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Original Article

Impact of donor-specific anti-HLA antibody on cardiac hemodynamics and graft function 3 years after pediatric heart transplantation: First results from the CTOTC-09 multi-institutional study

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ABSTRACT

The aim of this study (CTOTC-09) was to assess the impact of "preformed" (at transplant) donor-specific anti-HLA antibody (DSA) and first year newly detected DSA (ndDSA) on allograft function at 3 years after pediatric heart transplantation (PHTx). We enrolled children listed

Abbreviations: ACR, acute cellular rejection; AMR, antibody-mediated rejection; BNP, brain natriuretic peptide; CDC, complement-dependent cytotoxicity; CTOTC, Clinical Trials in Organ Transplantation in Children; DSA, donor-specific antibody; EST, estimate of the effect of a predictor variable on the modeled outcome; HLA, human leukocyte antigen; ISHLT, International Society for Heart and Lung Transplantation; MFI, median fluorescence intensity; ndDSA, newly detected DSA; OR, odds ratio; PCWP, pulmonary capillary wedge pressure; PHTx, pediatric heart transplant; PI, principal investigator; RHC, rejection with hemodynamic compromise.

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donor-specific antibodies
allograft function

at 9 North American centers. The primary end point was pulmonary capillary wedge pressure (PCWP) at 3 years posttransplant. Of 407 enrolled subjects, 370 achieved PHTx (mean age, 7.7 years; 57% male). Pre-PHTx sensitization status was nonsensitized (n = 163, 44%), sensitized/no DSA (n = 115, 31%), sensitized/DSA (n = 87, 24%), and insufficient DSA data (n = 5, 1%); 131 (35%) subjects developed ndDSA. Subjects with any DSA had comparable PCWP at 3 years to those with no DSA. There were also no significant differences overall between the 2 groups for other invasive hemodynamic measurements, systolic graft function by echocardiography, and serum brain natriuretic peptide concentration. However, in the multivariable analysis, persistent first-year DSA was a risk factor for 3-year abnormal graft function. Graft and patient survival did not differ between groups. In summary, overall, DSA status was not associated with worse allograft function or inferior patient and graft survival at 3 years, but persistent first-year DSA was a risk factor for late graft dysfunction.

1. Introduction

Pediatric heart transplant (PHTx) candidates are frequently sensitized to human leukocyte antigen (HLA) antigens, often with broad patterns of sensitization.[1,2] This reflects a high frequency of historical sensitizing events in this population, many of whom have undergone repair of congenital heart disease, not infrequently with use of homograft material for cardiac or vascular reconstruction.[2] In addition, we have demonstrated that approximately one-third of PHTx recipients develop newly detected donor-specific antibody (ndDSA) in the first year after transplantation.[3] The optimal strategy for managing these patients in the preoperative, perioperative, and postoperative periods remains largely unknown. In our Clinical Trials in Organ Transplantation in Children (CTOTC-04; NCT01005316) study, we showed that PHTx recipients who were highly sensitized at transplant and managed with perioperative antibody removal and an augmented immunosuppressive/immunomodulatory regimen had comparable 1-year patient and graft survival compared to nonsensitized transplant recipients.[4] We also demonstrated that first-year ndDSA was associated with increased risk of acute cellular rejection (ACR) but was not associated with early graft loss.[3] In this follow-up study (CTOTC-09; NCT02752789), we aimed to assess the impact of "preformed" donor-specific antibody (DSA) (at transplant) and first-year ndDSA on allograft function at 3 years as assessed by invasive cardiac hemodynamic assessment, echocardiography, and serum biomarkers. We hypothesized no detrimental effect of DSA on allograft function would be observed at 3 years posttransplant.

2. Materials and methods

2.1. Summary of study design

This multisite prospective study was part of the National Institutes of Health (NIH)–sponsored Clinical Trials in Organ Transplantation in Children (CTOTC) program (www.ctotc.org). The study was based on the original CTOTC-04 study design as previously reported.[2-4] These details include study design and organization, study sites, inclusion and exclusion criteria, study definitions, immunosuppression management, rejection surveillance and management, and use of core laboratories. Differences between CTOTC-04 and CTOTC-09 in terms of patient populations, study objectives, and primary and secondary end points are presented below. All sites received institutional review board approval, and informed consent and assent (if age appropriate) was obtained from all participants.

2.2. Patient population and clinical care guidelines

All eligible and consenting CTOTC-04 subjects at the 9 North American study sites were enrolled into CTOTC-09 for further follow-up, up to 5 years posttransplant. In addition, new subjects listed for transplantation at the end of CTOTC-04 enrollment were offered enrollment in CTOTC-09. Enrollment commenced in July 2014 and was completed in July 2016. Comparable with CTOTC-04, we attempted to recruit consecutive listed patients younger than 21 years at the time of listing and undergoing isolated orthotopic heart transplant at each study center. Multiorgan transplantation and failure to obtain informed consent were the only exclusion criteria. Donor selection was determined at the local clinical site as part of standard of care, and there was no protocol requirement for a negative virtual or complement-dependent cytotoxicity (CDC) crossmatch.[2] Pretransplant sensitization status was defined as presence of 1 or more anti-HLA antibodies with median fluorescence intensity (MFI) of $\geq$1000 using Luminex LABScreen single antigen beads (One Lambda; Thermo Fisher) utilizing the sample nearest (but prior) to transplant. For this report, all analyses are based on results of alloantibody testing in the Alloantibody Core Laboratory (University of Pittsburgh; principal investigator [PI]: A.Z.). Posttransplant DSA identification used the same single antigen methodology, and DSAs were defined as previously described.[4] Antibody analysis was performed in batches, and each patient's serum samples were tested in the same run by the same technologist to minimize technical issues. Interpretation of DSA specificity was performed by the Antibody Core Director (A.Z.) considering the assay limitations and possible false patterns. For

class II DQ DSAs, the specificity was assigned based on DQB1* and DQA1* pair. In patients with persistent DSA, we had the opportunity to observe the same pattern. Data from the Alloantibody Core were not made available to clinical sites during the course of the study, and there was no planned treatment for ndDSA in the absence of rejection findings. Echocardiographic, angiographic, and brain natriuretic peptide (BNP) measurements for this report were done at local site laboratories.

The standardized immunosuppression protocol has been described elsewhere.[2-4] In brief, all subjects received thymoglobulin induction therapy (total cumulative dose, 7.5 mg/kg) and maintenance immunosuppression with tacrolimus and mycophenolate mofetil. Corticosteroids were used only prior to the administration of each dose of thymoglobulin, and routine maintenance corticosteroids were not given for low immunologic risk subjects. CDC and/or flow-positive crossmatch-positive subjects or those with strongly positive virtual-positive crossmatch deemed at high risk for antibody-mediated rejection (AMR) by local site PI underwent a 1-fold to 3-fold intraoperative plasma exchange, a 5-day course of posttransplant plasma exchange/-plasmapheresis, and a course of posttransplant intravenous immunoglobulin (6 doses of 2 g/kg/dose given monthly). Maintenance immunosuppression in these high-risk subjects included maintenance corticosteroids for a minimum of 6 months, in addition to tacrolimus and mycophenolate mofetil. Rejection surveillance was by endomyocardial biopsy according to a standardized protocol and interpreted according to the guidelines of the International Society for Heart and Lung Transplantation (ISHLT),[5,6] and acute rejection was managed according to standardized guidelines as previously described.[2-4] However, if rejection advanced to graft dysfunction with hemodynamic compromise, further management was at the discretion of the treating physician. Cardiac catheterization for assessment of invasive cardiac hemodynamics and selective coronary angiography was performed per clinical protocol at 1, 3, and 5 years posttransplant and at any time when considered clinically indicated by the primary team managing the subject. Guidelines for obtaining, reviewing, interpreting, and reporting of hemodynamic measurements were provided to each site PI and cardiac catheterization laboratory.

2.3. Study end points analyzed in this report

The primary end point for CTOTC-09 is the pulmonary capillary wedge pressure (PCWP) obtained at cardiac catheterization at 3 years posttransplant. Secondary end points for this report include the following: echocardiographic assessment of systolic function (ejection fraction [EF], %); other hemodynamic measurements including mean pulmonary artery pressure, mean right atrial pressure, right and left ventricular end-diastolic pressures; cardiac index; and serum BNP. Other secondary end points included graft and patient survival, ACR, AMR, rejection with hemodynamic compromise (RHC), clinical rejection event (defined as a clinical event, irrespective of ISHLT grade, leading to an acute augmentation of immunosuppression), and posttransplant coronary artery disease defined by selective coronary angiography and evaluated according to the guidelines of ISHLT.[7]

For this report, all end points were analyzed at the 3-year posttransplant time point. Participants who died before 3 years posttransplant were not available in the analyses for the main 3-year outcomes. Data imputation was not performed for these participants regarding the 3-year end point.

2.4. Statistical considerations

The statistical analyses were performed on 370 transplanted subjects: 194 subjects previously enrolled in CTOTC-04 and 176 newly enrolled in CTOTC-09. Continuous variables were summarized with means, standard deviations, median, and IQR and categorical variables with counts and percentages. General linear models were used to evaluate the primary end point among subjects with known sensitization status at transplantation. Logistic regression analysis was used to evaluate predictors of 1 or more measures of abnormal hemodynamics or systolic function using the same population. The multivariable models were constructed using backward selection at significance level of 0.10, generating an estimate (EST) or odds ratio (OR) for the effect of each predictor on the modeled outcome. Two group comparisons were performed via *t* tests. Time-to-event analyses were performed using Kaplan-Meier plots and log-rank tests. All statistical analyses were performed using SAS version 9.4.

3. Results

3.1. Subject characteristics

The consort diagram (Fig. 1) follows the course of subjects within the study. Of 407 enrolled subjects, 370 achieved transplantation. Outcomes prior to transplantation (for those not achieving transplant) are also shown in Figure 1; 194 subjects were previously enrolled in CTOTC-04 and had continued follow-up without interruption within CTOTC-09. The remaining 176 subjects were newly enrolled in CTOTC-09, not having been previously enrolled in CTOTC-04. The mean age at transplantation was 7.7 years (range, 0-22 years), and 57% were male patients. Sensitization status at the time of transplant was as follows: nonsensitized (n = 163, 44%), sensitized/no DSA (n = 115, 31%), and sensitized/DSA (n = 87, 24%), with 5 (1%) having insufficient data to determine status. Thus, overall, more than half of all subjects had evidence of HLA sensitization at the time of transplantation. Among this sensitized group, 90 (44.6%) underwent perioperative desensitization per protocol based on sensitization profile at the time of transplantation, with 42 subjects continuing with the postoperative protocol for high-risk patients based on the results of DSA status at transplant and crossmatch results.[2] The baseline characteristics of subjects who underwent transplantation are shown in Table 1 stratified by sensitization status at the time of transplant.

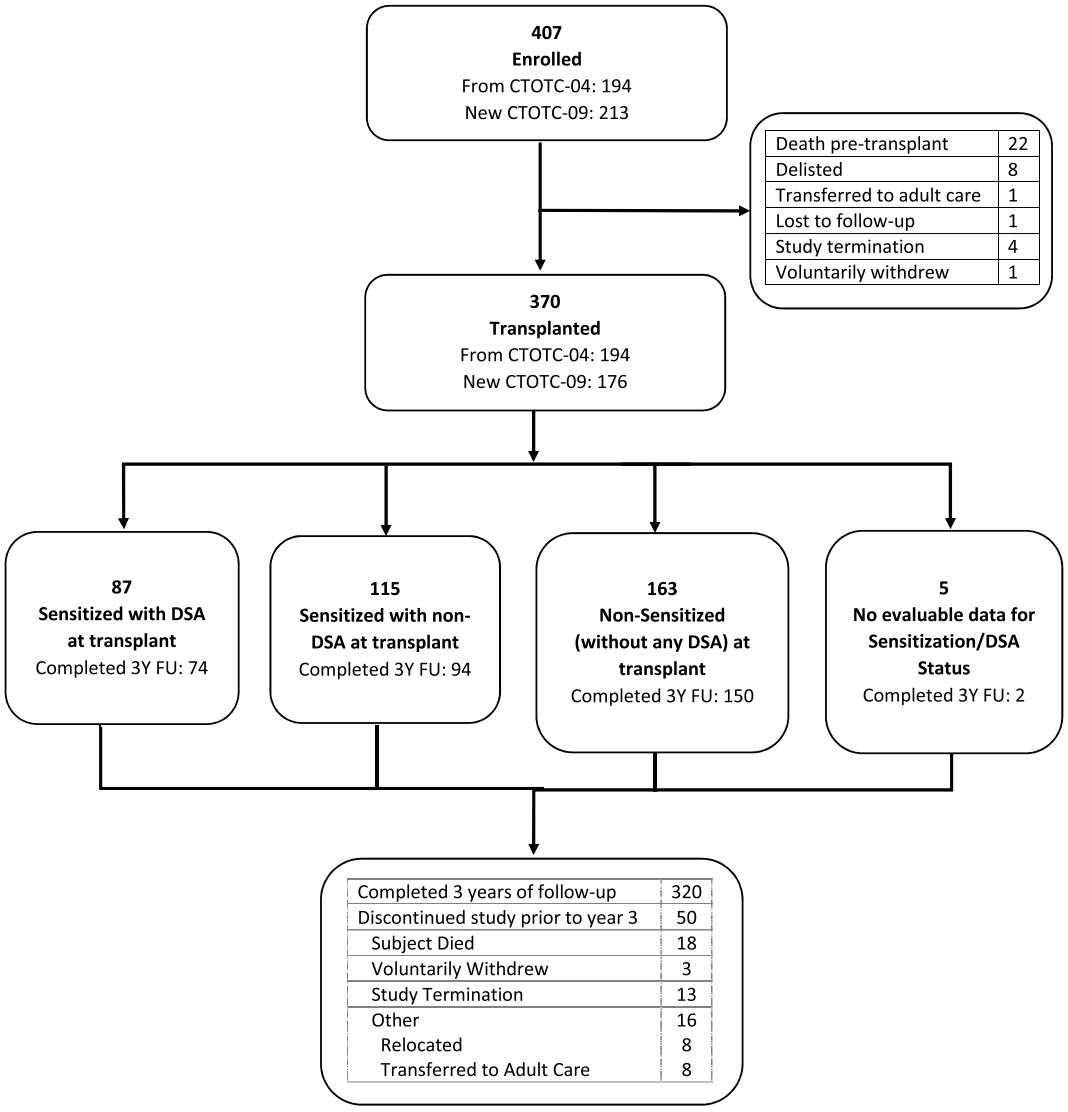

Figure 1. Consort diagram. Flow chart of all enrolled patients through the study: 194 patients previously enrolled in CTOTC-04 and 213 newly enrolled patients participated in the study. Reasons for discontinuation pretransplant and posttransplant, but prior to year 3 visit, are provided; 370 transplanted patients were classified based on central laboratory-determined sensitization status at transplant. Five of 370 patients had insufficient data to determine their baseline sensitization status. CTOTC, Clinical Trials in Organ Transplantation in Children; DSA, donor-specific antibody; FU, follow-up.

3.2. Subjects evaluable at 3 years posttransplant (primary end point)

PCWP obtained by cardiac catheterization at the time of the 3-year study visit was available in 291 of the 370 subjects (79%). Few differences in subject characteristics were identified between those with (n = 291) and without (n = 79) PCWP assessment at 3 years posttransplant. The group with invasive hemodynamic data available had a higher proportion of subjects with Hispanic or Latinx ethnicity ($P = .013$) and were more likely to be United Netwrork for Organ Sharing (UNOS) status 1A /hospitalized at the time of transplantation ($P < .001$ and $P = .038$, respectively) (Supplementary Table 1). Of note, those with and without 3-year hemodynamic data did not differ by pretransplant sensitization status ($P = .152$). Among the 79 without PCWP data, 50 did not complete the 3-year study visit due to subject death (n = 18), relocation or transfer to adult care (n = 16), study termination prior to reaching the 3-year visit (n = 13), and voluntary study withdrawal (n = 3). The remaining 29 subjects underwent a 3-year study visit but did not have available PCWP data. See full details of nonevaluable subjects in Supplementary Table 1. Of the 79 without evaluable PCWP data at 3 years, we examined 54 who underwent catheterization at a median of 2.1 years posttransplant (IQR 1.5, 2.5). Their PCWP was comparable to those with evaluable data at 3 years (median PCWP 11 vs 10 mm Hg with IQRs of 8, 14 and 8, 12 mm Hg, respectively). Information on diuretic use at 3 years was available for a subset of the subjects, and we noted no difference between those with (13/290) and without (1/27) evaluable PCWP (4.5% vs 3.7%, respectively).

Table 1
Baseline subject characteristics based on Alloantibody Core laboratory-determined sensitization status at transplantation.

	Sensitized with DSA (N = 87)	Sensitized without DSA (N = 115)	Nonsensitized (N = 163)	p-value[1]	Total[2] (N = 370)
Age at transplant (y)				0.064	
N	87	115	163		370
Mean (SD)	8.9 (6.31)	7.6 (6.26)	6.9 (7.00)		7.7 (6.63)
Median (IQR)	9.0 (3.0, 15.0)	8.0 (1.0, 13.0)	5.0 (0.0, 14.0)		7.0 (1.0, 14.0)
Range	(0.0-20.0)	(0.0-20.0)	(0.0-22.0)		(0.0-22.0)
Sex				0.304	
Female	31 (35.6%)	51 (44.3%)	74 (45.4%)		158 (42.7%)
Male	56 (64.4%)	64 (55.7%)	89 (54.6%)		212 (57.3%)
Ethnicity				0.024	
Hispanic or Latino	18 (20.7%)	15 (13.0%)	16 (9.8%)		50 (13.5%)
Not Hispanic or Latino	45 (51.7%)	75 (65.2%)	118 (72.4%)		240 (64.9%)
Unknown or not reported	24 (27.6%)	25 (21.7%)	29 (17.8%)		80 (21.6%)
Predominate race				0.333	
White	45 (51.7%)	68 (59.1%)	104 (63.8%)		221 (59.7%)
Black or African American	22 (25.3%)	24 (20.9%)	30 (18.4%)		76 (20.5%)
Asian	3 (3.4%)	3 (2.6%)	10 (6.1%)		16 (4.3%)
Native Hawaiian or Other Pacific Islander	0	0	1 (0.6%)		1 (0.3%)
More than 1 race	0	1 (0.9%)	0		1 (0.3%)
Unknown or not reported	17 (19.5%)	19 (16.5%)	18 (11.0%)		55 (14.9%)
Weight at transplant (kg)				0.017	
N	87	111	156		358
Mean (SD)	38.1 (27.83)	30.3 (23.47)	28.5 (25.35)		31.6 (25.60)
Median (IQR)	32.6 (14.0, 52.4)	22.3 (9.5, 47.9)	19.2 (7.8, 42.4)		23.1 (9.8, 49.3)
Range	(5.9-134.2)	(3.0-100.6)	(3.0-118.6)		(3.0-134.2)
Height at transplant (cm)				0.017	
N	78	102	146		330
Mean (SD)	126.7 (38.87)	117.0 (40.40)	110.1 (42.94)		116.6 (41.59)
Median (IQR)	130.1 (92.0, 163.0)	121.5 (76.0, 155.0)	109.0 (67.0, 153.0)		118.0 (77.0, 156.0)
Range	(58.0-193.0)	(45.7-191.0)	(7.9-182.9)		(7.9-193.0)
Status at time of transplant					
UNOS status at transplant				0.469	
1A	73 (83.9%)	94 (81.7%)	138 (84.7%)		308 (83.2%)
1B	8 (9.2%)	15 (13.0%)	20 (12.3%)		45 (12.2%)
2	6 (6.9%)	6 (5.2%)	4 (2.5%)		16 (4.3%)
Missing	0	0	1 (0.6%)		1 (0.3%)
Hospitalized at transplant				0.150	
Yes	53 (60.9%)	77 (67.0%)	118 (72.4%)		249 (67.3%)
No	34 (39.1%)	38 (33.0%)	44 (27.0%)		120 (32.4%)

(*continued on next page*)

Table 1 (*continued*)

	Sensitized with DSA (N = 87)	Sensitized without DSA (N = 115)	Nonsensitized (N = 163)	p-value[1]	Total[2] (N = 370)
Missing	0	0	1 (0.6%)		1 (0.3%)
Intensive care unit at transplant				0.009	
Yes	26 (29.9%)	58 (50.4%)	76 (46.6%)		160 (43.2%)
No	61 (70.1%)	57 (49.6%)	87 (53.4%)		210 (56.8%)
Ventilator at transplant				0.279	
Yes	7 (8.0%)	11 (9.6%)	23 (14.1%)		41 (11.1%)
No	80 (92.0%)	104 (90.4%)	140 (85.9%)		329 (88.9%)
ECMO at transplant				0.366	
Yes	4 (4.6%)	2 (1.7%)	8 (4.9%)		14 (3.8%)
No	83 (95.4%)	113 (98.3%)	155 (95.1%)		356 (96.2%)
VAD at transplant				0.950	
Yes	15 (17.2%)	18 (15.7%)	26 (16.0%)		60 (16.2%)
No	72 (82.8%)	97 (84.3%)	137 (84.0%)		310 (83.8%)
MCS at transplant				0.689	
Yes	19 (21.8%)	20 (17.4%)	34 (20.9%)		74 (20.0%)
No	68 (78.2%)	95 (82.6%)	129 (79.1%)		296 (80.0%)
Prior sensitizing event					
Prior sensitizing event				0.027	
Yes	74 (85.1%)	79 (68.7%)	120 (73.6%)		278 (75.1%)
No	13 (14.9%)	36 (31.3%)	43 (26.4%)		92 (24.9%)
Prior sensitizing: surgery				<0.001	
Yes	66 (75.9%)	57 (49.6%)	78 (47.9%)		204 (55.1%)
No	21 (24.1%)	58 (50.4%)	85 (52.1%)		166 (44.9%)
Prior sensitizing: VAD				0.998	
Yes	20 (23.0%)	26 (22.6%)	37 (22.7%)		85 (23.0%)
No	67 (77.0%)	89 (77.4%)	126 (77.3%)		285 (77.0%)
Prior sensitizing: ECMO				0.048	
Yes	20 (23.0%)	12 (10.4%)	24 (14.7%)		56 (15.1%)
No	67 (77.0%)	103 (89.6%)	139 (85.3%)		314 (84.9%)
Prior sensitizing: any MCS (VAD or ECMO)				0.275	
Yes	32 (36.8%)	31 (27.0%)	56 (34.4%)		121 (32.7%)
No	55 (63.2%)	84 (73.0%)	107 (65.6%)		249 (67.3%)
Prior sensitizing: blood transfusion				0.002	
Yes	61 (70.1%)	53 (46.1%)	85 (52.1%)		203 (54.9%)
No	26 (29.9%)	62 (53.9%)	78 (47.9%)		167 (45.1%)
Prior sensitizing: homograft placement				<0.001	
Yes	21 (24.1%)	12 (10.4%)	10 (6.1%)		45 (12.2%)
No	66 (75.9%)	103 (89.6%)	153 (93.9%)		325 (87.8%)

(*continued on next page*)

Table 1 (*continued*)

	Sensitized with DSA (N = 87)	Sensitized without DSA (N = 115)	Nonsensitized (N = 163)	p-value[1]	Total[2] (N = 370)
Prior sensitizing: prior pregnancy				0.702	
Yes	0	1 (0.9%)	1 (0.6%)		2 (0.5%)
No	87 (100.0%)	114 (99.1%)	162 (99.4%)		368 (99.5%)
Prior sensitizing: prior transplant				0.214	
Yes	6 (6.9%)	4 (3.5%)	4 (2.5%)		14 (3.8%)
No	81 (93.1%)	111 (96.5%)	159 (97.5%)		356 (96.2%)

Abbreviations: ECMO, Extracorporeal membrane oxygenation; MCS, mechanical circulatory support; VAD, ventricular assist device.

[1] P value results from Cochran-Mantel-Haenszel test for categorical variables and ANOVA for continuous variables.

[2] 5 Subjects did not have sufficient data to evaluate their sensitization DSA status but were included in the Total column.

3.3. Characteristics of subjects and DSA profiles at transplant and during first year posttransplant

Overall, 169 of 370 (46%) subjects had DSA prior to transplantation and/or developed ndDSA posttransplant and 1 had (0.3%) insufficient data to determine pretransplant DSA status or posttransplant ndDSA status (Table 2). Of these 169 subjects, 38 (22%) had DSA prior to transplantation but did not develop ndDSA; 82 subjects (49%) developed ndDSA in the first year after transplantation without preformed DSA; and 49 subjects (29%) who had DSA prior to transplantation also developed ndDSA. Furthermore, among the 169 subjects with DSA (at transplant and/or ndDSA), 66 (39%) had at least 1 DSA with MFI of $\geq$8000 and 75 (44%) had the sum of the MFIs of all DSA of $\geq$8000. Among the 82 subjects who did not have DSA at transplantation but who developed ndDSA in the first year after transplantation, 61 (74%) subjects developed ndDSA in the first 6 weeks after transplantation, and 33 (26%) subjects developed ndDSA beyond 6 weeks. The characteristics of DSA at transplant and ndDSA identified during the first year after transplantation are summarized in Table 2.

3.4. Primary outcome and other measures of graft function at 3 years after transplantation

The predetermined study primary end point was assessment of PCWP obtained during invasive cardiac catheterization at 3 years after transplantation. In addition, to provide a multimodal assessment of graft function, we also assessed echocardiographic assessment of systolic function (EF) and serum measurement of BNP (with logarithmic transformation). Figure 2 demonstrates PCWP, EF, and $\log_{10}$ BNP for subjects who underwent transplantation stratified by presence or absence of DSA at transplant and/or during the first year following transplantation. Thus, this analysis incorporates exposure of the graft to preformed and/or newly detected DSA in the first year posttransplant. For each variable, there was no difference between the DSA and the non-DSA groups (Fig. 2). Similarly, there were no significant differences between the 2 groups for other hemodynamic measurements (pulmonary artery, right atrial, right and left ventricular end-diastolic pressures; all $P > .100$) nor for cardiac index ($P = .363$) (Supplementary Table 2).

A second analysis was performed to identify any associations of ndDNA in the first year posttransplant with PCWP, EF, or log 10 BNP, regardless of the presence of DSA at transplant. Within each mutually exclusive group at transplant (nonsensitized/sensitized-no DSA/sensitized with DSA), we compared subjects with and without ndDSA in the first year after transplant and found no differences between the groups (Fig. 3).

3.5. Risk factors for elevated PCWP and abnormal graft function

Variables assessed as predictors of higher PCWP in a univariable model are tabulated in Supplementary Table 3. In the multivariable model (Table 3), increasing weight per kilogram (Estimate [EST] = 0.04; 95% CI = 0.02-0.07; $P = .001$) was found to be associated with higher PCWP. Posttransplant coronary artery disease, which was present in only 15 subjects, was also associated with higher PCWP (EST = 1.83; 95% CI = 0.13-3.54; $P = .035$). Of note, the presence of DSA at transplant and/or during the first-year posttransplant did not predict higher PCWP ($P = .855$). Refining the model (model 2, Table 3) using the presence or absence of the sum of MFI of $\geq$8000 for all DSA at any given time point pretransplant or during first year posttransplant also did not identify DSA as a predictor of higher PCWP (EST = 0.36; 95% CI = −0.51 to 1.24; $P = .416$).

We then explored in univariable analysis the same variables that were used for prediction of higher PCWP (Supplementary Table 3) but this time for the prediction of abnormal hemodynamics/systolic function. For this analysis, abnormal graft function was defined as the presence of 1 or more invasive hemodynamic measurements above normal range for age; or EF >2 standard deviations below normal for age. Of 318 subjects with hemodynamic assessment and echocardiogram at 3 years, 58 subjects (18%) had 1 or more measurements outside the specified normal range. In the final multivariable model, only weight (OR = 1.04; 95% CI = 1.02-1.05; $P < .001$ per kilogram

Table 2
Characteristics of preformed or newly detected donor-specific antibodies.

	Subjects with evaluable DSA data (N = 369)			
	Preformed DSA only (N = 38)	Newly detected DSA only in year 1 (N = 82)	Both preformed and newly detected DSA in year 1 (N = 49)	Preformed and/or newly detected DSA in year 1 (N = 169)
Class I only	18 (47.4%); (4.9%)	38 (46.3%); (10.3%)	8 (16.3%); (2.2%)	64 (37.9%); (17.3%)
Class II only	14 (36.8%); (3.8%)	18 (22.0%); (4.9%)	1 (2.0%); (0.3%)	33 (19.5%); (8.9%)
Both class I and II	6 (15.8%); (1.6%)	26 (31.7%); (7.0%)	40 (81.6%); (10.8%)	72 (42.6%); (19.5%)
Maximum MFI of DSA <4000	24 (63.2%); (6.5%)	47 (57.3%); (12.7%)	9 (18.4%); (2.4%)	80 (47.3%); (21.7%)
Maximum MFI of DSA 4000-7999	4 (10.5%); (1.1%)	11 (13.4%); (3.0%)	8 (16.3%); (2.2%)	23 (13.6%); (6.2%)
Maximum MFI of DSA >=8000	10 (26.3%); (2.7%)	24 (29.3%); (6.5%)	32 (65.3%); (8.7%)	66 (39.1%); (17.9%)
Maximum Sum of MFI of DSA <4000	20 (52.6%); (5.4%)	39 (47.6%); (10.6%)	5 (10.2%); (1.4%)	64 (37.9%); (17.3%)
Maximum Sum of MFI of DSA 4000-7999	6 (15.8%); (1.6%)	15 (18.3%); (4.1%)	9 (18.4%); (2.4%)	30 (17.8%); (8.1%)
Maximum Sum of MFI of DSA >=8000	12 (31.6%); (3.3%)	28 (34.1%); (7.6%)	35 (71.4%); (9.5%)	75 (44.4%); (20.3%)

Note: 2 percentages are presented. The first one was based off the column (N). The second one was based off the # of subjects with at least one DSA sample available pre- or posttransplant (N = 369). One transplanted subject did not have sufficient sample for DSA determination.

weight increase) and the presence of any prior episode of RHC (OR = 11.13; 95% CI = 1.87-66.18; $P = .008$) were associated with 1 or more measures of abnormal hemodynamics or abnormal EF. The presence of coronary artery disease approached significance (OR = 3.18; 95% CI = 0.83-12.09; $P = .090$). Of note, the overall presence of DSA at transplant and/or during the first-year posttransplant did not predict abnormal hemodynamics or reduced EF (Table 3). A post hoc analysis found no evidence of significant collinearity among DSA, CAV, and RHC.

We then examined the effect of transient vs persistent DSA (defined as 2 or more occurrences in the first year posttransplant) and found no effect on PCWP, but there was an association with abnormal hemodynamics/systolic function (OR = 2.76; $P = .001$). This effect was maintained in the multivariate model (OR = 3.19; 95% CI = 1.56-6.49; $P = .001$ (Table 3). Furthermore, we examined the effect of class I vs class II vs no DSA in the first year posttransplant and found no association with PCWP ($P = .272$) or abnormal hemodynamics/systolic function ($P = .153$). The analysis was repeated using any class II (ie, with or without class I), vs no class II, and again no association with PCWP ($P = .132$) or abnormal hemodynamics ($P = .231$) was observed.

3.6. Death, retransplantation, and rejection outcomes

Freedom from the composite of death, retransplantation, and RHC (the primary end point in the CTOTC-04 study) stratified by pretransplant sensitization status is shown in Figure 4A for the CTOTC-09 cohort. The individual components of the composite are shown in Figure 4B-D. Death and retransplantation did not differ by sensitization status at transplant. However, freedom from RHC was different across the sensitization groups ($P = .011$), with the lowest freedom from rejection observed in subjects with 1 or more moderate/high strength DSA (MFI $\geq$ 4000) at transplantation ($P = .005$ vs nonsensitized subjects) (Fig. 4C). Freedom from AMR was lower in subjects with pretransplant sensitization (Fig. 5A) ($P < .001$) and was the lowest in those with pretransplant DSA MFI of $\geq$4000 ($P < .001$ vs nonsensitized subjects). Freedom from ACR did not differ across the pretransplant sensitization groups.

4. Discussion

4.1. Understanding the long-term consequences of DSAs

In our CTOTC-04 study, we demonstrated that sensitized pediatric heart candidates with DSA (including those with positive cytotoxicity crossmatch) could achieve 1-year graft and patient survival comparable with nonsensitized candidates when managed with perioperative antibody removal and augmented immunosuppression.[4] We further demonstrated that acceptance of donor organs irrespective of the crossmatch result leads to comparable waitlist mortality between sensitized and nonsensitized candidates, a finding that differs from traditional approaches of waiting for a negative crossmatch.[1,8,9]

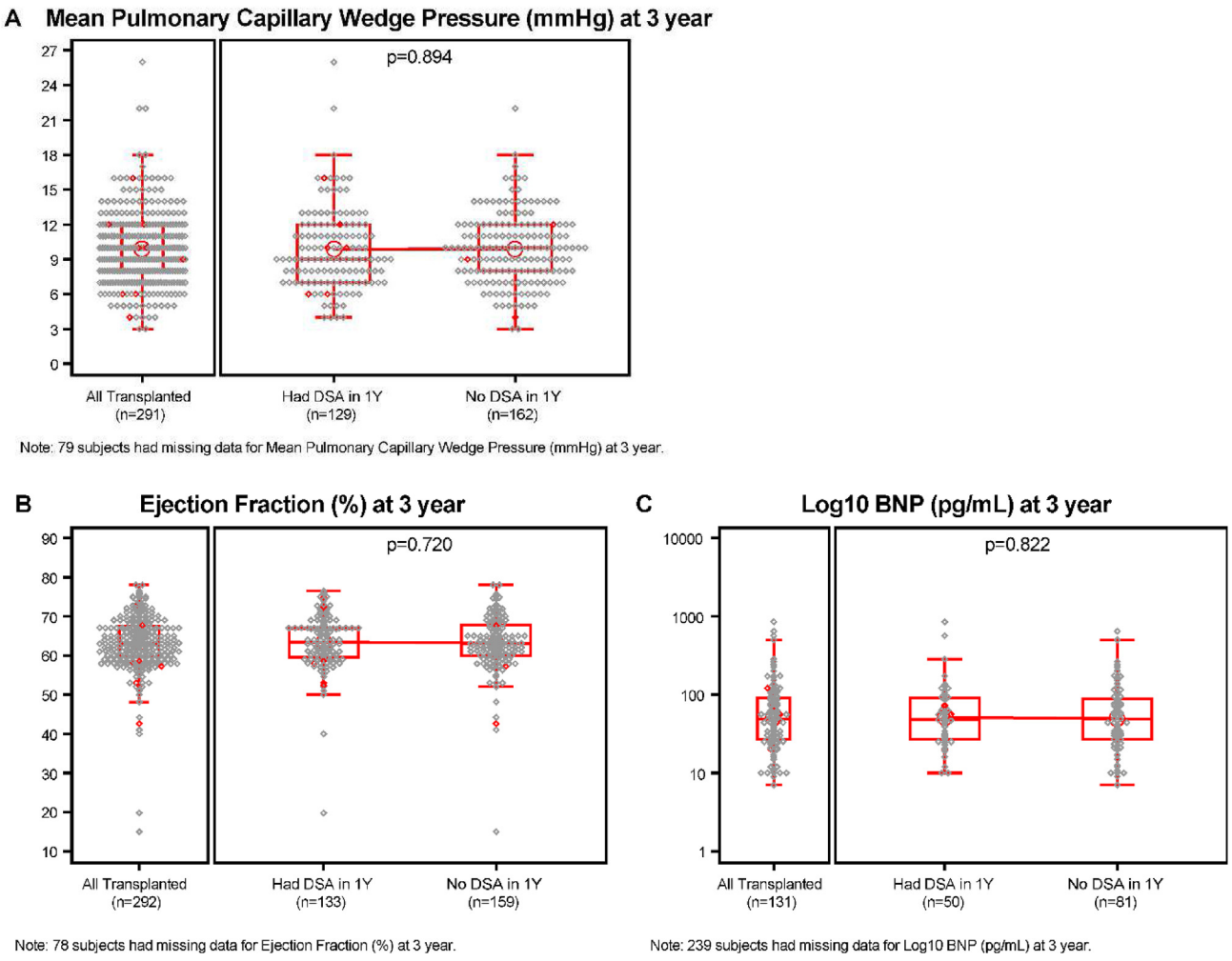

Figure 2. Box plots of hemodynamic measurements by preformed and/or newly detected DSA in first year posttransplant. Hemodynamic measurements at 3 years posttransplant are plotted for (A) mean pulmonary capillary wedge pressure (mm Hg) (291 patients with available data), (B) ejection fraction (%) (292 patients with available data), and (C) $\log_{10}$ brain natriuretic peptide (BNP) (pg/mL) (131 patients with available data). Each subpart (A-C) includes 2 panels. The left-hand panel presents 1 box plot of all patients who underwent transplantation and had the specific measurement available at year 3, and the right-hand panel presents the measurement stratified by whether or not the patient had preformed and/or newly detected DSA in the first-year posttransplant (labeled as Had DSA in 1Y vs No DSA in 1Y). The length of the box represents the interquartile range of 25th and 75th percentiles, the circle in the box represents the group mean, the horizontal line in the box represents the group median, and the whiskers from the box represents the 5th and 95th percentiles. The means of 2 groups were connected by a line; 9 patients who had acute rejection at the time of collection of their 3-year hemodynamic measurements are highlighted in red. The *P* values result from *t* tests to compare the mean difference between 2 groups. DSA, donor-specific antibody.

The CTOTC-04 study focused on follow-up at 1 year posttransplant and on outcomes for those most at risk for adverse events, that is, those highly sensitized with a positive donor-specific CDC. We also observed that pretransplant sensitization was highly prevalent.[2,4] Furthermore, one-third of subjects who underwent transplantation were noted to have ndDSA in the first year after transplantation.[3] Thus, many more patients are at risk for DSA-mediated graft damage than just the small population with positive CDC crossmatch, and these patients should be carefully followed up within the setting of prospective cohort studies (or clinical trials). These goals were incorporated into CTOTC-09, which was designed to study a large cohort of PHTx recipients whose allografts are exposed to DSA, whether present at the time of transplantation or which developed posttransplant.

4.2. Key observations from the CTOTC-09 study

In the CTOTC-09 study, we again noted that over half of the subjects were sensitized at transplantation, with almost a quarter of the entire cohort having DSA at transplantation. Furthermore, one-third of all subjects had ndDSA identified in the first year after transplantation, and a substantial proportion of subjects had one or more ndDSA at high strength (MFI $\geq$ 8000). Our careful analyses of graft function failed to demonstrate any relationship overall between DSA and graft function at 3 years posttransplant. This was performed first by comparing 2 groups: those with and those without any DSA, whether preformed at transplant and/or newly detected in the first year thereafter. To define any specific contribution of ndDSA to graft function at 3 years, we reanalyzed

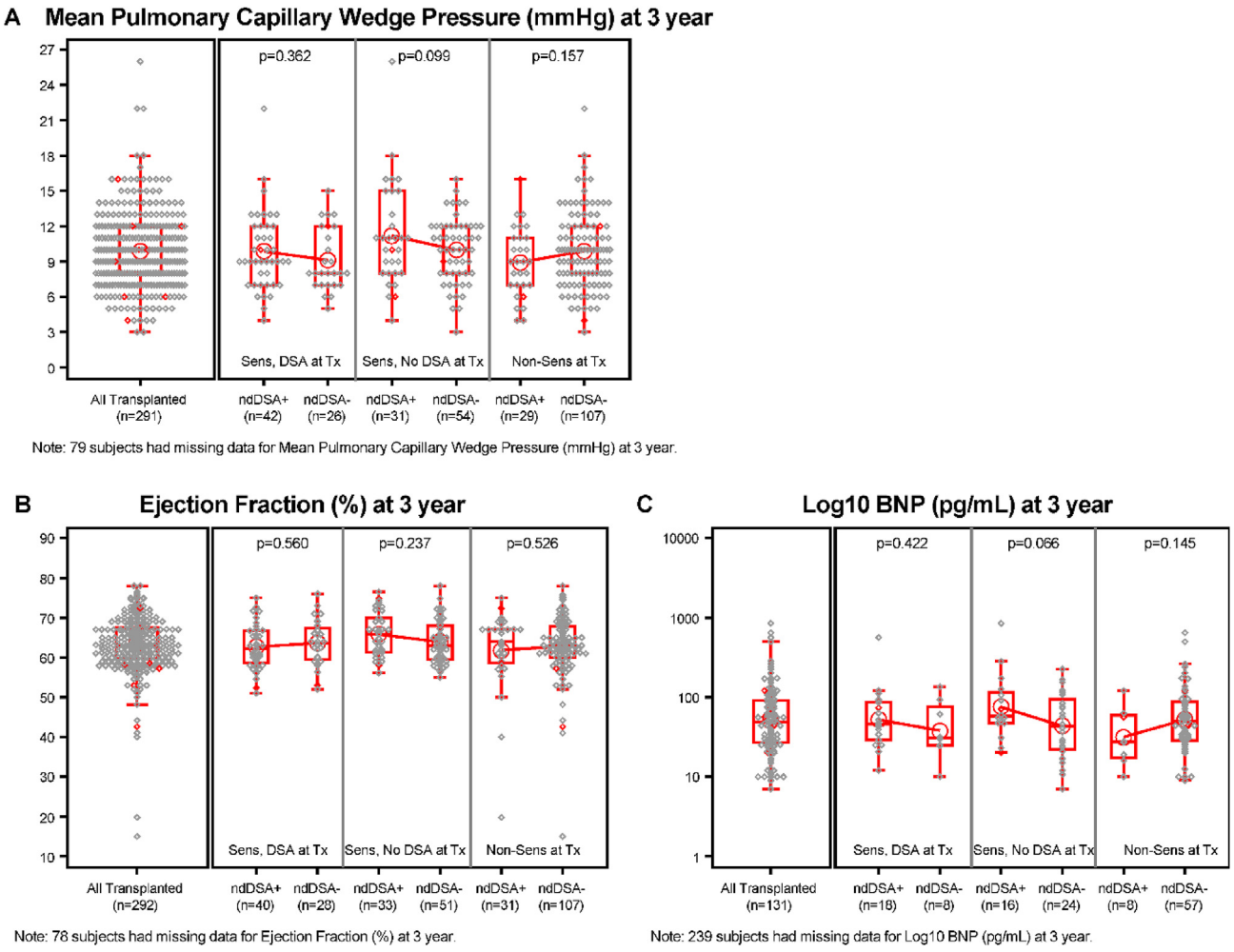

Figure 3. Box plots of hemodynamic measurements by sensitized status at transplant and newly formed DSA in 1-year posttransplant. Hemodynamic measurements at 3 years posttransplant are plotted for (A) mean pulmonary capillary wedge pressure (mm Hg) (291 patients with available data), (B) ejection fraction (%) (292 patients with available data), and (C) $\log_{10}$ brain natriuretic peptide (BNP) (pg/mL) (131 patients with available data). Each subpart (A-C) includes 2 panels. The left-hand panel presents 1 box plot of all patients who underwent transplantation and had the specific measurement available at year 3, and the right-hand panel presents the measurement stratified by 3 groups of sensitization status at transplantation; sensitized with DSA, sensitized without DSA, and nonsensitized (labeled as Sens, DSA at Tx vs Sens, No DSA at Tx vs Non-Sens at Tx) and within each sensitization status group, patients were stratified by whether the patient had newly detected DSA in the first year posttransplant (labeled as ndDSA+ vs. ndDSA−). The length of the box represents the interquartile range of 25th and 75th percentiles, the circle in the box represents the group mean, the horizontal line in the box represents the group median, and the whiskers from the box represents the 5th and 95th percentiles. The means of 2 groups were connected by a line; 9 patients who experienced acute rejections at the time of 3 years hemodynamic measurements were collected are highlighted in red. The *P* values result from general linear regression models to test the mean difference between 2 subgroups are at the top of each panel. DSA, donor-specific antibody; ndDSA, newly detected DSA.

the data set looking at the impact of ndDSA (vs no ndDSA) among the 3 mutually exclusive groups at the time of transplant (nonsensitized; sensitized/no DSA; sensitized/DSA). Again, we did not observe any impact of ndDSA on PCWP and graft dysfunction at 3 year posttransplant. Furthermore, we evaluated 2 other independently measured modalities of graft function: echocardiographically derived EF and serum BNP levels. Again, no relationship between overall DSA and graft function was observed. Thus, 3 independently performed measures of graft function/status (hemodynamics, imaging, and serum biomarker) failed to identify any overall relationship between DSA and graft status at 3 years. Although we did not find an overall association between first-year DSA (preformed at transplant and/or ndDSA in the first year) and graft function at 3 years posttransplant, we did detect an association between persistent first-year DSA and abnormal graft function (although not with the primary end point of PCWP). This is consistent with other studies that suggest that persistent posttransplant de novo DSA may be an important marker of late graft dysfunction.[10,11]

4.3. Implications of study findings

Clinicians are faced with the challenge that many children on the heart transplant waitlist are sensitized against HLA antigens, many with 1 or more high-strength antibodies and often with broad patterns of sensitization. Findings from the current study

Table 3
Multivariable analysis results for pulmonary capillary wedge pressure and adverse graft function at 3 years posttransplant.

Risk factors	PCWP[1]				Adverse graft function (>=1 abnormal hemodynamic measurement)[2]	
	Model 1		Model 2			
	Estimate (95% CI)	p value	Estimate (95% CI)	p value	Odds ratio (95% CI)	p value
Number of observations Used	288	0.005	288	0.004	315	<0.001
Age at 3 y post tx (y)	-0.13 (−0.24, −0.02)	0.017	-0.13 (−0.24, −0.03)	0.016		
Weight at 3 y post tx (kg)	0.04 (0.02, 0.07)	0.001	0.04 (0.02, 0.07)	0.001	1.04 (1.02, 1.05)	<0.001
Any rejection with hemodynamic compromise upto 3 y Yes vs No					11.13 (1.87, 66.18)	0.008
Coronary artery disease upto 3 years Yes vs No	1.83 (0.13, 3.54)	0.035	1.84 (0.14, 3.54)	0.034	3.18 (0.83, 12.09)	0.090
Preformed and/or newly formed DSA in first year post-tx Yes vs No	-0.07 (−0.81, 0.67)	0.855				
Preformed and/or newly formed DSA using cumulative MFI of C1 & C2 >=8000 in first year post-tx Yes vs No			0.36 (-0.51, 1.24)	0.416	3.19 (1.56,6.49) 001	0.001
Persistent vs. transient/none DSA in 1st year posttransplant						

[1] The models were selected using backward selection at significance level of 0.10. General linear model was used for estimates and p-values.

[2] The model was selected using backward selection at significance level of 0.10. Logistic regression model was used to estimate odds ratios and p-values.

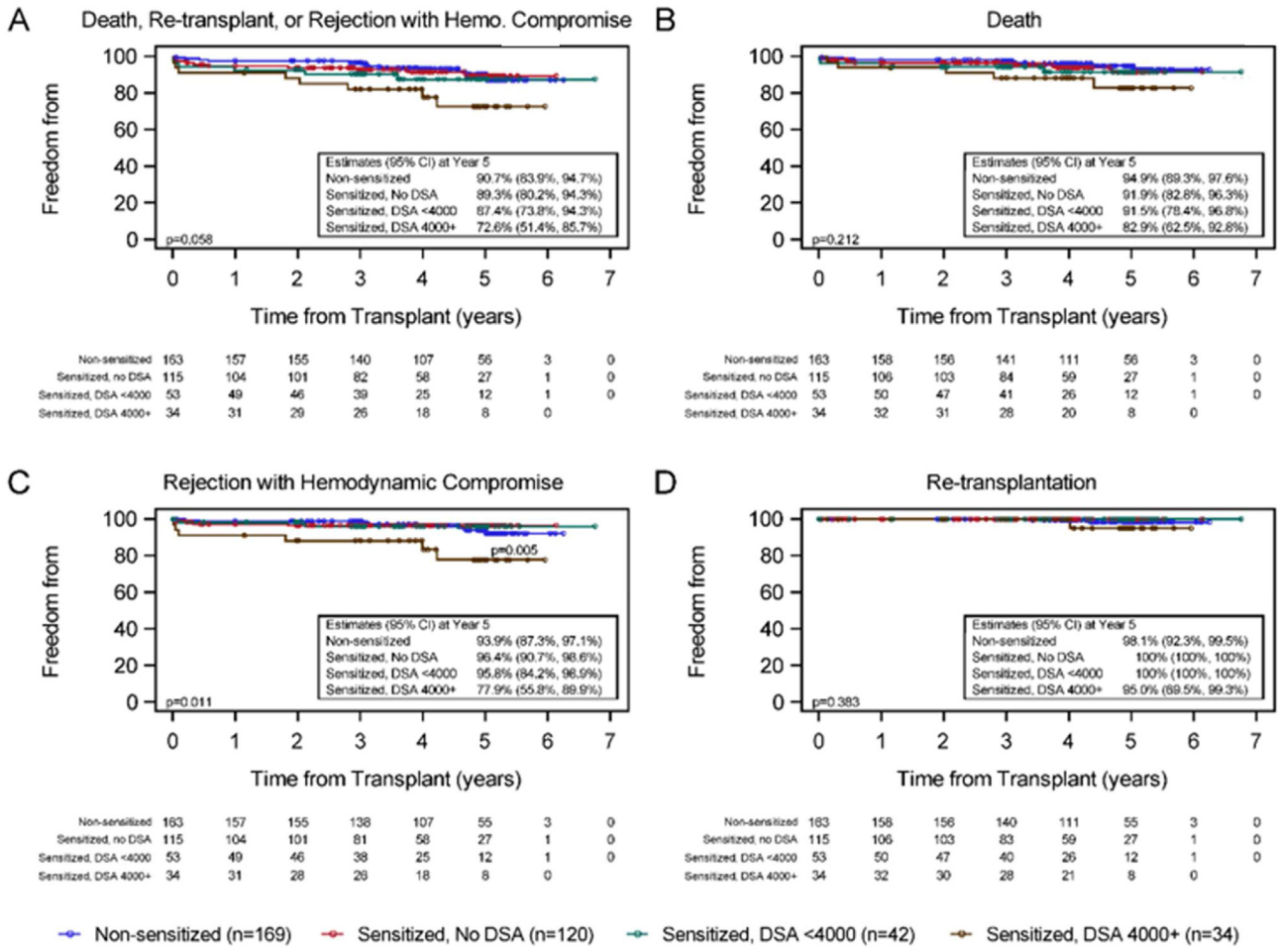

Figure 4. Freedom from the composite of death, retransplantation, or rejection with hemodynamic compromise stratified by sensitization status and highest MFI of DSA at transplantation based on Alloantibody Core Laboratory testing. Probability of freedom from the composite of death, retransplantation, or rejection with hemodynamic compromise (A) and its constituent components (B-D) stratified by sensitization status and highest MFI of DSA at transplantation based on Alloantibody Core Laboratory testing. The number of patients at risk is presented annually posttransplant. Kaplan-Meier survival analysis was used to produce the estimates with a 95% CI for each group; *P* values reported from the log-rank test shown in the bottom left corner of each panel. An additional *P* value (*P* =.005) is shown for pairwise comparison of rejection with hemodynamic compromise comparing subjects sensitized with 1 or more DSA with MFI 4000+ with nonsensitized subjects (C). DSA, donor-specific antibody; MFI, median fluorescence intensity.

(and from CTOTC-04) suggest that children should not automatically be excluded from transplantation in the setting of a positive virtual (or actual) crossmatch if the chance of waitlist mortality is considered to be substantial. We acknowledge however that this study was not designed to determine whether there are any absolute contraindications to transplantation in this setting. Ongoing analysis of samples from CTOTC-09 will determine whether certain antibody characteristics (eg, titer, immunoglobulin G subclass, and complement-fixing ability) can better define the risk of adverse graft outcomes after transplantation across a positive crossmatch.[12]

A number of prior reports in the pediatric and adult heart transplant literature suggest that sensitization at transplant with DSA and posttransplant ndDSA are associated with adverse graft outcomes, including acute and chronic graft dysfunction, posttransplant coronary artery disease, graft loss, and death.[10,11,13-15] In particular, persistent DSA,[10,11] class II DSA,[11,14] complement-fixing DSA,[13] and late-onset ndDSA after 1 year,[15] all carried adverse prognosis across differing studies. We identified 2 recent reports with findings similar to our own following adult thoracic transplantation. Sommer et al[16] reported the experience of the Hannover and Heidelberg teams managing sensitized adult heart transplant recipients. Their perioperative management and maintenance immunosuppressive regimens were similar to ours, although with the addition of intraoperative tocilizumab and a single dose of rituximab on day 5 posttransplant. Excellent outcomes were observed at 1 year, similar to those seen in CTOTC-04 and CTOTC-09. In another recent report, Aversa et al[17] from the Toronto Lung Transplant Program managed sensitized lung recipients with a protocol very similar to CTOC-04/09. They found no difference in long-term graft survival and freedom from chronic lung allograft dysfunction among subjects who were nonsensitized, sensitized without DSA, or

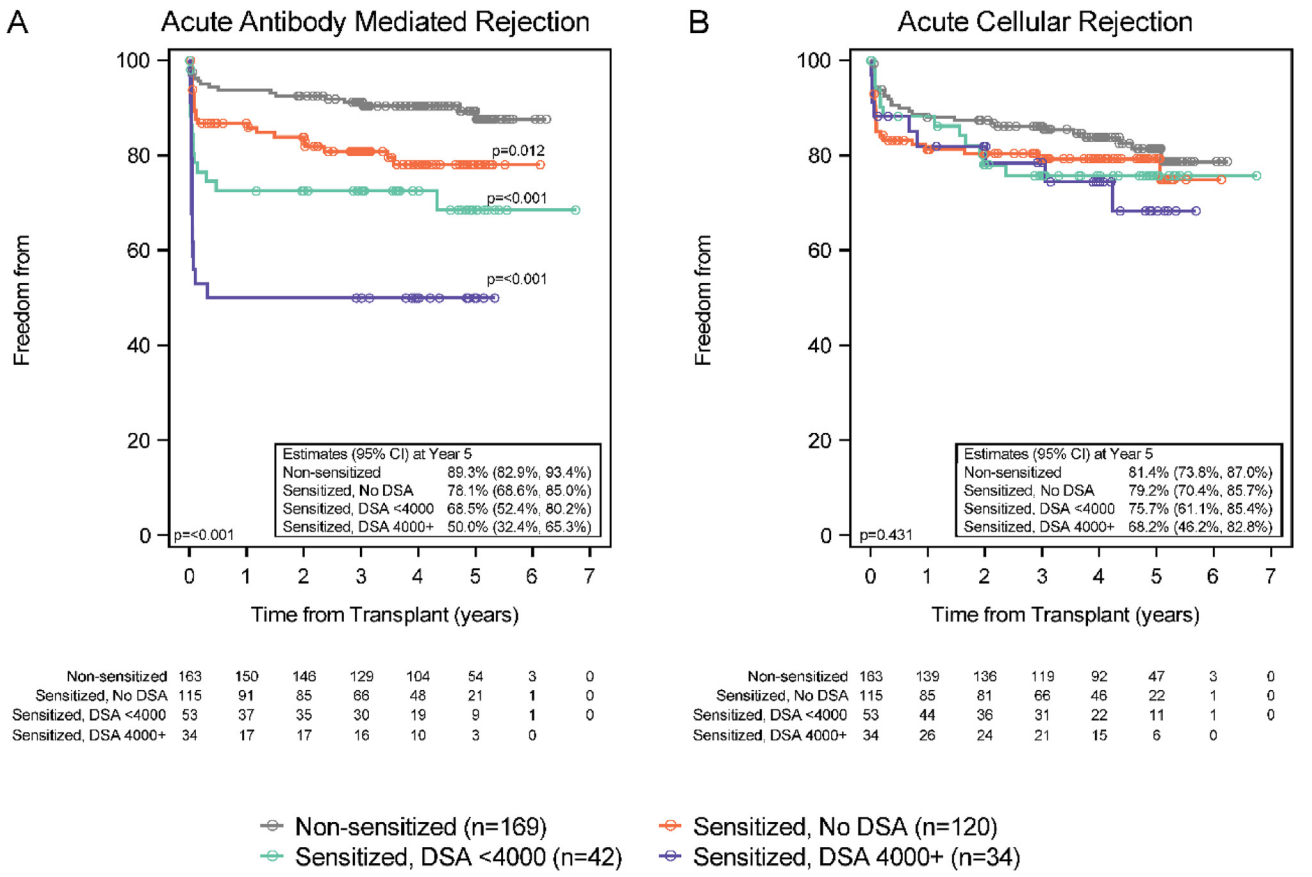

Figure 5. Freedom from acute antibody-mediated rejection and acute cellular rejection stratified by sensitization status and highest MFI of DSA at transplantation based on Alloantibody Core Laboratory testing. Probability of freedom from acute antibody-mediated rejection (A) and acute cellular rejection (B) stratified by sensitization status and highest MFI of DSA at transplantation based on Alloantibody Core Laboratory testing. The number of patients at risk is presented annually posttransplant. Kaplan-Meier survival analysis was used to produce the estimates with 95% CIs for each group; *P* values reported from the log-rank test comparing across all 4 groups and pairwise comparisons with the nonsensitized group. DSA, donor-specific antibody; MFI, median fluorescence intensity.

sensitized with DSA at the time of transplantation. The median follow-up in that cohort was 6.7 years.

It is unclear why our overall findings, and those of Sommer et al[16] and Avers et al,[17] are different from the literature (although with similarity in relation to impact of persistent first-year antibodies).[10,11] In the absence of randomized controlled trials, we can only conjecture that it is the specific immunomodulatory and immunosuppressive strategies employed that are critical to achieving excellent outcomes in high-risk populations. Early antibody removal (including intraoperative plasma exchange) may result in exposure of the donor vascular endothelium to low titer/concentration of DSA during weaning from cardiopulmonary bypass. There is a growing literature to suggest that initial exposure of vascular endothelium to low concentration DSA results in enhanced expression of survival genes and decreased expression of adhesion molecules that protects the endothelium during subsequent re-exposure with high concentration DSA[18-20] (as may occur following cessation of plasmapheresis/exchange). However, if this does reflect a degree of graft accommodation (ie, the resistance of an allograft to the acute pathologic effects of graft specific antibodies and complement fixation),[20,21] then the accommodation must be transient and/or incomplete in many subjects since we did observe graft injury in the form of AMR.

4.4. Study limitations

We did not randomize subjects to any specific therapy, so we cannot prove which specific interventions performed in this study, if any, led to the satisfactory outcomes observed among subjects sensitized with DSA at the time of transplantation. The follow-up remains relatively short. The low prevalence of coronary disease may be an underestimate as we did not perform intravascular ultrasound assessments of the coronary arteries since this is not standard of care in the study centers and carries risk of morbidity (and even mortality) in small recipients (a high proportion of this cohort). Nonetheless, the overall incidence of coronary disease diagnosed by angiography is very low at this time, despite the large number of subjects with DSA. Further follow-up is necessary to determine if freedom from moderate or severe coronary artery disease is comparable between those with and without

DSA over the life of the graft. Such a follow-up study is planned. We also recognize that the predictive value for clinical outcomes may be enhanced by more detailed characterization of pretransplant and posttransplant DSA. Such studies are already underway. Other ongoing studies from this cohort include the assessment of more subtle degrees of graft dysfunction (including diastolic dysfunction) by detailed evaluation of serial echocardiograms in the Echocardiography Core laboratory as well as serial assessment of coronary angiograms by the Angiography Core laboratory out to 5 years posttransplant. A final important limitation is that we could only assess the primary and secondary end points for those subjects alive and evaluable at 3 years posttransplant—a potential cause of bias. However, a careful evaluation of evaluable and nonevaluable subjects demonstrated that less than a quarter of nonevaluable subjects were deceased at 3 years, and DSA status at transplant did not differ between those who were and who were not evaluable for the primary end point.

We would stress, however, that our findings do not mean that DSA never impact graft function, cause graft failure, or lead to chronic graft vasculopathy. We also did observe AMR in a significant proportion of subjects with high-strength DSA. We simply present our data at 3 years within CTOTC-09 and acknowledge that other immune modulatory and immunosuppression protocols may not achieve comparable results.

5. Conclusions

Among PHTx recipients managed with a predefined protocol of care, we found overall no difference in graft function at 3 years between those with DSA (at transplant and/or with newly detected DSA in the first-year posttransplant) compared to those without DSA. In multivariable models, DSA status in the first-year posttransplant was not determined to be a risk factor for elevated PCWP at 3 years posttransplant, but we did observe that persistent first year DSA was a risk factor for late graft dysfunction. Ongoing follow-up will determine if these encouraging findings persist long-term.

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Data availability

All source data from the CTOTC-09 study will be made publicly available according to the NIH policy.

Declaration of Competing Interest

The authors declare the following financial interests/personal relationships which may be considered as potential competing interests: Steven Webber reports financial support was provided by National Institutes of Health.

Disclosure

The authors of this manuscript have no conflicts of interest to disclose as described by the *American Journal of Transplantation*.

Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.ajt.2023.08.006.

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References

1. Mahle WT, Tresler MA, Edens RE, et al. Allosensitization and outcomes in pediatric heart transplantation. *J Heart Lung Transplant*. 2011;30(11):1221–1227.
2. Zuckerman WA, Zeevi A, Mason KL, et al. Study rationale, design, and pretransplantation alloantibody status: A first report of Clinical Trials in Organ Transplantation in Children-04 (CTOTC-04) in pediatric heart transplantation. *Am J Transplant*. 2018;18(9):2135–2147.
3. Dipchand AI, Webber S, Mason K, et al. Incidence, characterization, and impact of newly detected donor-specific anti-HLA antibody in the first year after pediatric heart transplantation: a report from the CTOTC-04 study. *Am J Transplant*. 2018;18(9):2163–2174.
4. Webber S, Zeevi A, Mason K, et al. Pediatric heart transplantation across a positive crossmatch: first year results from the CTOTC-04 multi-institutional study. *Am J Transplant*. 2018;18(9):2148–2162.
5. Stewart S, Winters GL, Fishbein MC, et al. Revision of the 1990 working formulation for the standardization of nomenclature in the diagnosis of heart rejection. *J Heart Lung Transplant*. 2005;24(11):1710–1720.
6. Berry GJ, Burke MM, Andersen C, et al. The 2013 International Society for Heart and Lung Transplantation Working Formulation for the

standardization of nomenclature in the pathologic diagnosis of antibody-mediated rejection in heart transplantation. *J Heart Lung Transplant*. 2013;32(12):1147–1162.
7. Mehra MR, Crespo-Leiro MG, Dipchand A, et al. International Society for Heart and Lung Transplantation working formulation of a standardized nomenclature for cardiac allograft vasculopathy—2010. *J Heart Lung Transplant*. 2010;29(7):717–727.
8. Feingold B, Bowman P, Zeevi A, et al. Survival in allosensitized children after listing for cardiac transplantation. *J Heart Lung Transplant*. 2007; 26(6):565–571.
9. Feingold B, Park SY, Comer DM, Moore CG, Webber SA, Bryce CL. Outcomes after listing with a requirement for a prospective crossmatch in pediatric heart transplantation. *J Heart Lung Transplant*. 2013;32(1):56–62.
10. Tran A, Fixler D, Huang R, Meza T, Lacelle C, Das BB. Donor-specific HLA alloantibodies: impact on cardiac allograft vasculopathy, rejection, and survival after pediatric heart transplantation. *J Heart Lung Transplant*. 2016;35(1):87–91.
11. Irving C, Carter V, Parry G, Hasan A, Kirk R. Donor-specific HLA antibodies in paediatric cardiac transplant recipients are associated with poor graft survival. *Pediatr Transplant*. 2011;15(2):193–197.
12. Zhang R. Donor-specific antibodies in kidney transplant recipients. *Clin J Am Soc Nephrol*. 2018;13(1):182–192.
13. Zhang Q, Hickey M, Drogalis-Kim D, et al. Understanding the correlation between DSA, complement activation, and antibody-mediated rejection in heart transplant recipients. *Transplantation*. 2018;102(10):e431–e438.
14. Clerkin KJ, Farr MA, Restaino SW, et al. Donor-specific anti-HLA antibodies with antibody-mediated rejection and long-term outcomes following heart transplantation. *J Heart Lung Transplant*. 2017;36(5): 540–545.
15. Ho EK, Vlad G, Vasilescu ER, et al. Pre- and posttransplantation allosensitization in heart allograft recipients: major impact of de novo alloantibody production on allograft survival. *Hum Immunol*. 2011;72(1): 5–10.
16. Sommer W, Avsar M, Aburahma K, et al. Heart transplantation across preformed donor-specific antibody barriers using a perioperative desensitization protocol. *Am J Transplant*. 2022;22(8): 2064–2076.
17. Aversa M, Martinu T, Patriquin C, et al. Long-term outcomes of sensitized lung transplant recipients after peri-operative desensitization. *Am J Transplant*. 2021;21(10):3444–3448.
18. Sharma M, Webber SA, Zeevi A, Mohanakumar T. Molecular events contributing to successful pediatric cardiac transplantation in HLA sensitized recipients. *Hum Immunol*. 2019;80(4):248–256.
19. Narayanan K, Jaramillo A, Phelan DL, Mohanakumar T. Pre-exposure to sub-saturating concentrations of HLA class I antibodies confers resistance to endothelial cells against antibody complement-mediated lysis by regulating Bad through the phosphatidylinositol 3-kinase/Akt pathway. *Eur J Immunol*. 2004;34(8):2303–2312.
20. Fukami N, Ramachandran S, Narayanan K, et al. Mechanism of accommodation in a sensitized human leukocyte antigen transgenic murine cardiac transplant model. *Transplantation*. 2012;93(4): 364–372.
21. Koch CA, Khalpey ZI, Platt JL. Accomodation: preventing injury in transplantation and disease. *J Immunol*. 2004;172(9):5143–5148.