reasons for allograft failure. The majority of events occurred within the first six months post-transplant (57/70, 81%). Primary non-function of the transplanted kidney accounted for 32/70 (46%) of allograft failures. The dominant causes of primary non-function were vascular and surgical problems (18/32, 56%), mainly renal artery or vein thrombosis and hemorrhage, and ischemia-reperfusion injury/graft quality (8/32, 25%). The reasons for allograft failure were not significantly different between transplants with and without pre-transplant DSA (Table 2).

Reasons for patient's death within the first year

Compared with allograft failures, death with a functioning graft occurred more equally distributed over the course of the first year (Fig. 2b). The leading causes of death were infections (38%), followed by unobserved deaths (19%), mostly in patients' domestic environment and without an autopsy performed to clarify the reason of death. Cardiovascular reasons (10%) were overall not frequent, even though it cannot be excluded that the majority of unobserved deaths resulted from cardiovascular events. Five patients (10%) died following gastrointestinal diseases and only four patients (8%) died of malignancy, among that one patient suffered from a post-transplantation lymphoproliferative disease (PTLD). Again, we did not observe a statistically significant difference regarding the reasons for patient death between pre-transplant DSA-positive and DSA-negative transplants (Table 2).

Impact of DSA characteristics on first-year transplant outcomes

Having shown that the presence or absence of pre-transplant DSA did not correlate with the investigated first-year renal outcomes, we next focused on DSA characteristics and their correlation with the outcomes of interest. Neither HLA class nor number of DSA, median MFI of the dominant DSA or median cumulative MFI of all DSA were statistically significantly different among the investigated outcomes (Table 3).

In our cohort of transplants with pre-transplant DSA, 45 (11%) had only historical but no current DSA. However, as shown in Table 3, these transplants were equally distributed among the investigated outcomes. When we excluded transplants with only historical DSA from the analysis addressing DSA characteristics, results remained unchanged (data not shown).

Differential impact of rejection episodes in transplants with and without pre-transplant DSA

Next, we focused on the occurrence of biopsy-proven rejection within the first year post-transplant. As detailed in Table 4, pre-transplant DSA-positive transplants underwent more frequently allograft biopsies and experienced significantly more often rejection episodes as compared to patients without pre-transplant DSA. This difference was primarily driven by a significantly higher proportion of ABMR. While T cell-mediated rejection (TCMR) occurred at a frequency of around 15% in both groups, pre-transplant DSA-positive transplants showed significantly more often ABMR as well as mixed rejection episodes in allograft biopsies ($P < 0.0001$). Interestingly, this did not impact allograft

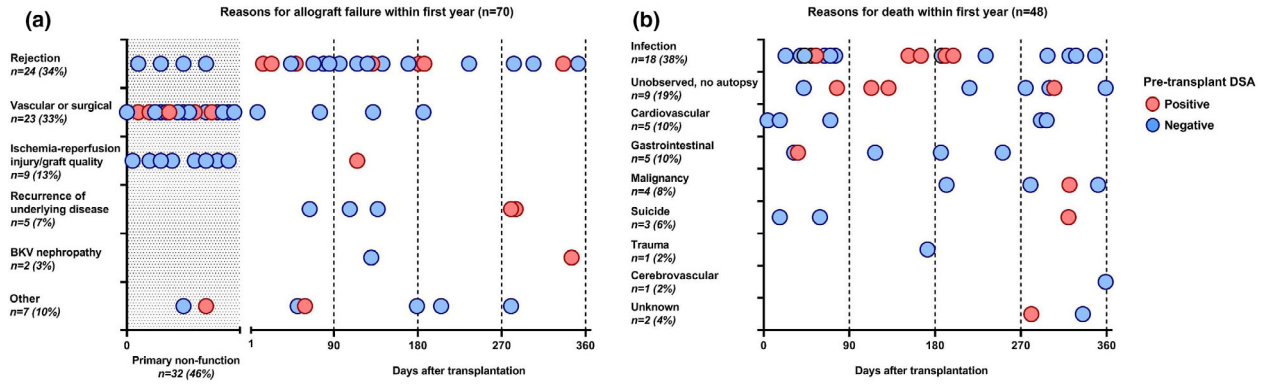

Figure 2 Reasons for allograft failure (a) and death (b) over the course of the first year post-transplant in transplants with and without pre-transplant donor-specific HLA antibodies (DSA). Abbreviations: BKV, BK polyomavirus; DSA, donor-specific HLA antibodies.

Table 3. Correlation of pre-transplant DSA characteristics with the composite outcome, as well as with the individual outcomes graft failure, poor allograft function and death in $n = 411$ pre-transplant DSA-positive transplants.

	Composite outcome reached			Allograft failure			Poor allograft function			Death		
Pre-transplant DSA characteristics	Yes ($n = 45$)	No ($n = 366$)	*P*-value	Yes ($n = 18$)	No ($n = 393$)	*P*-value	Yes ($n = 14$)	No ($n = 397$)	*P*-value	Yes ($n = 13$)	No ($n = 398$)	*P*-value
Class, % I/II/I+II	22/58/20	31/52/17	0.44	22/56/22	31/52/17	0.69	14/64/22	31/52/17	0.41	31/54/15	30/53/17	0.99
Number, % 1/2/>2	64/20/16	63/22/15	0.96	66/17/17	63/22/15	0.87	64/22/14	63/22/15	1.00	62/23/15	63/22/15	0.99
Dominant DSA MFI, median (IQR)	1916 (1130–5390)	1611 (925–3724)	0.29	1981 (1083–6427)	1621 (927–3747)	0.32	2471 (1059–6070)	1631 (933–3681)	0.36	1679 (1058–2507)	1634 (936–3834)	0.81
Cumulative MFI, median (IQR)	1965 (1238–6251)	1808 (957–4572)	0.34	2412 (1305–7198)	1810 (983–4626)	0.39	2667 (1059–7586)	1854 (1001–4537)	0.44	1916 (1238–2920)	1852 (997–4671)	0.91
Only historical DSA, no (%)	5 (11%)	40 (11%)	0.97	2 (11%)	43 (11%)	0.98	1 (7%)	44 (11%)	0.64	2 (15%)	43 (11%)	0.60

DSA, donor-specific HLA antibodies; IQR, interquartile range; MFI, mean fluorescence intensity.

function at one year when comparing serum creatinine and eGFR in transplants that were still functioning at this time (Table 4). In parallel, pre-transplant DSA status only mildly affected one-year death-censored graft survival, but was clearly associated with an inferior long-term allograft survival (Fig. 3a). However, when the groups were subdivided into those that did and did not experience biopsy-proven rejection within the first year, the combination of presence of pre-transplant DSA and rejection was associated with the overall worst death-censored graft survival ($P < 0.0001$; Fig. 3b). Importantly, this difference was already evident at one year and continued thereafter. Even though also DSA-negative allografts experiencing biopsy-proven rejection showed an inferior death-censored graft survival at one year and beyond, we still observed a significantly worse death-censored graft survival in DSA-positive transplants when directly comparing both groups ($P = 0.04$). These results did not change when we classified transplants with only historical but no current DSA as DSA-negative (Figure S2).

Predictors for first-year rejection in pre-transplant DSA-positive transplants

In total, 111/411 (27%) DSA-positive transplants developed biopsy-proven rejection within the first year. Table 5 details the univariable and multivariable analysis correlating various dichotomous immunological parameters, as well as re-transplantation status with the occurrence of any first year rejection. In this model, cumulative MFI of DSA was the only independent predictor for occurrence of first year rejection (odds ratio [OR] 1.07 [95% confidence interval (CI) 1.03–1.12]; $P = 0.0007$). However, when performing ROC analysis, the area under the curve (AUC) of cumulative DSA MFI in predicting any rejection within the first year post-transplant was only 0.59 with a cutoff MFI of 5185. AUC did not improve when we analyzed dominant DSA MFI (AUC 0.59; cutoff MFI 2873), indicating that both cumulative and dominant DSA MFI have only a moderate predictive value for first-year rejection (Fig. 4).

Discussion

This nationwide multicenter study identified 10% of renal transplants with a poor first-year renal transplant outcome defined as allograft failure, poor allograft function or death of the patient. Our main finding is that presence of pre-transplant DSA had no impact on the

Table 4. Occurrence of rejection and allograft survival in transplants with and without pre-transplant donor-specific HLA antibodies (DSA).

	DSA (n = 411)	No DSA (n = 1804)	P-value
Transplant biopsies in the first year			
Median	1 (0–2)	1 (0–1)	0.03
% 0/1/2/3/>3	39/33/18/7/3	44/32/18/5/1	0.006
Rejection episodes in the first year			
Any rejection	111 (27%)	287 (16%)	<0.0001
No of rejection episodes, % 0/1/2/>2	73/18/6/3	84/13/2/1	<0.0001
Any TCMR	56 (14%)	265 (15%)	0.58
Any ABMR	75 (18%)	31 (2%)	<0.0001
Rejection phenotypes in the first year			
First rejection phenotype	n = 111	n = 287	
TCMR I	12 (11%)	115 (40%)	<0.0001
TCMR II	30 (27%)	140 (49%)	
TCMR III	0	3 (1%)	
Active ABMR	49 (44%)	25 (9%)	
Chronic active ABMR	5 (4%)	1 (0%)	
Mixed rejection	15 (14%)	3 (1%)	
Most severe TCMR phenotype	n = 56	n = 265	
TCMR IA	10 (18%)	94 (35%)	0.09
TCMR IB	1 (2%)	8 (3%)	
TCMR IIA	42 (75%)	147 (55%)	
TCMR IIB	2 (3%)	13 (5%)	
TCMR III	1 (2%)	3 (1%)	
Most severe ABMR phenotype	n = 75	n = 31	
Active ABMR	70 (93%)	30 (97%)	0.49
Chronic active ABMR	5 (7%)	1 (3%)	
Allograft function at one year	n = 380	n = 1717	
Serum creatinine, µmol/l	122 (98–152)	123 (101–151)	0.42
eGFR (CKD-EPI), ml/min/1.73 m^2	50 (37–65)	52 (39–65)	0.22
Graft survival			
1 year	92%	95%	0.001
5 years	78%	84%	
10 years	55%	68%	
Death-censored graft survival			
1 year	96%	97%	0.0006
5 years	88%	92%	
10 years	71%	86%	

eGFR, estimated glomerular filtration rate; TCMR, T cell-mediated rejection; ABMR, antibody-mediated rejection.

investigated poor first-year renal transplant outcomes and only a mild impact on first-year death-censored allograft survival.

Most previously published studies described the effect of pre-transplant DSA on the long-term without specifically addressing their impact on first-year renal outcomes. Frequently, they consisted mainly of deceased donor transplants [9,20] and only few studies included also a substantial proportion of living transplants [21–23]. Compared with results from our study, both Ziemann *et al.* and Kamburova *et al.* reported a more pronounced impact of pre-transplant DSA on first-year allograft survival than in our study. However, there were differences in patient management and DSA-positive patients in our cohort received more frequently a T cell-depleting induction therapy. Together with a high proportion (86%) of DSA-positive transplants treated with an tacrolimus-based maintenance immunosuppression, this might have counterbalanced the risk conferred by pre-transplant DSA [24].

In our cohort, the absolute frequency of allograft failure within the first year was low (i.e. 3%). Another 5% of transplants showed a poor allograft function at one year. As in previous studies [24–26], such severely

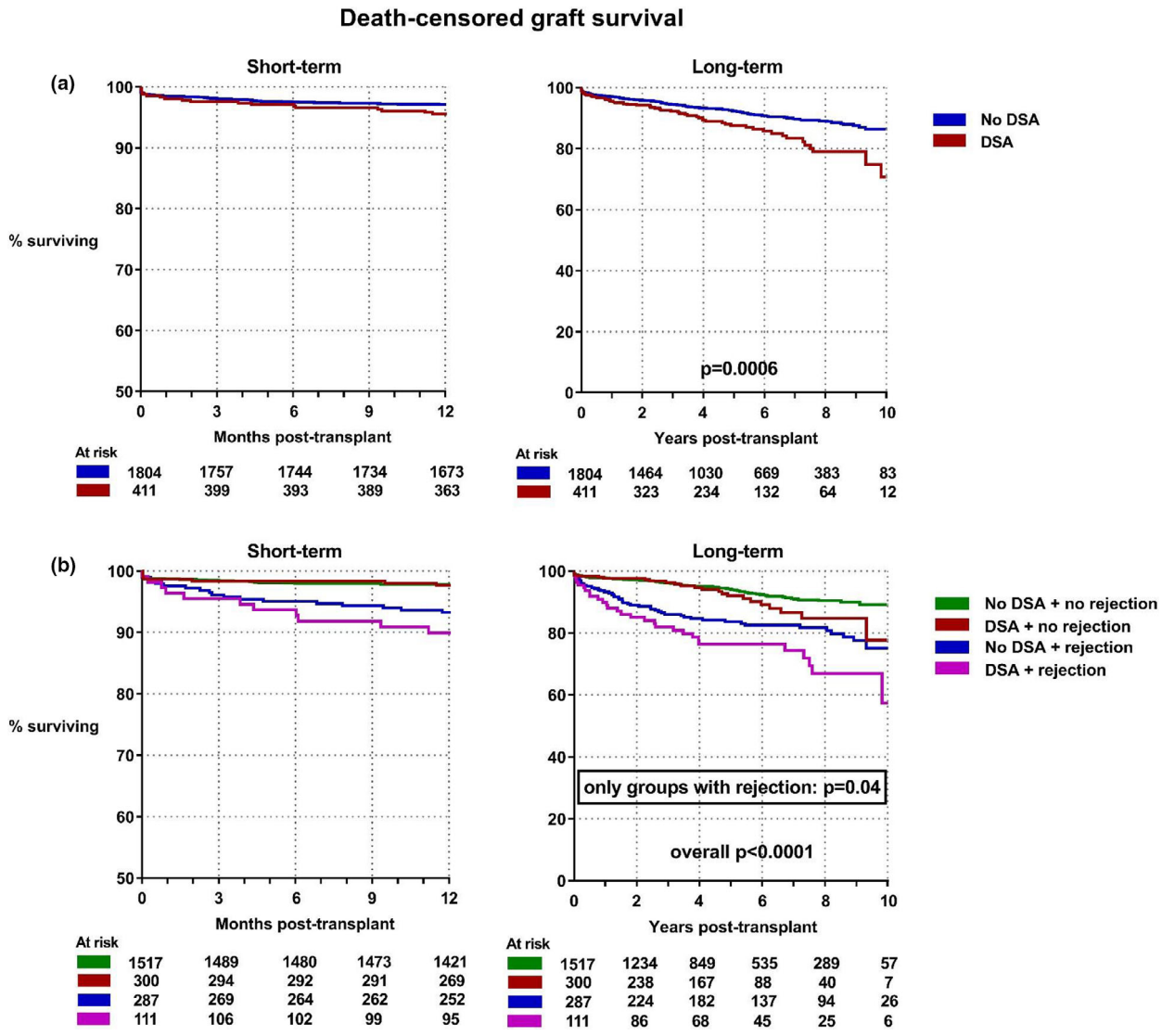

Figure 3 (a) Death-censored graft survival in transplants with and without pre-transplant donor-specific HLA antibodies (DSA). (b) Death-censored graft survival in transplants with and without pre-transplant donor-specific HLA antibodies (DSA), grouped by presence or absence of biopsy-proven rejection within the first year post-transplant.

impaired one-year allograft function correlated well with a decreased mid- and long-term allograft survival in our cohort and around 20% of allografts already failed until the end of the second year post-transplant. Neither the frequency of allograft failure and poor allograft function nor the reasons for allograft failure were significantly different with respect to presence of pre-transplant DSA. Still, rejection (34%) was, followed by vascular/surgical complications (33%), the most frequent reason for allograft failure within the first year. Compared with two previous European studies [13,27], this frequency is higher in our study, which might be related to the strict attribution to a dominant causative group, including allografts with primary non-function. We also investigated the timely occurrence of allograft failures over the course of the first year. Among transplants that either never took up function (n = 32, 46%) or failed until day 90 post-transplant (additional n = 11, 16%), we found a predominance of non-immunological reasons, which emphasizes their importance in the very early period post-transplant. From our perspective, it might be challenging to further improve early outcomes, since vascular procedure-related complications are often difficult to predict and may predominantly be influenced by donor organ and recipient factors [28,29]. In addition, allograft losses due to an ischemic injury and/or poor graft quality are also inherently linked to strategies aiming at increasing the donor pool by acceptance

Transplant International 2021; 34: 2755–2768

Table 5. Univariable and multivariable analysis for prediction of rejection within the first year post-transplant in pre-transplant DSA-positive transplants (n = 411).

Parameter	Rejection within first year (n = 111)	No rejection within first year (n = 300)	Univariable P-value	Multivariable OR (95% CI); P-value
Any class II DSA, no (%)	75 (68%)	211 (70%)	0.59	0.75 (0.46–1.24); P = 0.27
Multiple DSA, no (%)	49 (44%)	102 (34%)	0.06	1.01 (0.60–1.72); P = 0.96
Cumulative MFI, median (IQR)	2595 (1267–7518)	1800 (890–4126)	0.008	1.07 (1.03–1.12); P = 0.0007
Only current DSA, no (%)	98 (88%)	268 (89%)	0.76	0.62 (0.30–1.27); P = 0.20
A/B/DRB1 Mismatches, median (IQR)	4 (3–5)	4 (3–5)	0.97	1.03 (0.86–1.23); P = 0.71
Previous organ transplant, no (%)	61 (55%)	120 (40%)	0.007	1.54 (0.96–2.48); P = 0.07

CI, confidence interval; DSA, donor-specific HLA antibodies; IQR, interquartile range; MFI, mean fluorescence intensity; OR, Odds Ratio.

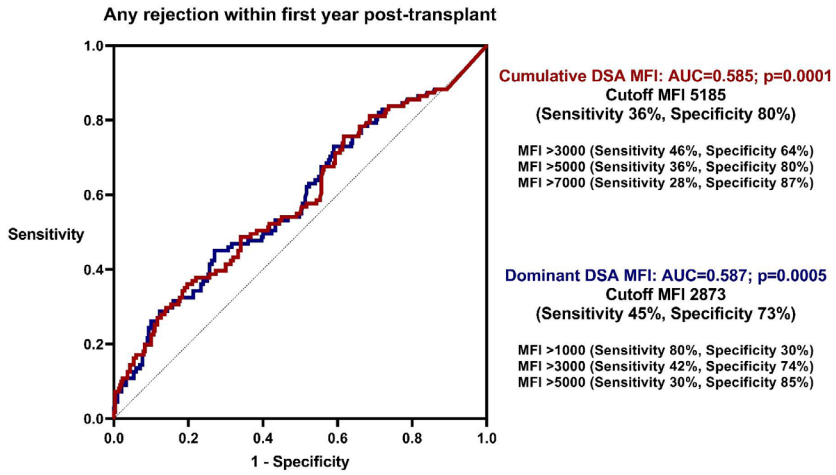

Figure 4 Receiver operating characteristic (ROC) statistics, area under the curve (AUC) values and diagnostic properties of cumulative and dominant DSA MFI for prediction of any rejection within the first year post-transplant. Abbreviations: AUC, area under the curve; DSA, donor-specific HLA antibodies; MFI, mean fluorescence intensity.

of rather marginal and extended criteria donors [30]. However, as indicated for instance by reasonable outcomes in the setting of donation after circulatory death programs [31], it seems worth accepting that allocation of such organs may not always be successful on an individual level.

Death of the patient within the first year was a rare event (i.e. 2%) in our study and similar to previously published studies [13,32,33]. As in the study of Farrugia *et al.* [32], infections were the leading cause of death, with an absolute risk of death from infection of 0.8% within the first year. Patients with pre-transplant DSA were not at higher risk for first-year death in our study. In addition, we did not observe a correlation between the use of a T cell-depleting therapy and death from infection (data not shown) but could not analyze the intensity of maintenance immunosuppression as well as rejection treatments in this regard. Cardiovascular and malignant diseases accounted for cumulative less than 20% of all deaths. Despite the fact that our analysis might underestimate the frequency of cardiovascular reasons for death due to a relative high number of unobserved and unclarified deaths, results suggest that the current standard of pre-transplant screening for cardiovascular diseases and malignancy is accurately able to prevent a fatal first-year outcome resulting from a pre-existing comorbidity in the vast majority of transplant recipients.

While we were unable to find an association between pre-transplant DSA status and the frequency of a poor first year outcome as defined in our study, the occurrence of first-year rejection differentially influenced the allograft survival. DSA-positive transplants with rejection showed the overall poorest allograft survival, which was significantly worse than in transplants with rejection but without evidence of pre-transplant DSA. This

difference in allograft survival was already visible in the first year and was primarily driven by a higher frequency of ABMR. Extended analysis of immunological parameters associated with first-year rejection in DSA-positive transplants identified only cumulative DSA MFI as a positive predictor, albeit with weak association as indicated by a low AUC in the ROC analysis. Even though DSA-positive transplants had a class II DSA in roughly 70%, we did not observe a predictive value of presence of class II DSA. From our perspective, these data underline important but largely unresolved challenges in transplanting patients in the presence of SAB-defined DSA. First, ABMR remains difficult to treat and may respond poorly to treatment [34]. Secondly, there is a current lack of reliable parameters to predict first-year ABMR in pre-transplant DSA-positive transplants, emphasizing that factors influencing the pathogenicity of DSA are still poorly understood [35,36]. Beside better characterization of additional antibody properties such as antibody epitope specificity and antibody affinity [37,38], it might also be promising to elucidate determinants of HLA antibody augmentation and renewal, since post-transplant persistence of DSA has also previously been associated with decreased allograft survival [39,40].

Our study has some important strengths. First, the study population is a large and unselected nationwide multicenter cohort, comprising about 92% of all Swiss transplants performed since 2008 and representing a real-life setting. Secondly, all transplants of our cohort had accurate and complete DSA assignment by virtual cross-match. Despite the fact that transplants were performed over almost one decade, HLA antibodies were in more than 99% of transplants determined by Luminex SAB technology. Since a rather low MFI cutoff was used, it seems unlikely that DSA were missed. Third, all rejection episodes were biopsy-proven and carefully recorded. Fourth, reasons for allograft failure and patient death were precisely determined and, if necessary, evaluated by individual chart review, providing the most accurate attribution to the different causative groups.

There are also some limitations applying to our study. While this multicenter cohort enabled us to study a large number of renal allograft recipients, there were center-specific differences in patient management, as well as selection of both induction and maintenance immunosuppression. Since some HLA data were retrospectively completed, we cannot exclude that presence of pre-transplant DSA was unknown in these cases at the time of transplantation and would have potentially altered selection of immunosuppression and post-transplant management. For definition of DSA, a rather low MFI cutoff was used. While this strategy bears the risk of overestimating DSA frequency, it reduces the risk of missing relevant DSA and was recently shown to correlate best with the impact on renal allograft survival [41]. *De novo* DSA occurring within the first year post-transplant were not investigated. Our analysis included both protocol and indication biopsies. However, not all centers performed protocol biopsies. Therefore, we might have underestimated the overall rejection frequency. Lastly, detailed information on treatment of rejection episodes and its success was not available, precluding a detailed analysis of association between immunosuppression and allograft failure due to rejection, as well as infection as a cause of patient's death.

In conclusion, this multicenter study shows that presence of pre-transplant DSA per se did not affect first-year renal transplant outcomes. However, DSA-positive transplants had a higher risk to develop biopsy-proven rejection episodes during the first year. Among all transplants, the combination of presence of pre-transplant DSA and first-year rejection was associated with the worst short- and long-term allograft survival. Since rejection remains poorly predictable in DSA-positive transplants, our results suggest an intense clinical surveillance of this subgroup that might benefit from early and aggressive rejection treatment.

Authorship

CW: designed and performed the research, analyzed the data, and wrote the article. PA, DS, UW, KH, SF-L, VA, AS, KS, TS, and JN: collected the data and critically reviewed the manuscript. SS: designed the research, collected and analyzed the data, and wrote the article.

Funding

The Swiss Transplant Cohort Study is supported by the Swiss National Science Foundation (https://www.snf.ch), Unimedsuisse (https://www.unimedsuisse.ch) and the Transplant Centers.

Conflicts of interest

The authors declare no conflicts of interest.

Acknowledgments

This study has been conducted in the framework of the Swiss Transplant Cohort Study, supported by the Swiss National Science Foundation and the Swiss University

Hospitals (G15) and transplant centers. The members of the Swiss Transplant Cohort Study are: Patrizia Amico, Andres Axel, John-David Aubert, Vanessa Banz, Beckmann Sonja, Guido Beldi, Christoph Berger, Ekaterine Berishvili, Isabelle Binet, Pierre-Yves Bochud, Sanda Branca, Heiner Bucher, Thierry Carrel, Emmanuelle Catana, Yves Chalandon, Sabina De Geest, Olivier De Rougemont, Michael Dickenmann, Joëlle Lynn Dreifuss, Michel Duchosal, Thomas Fehr, Sylvie Ferrari-Lacraz, Nicola Franscini, Christian Garzoni, Paola Gasche Soccal, Christophe Gaudet, Déla Golshayan, Nicolas Goossens, Karine Hadaya, Jörg Halter, Dominik Heim, Christoph Hess, Sven Hillinger, Hans Hirsch, Patricia Hirt, Günther Hofbauer, Uyen Huynh-Do, Franz Immer, Michael Koller (Head of the data center), Mirjam Laager, Bettina Laesser, Roger Lehmann, Alexander Leichtle, Christian Lovis, Oriol Manuel, Hans-Peter Marti, Pierre Yves Martin, Michele Martinelli, Valérie McLin, Katell Mellac, Aurélia Merçay, Karin Mettler, Nicolas Mueller (Chairman Scientific Committee), Antonia Müller, Thomas Müller, Ulrike Müller-Arndt, Beat Müllhaupt, Mirjam Nägeli, Graziano Oldani, Manuel Pascual (Executive office), Klara Posfay-Barbe, Juliane Rick, Anne Rosselet, Simona Rossi, Silvia Rothlin, Frank Ruschitzka, Urs Schanz, Stefan Schaub, Aurelia Schnyder, Macé Schuurmans, Thierry Sengstag, Federico Simonetta, Katharina Staufer, Susanne Stampf, Jürg Steiger (Head, Excecutive office), Guido Stirniman, Ueli Stürzinger, Christian Van Delden (Executive office), Jean-Pierre Venetz, Jean Villard, Julien Vionnet, Madeleine Wick (STCS coordinator), Markus Wilhlem, Patrick Yerly.

Data availability statement

The data that support the findings of this study are available from the corresponding author upon reasonable request.

SUPPORTING INFORMATION

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

Figure S1. Graft survival, death-censored graft survival, and patient survival in allografts with and without poor function at one year (defined by an eGFR ≤25 ml/min/1.73 m^2).

Figure S2. (A) Death-censored graft survival in transplants with and without current pre-transplant donor-specific HLA antibodies (DSA). In this analysis, transplants with only historical DSA were considered as having no DSA. (B) Death-censored graft survival in transplants with and without current pre-transplant donor-specific HLA antibodies (DSA), grouped by presence or absence of biopsy-proven rejection within the first year post-transplant. In this analysis, transplants with only historical DSA were considered as having no DSA.

REFERENCES

1. Sayegh MH, Carpenter CB. Transplantation 50 years later–progress, challenges, and promises. *N Engl J Med* 2004; **351**: 2761.
2. Gondos A, Dohler B, Brenner H, Opelz G. Kidney graft survival in Europe and the United States: strikingly different long-term outcomes. *Transplantation* 2013; **95**: 267.
3. Lamb KE, Lodhi S, Meier-Kriesche HU. Long-term renal allograft survival in the United States: a critical reappraisal. *Am J Transplant* 2011; **11**: 450.
4. Wekerle T, Segev D, Lechler R, Oberbauer R. Strategies for long-term preservation of kidney graft function. *Lancet* 2017; **389**: 2152.
5. Coemans M, Susal C, Dohler B, *et al.* Analyses of the short- and long-term graft survival after kidney transplantation in Europe between 1986 and 2015. *Kidney Int* 2018; **94**: 964.
6. Helal I, Abderrahim E, Ben Hamida F, *et al.* The first year renal function as a predictor of long-term graft survival after kidney transplantation. *Transplant Proc* 2009; **41**: 648.
7. Salvadori M, Rosati A, Bock A, *et al.* Estimated one-year glomerular filtration rate is the best predictor of long-term graft function following renal transplant. *Transplantation* 2006; **81**: 202.
8. Lefaucheur C, Loupy A, Hill GS, *et al.* Preexisting donor-specific HLA antibodies predict outcome in kidney transplantation. *J Am Soc Nephrol* 2010; **21**: 1398.
9. Mohan S, Palanisamy A, Tsapepas D, *et al.* Donor-specific antibodies adversely affect kidney allograft outcomes. *J Am Soc Nephrol* 2012; **23**: 2061.
10. Wehmeier C, Hoenger G, Cun H, *et al.* Donor specificity but not broadness of sensitization is associated with antibody-mediated rejection and graft loss in renal allograft recipients. *Am J Transplant* 2017; **17**: 2092.
11. De Broe ME, Elseviers MM. Analgesic nephropathy. *N Engl J Med* 1998; **338**: 446.
12. Hamed MO, Chen Y, Pasea L, *et al.* Early graft loss after kidney transplantation: risk factors and consequences. *Am J Transplant* 2015; **15**: 1632.
13. Helantera I, Raiha J, Finne P, Lempinen M. Early failure of kidney transplants in the current era-a national cohort study. *Transpl Int* 2018; **31**: 880.
14. Phelan PJ, O'Kelly P, Tarazi M, *et al.* Renal allograft loss in the first postoperative month: causes and consequences. *Clin Transplant* 2012; **26**: 544.
15. Dunn TB, Noreen H, Gillingham K, *et al.* Revisiting traditional risk factors

for rejection and graft loss after kidney transplantation. *Am J Transplant* 2011; **11**: 2132.
16. Huber L, Lachmann N, Niemann M, *et al.* Pretransplant virtual PRA and long-term outcomes of kidney transplant recipients. *Transpl Int* 2015; **28**: 710.
17. von Moos S, Cippa PE, van Breemen R, Mueller TF. HLA antibodies are associated with deterioration of kidney allograft function irrespective of donor specificity. *Hum Immunol* 2021; **82**: 19.
18. Lan JH, Kadatz M, Chang DT, Gill J, Gebel HM, Gill JS. Pretransplant calculated panel reactive antibody in the absence of donor-specific antibody and kidney allograft survival. *Clin J Am Soc Nephrol* 2021; **16**: 275.
19. Loupy A, Haas M, Solez K, *et al.* The Banff 2015 kidney meeting report: current challenges in rejection classification and prospects for adopting molecular pathology. *Am J Transplant* 2017; **17**: 28.
20. Loupy A, Lefaucheur C, Vernerey D, *et al.* Complement-binding anti-HLA antibodies and kidney-allograft survival. *N Engl J Med* 2013; **369**: 1215.
21. Kamburova EG, Wisse BW, Joosten I, *et al.* Differential effects of donor-specific HLA antibodies in living versus deceased donor transplant. *Am J Transplant* 2018; **18**: 2274.
22. Orandi BJ, Garonzik-Wang JM, Massie AB, *et al.* Quantifying the risk of incompatible kidney transplantation: a multicenter study. *Am J Transplant* 2014; **14**: 1573.
23. Ziemann M, Altermann W, Angert K, *et al.* Preformed donor-specific HLA Antibodies in living and deceased donor transplantation: a multicenter study. *Clin J Am Soc Nephrol* 2019; **14**: 1056.
24. Baechler K, Amico P, Hoenger G, *et al.* Efficacy of induction therapy with ATG and intravenous immunoglobulins in patients with low-level donor-specific HLA-antibodies. *Am J Transplant* 2010; **10**: 1254.
25. Kasiske BL, Israni AK, Snyder JJ, Skeans MA. Patient Outcomes in Renal Transplantation I. The relationship between kidney function and long-term graft survival after kidney transplant. *Am J Kidney Dis* 2011; **57**: 466.
26. Schnitzler MA, Johnston K, Axelrod D, Gheorghian A, Lentine KL. Associations of renal function at 1-year after kidney transplantation with subsequent return to dialysis, mortality, and healthcare costs. *Transplantation* 2011; **91**: 1347.
27. Van Loon E, Senev A, Lerut E, *et al.* Assessing the complex causes of kidney allograft loss. *Transplantation* 2020; **104**: 2557.
28. Kulu Y, Fathi P, Golriz M, *et al.* Impact of surgeon's experience on vascular and haemorrhagic complications after kidney transplantation. *Eur J Vasc Endovasc Surg* 2019; **57**: 139.
29. Wolff T, Schumacher M, Dell-Kuster S, *et al.* Surgical complications in kidney transplantation: no evidence for a learning curve. *J Surg Educ* 2014; **71**: 748.
30. Foroutan F, Friesen EL, Clark KE, *et al.* Risk factors for 1-year graft loss after kidney transplantation: systematic review and meta-analysis. *Clin J Am Soc Nephrol* 2019; **14**: 1642.
31. Snoeijs MG, Winkens B, Heemskerk MB, *et al.* Kidney transplantation from donors after cardiac death: a 25-year experience. *Transplantation* 2010; **90**: 1106.
32. Farrugia D, Cheshire J, Begaj I, Khosla S, Ray D, Sharif A. Death within the first year after kidney transplantation–an observational cohort study. *Transpl Int* 2014; **27**: 262.
33. Gill JS, Pereira BJ. Death in the first year after kidney transplantation: implications for patients on the transplant waiting list. *Transplantation* 2003; **75**: 113.
34. 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.
35. Courant M, Visentin J, Linares G, *et al.* The disappointing contribution of anti-human leukocyte antigen donor-specific antibodies characteristics for predicting allograft loss. *Nephrol Dial Transplant* 2018; **33**: 1853.
36. Valenzuela NM, Schaub S. The biology of IgG subclasses and their clinical relevance to transplantation. *Transplantation* 2018; **102**(1S Suppl. 1): S7.
37. Daga S, Moyse H, Briggs D, *et al.* Direct quantitative measurement of the kinetics of HLA-specific antibody interactions with isolated HLA proteins. *Hum Immunol* 2018; **79**: 122.
38. Kramer CSM, Franke-van Dijk MEI, Priddey AJ, *et al.* Recombinant human monoclonal HLA antibodies of different IgG subclasses recognising the same epitope: excellent tools to study differential effects of donor-specific antibodies. *HLA* 2019; **94**: 415.
39. Wehmeier C, Karahan GE, Krop J, *et al.* Donor-specific B cell memory in alloimmunized kidney transplant recipients: first clinical application of a novel method. *Transplantation* 2020; **104**: 1026.
40. Kimball PM, Baker MA, Wagner MB, King A. Surveillance of alloantibodies after transplantation identifies the risk of chronic rejection. *Kidney Int* 2011; **79**: 1131.
41. Wisse BW, Kamburova EG, Joosten I, *et al.* Toward a sensible single-antigen bead cutoff based on kidney graft survival. *Transplantation* 2019; **103**: 789.