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A Novel Method of Measuring Cardiac Preservation Injury Demonstrates University of Wisconsin Solution Is Associated with Less Ischemic Necrosis than Celsior in Early Cardiac Allograft Biopsies

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Abstract

Purpose—No consensus exists on the optimal heart preservative solution (HPS) for cardiac allograft preservation. The significance of varying degrees of acute ischemic necrosis (AIN) in early transplant biopsies is unknown. We investigated the effects of HPS on early cardiac histopathology by developing a novel grading system of AIN.

Methods—We retrospectively reviewed our institutional database of orthotopic heart transplants (OHT) identifying all hearts preserved with University of Wisconsin (UW) or Celsior (CS) solutions. A single blinded cardiovascular pathologist graded AIN severity on early post-transplant biopsies. Primary stratification was by HPS. Multivariable models examined mortality, AIN grade, primary graft dysfunction (PGD) and right heart failure (RHF).

Results—From 1996–2010, 174 adult OHT were preserved with UW (42) or CS (132) from which 431 biopsies were reviewed. UW and CS had similar 30-day ($p=0.79$) and 1-year mortality ($p=0.92$). CS was associated with significantly more AIN on the 1st ($p=0.02$) and 2nd ($p=0.04$) biopsies. This association persisted on multivariable analysis (1st biopsy OR: 2.93[1.26–6.83], $p=0.01$ and 2nd biopsy OR: 2.08[0.99–4.34], $p=0.05$). When stratified by AIN score, 30-day and 1-year mortality were similar ($p>0.05$). However, on adjusted analysis, increasing AIN score on the 1st biopsy was strongly associated with an increased incidence of PGD (OR: 1.59[1.02–2.47], $p=0.04$) and RHF (OR: 2.45[1.14–5.27], $p=0.02$).

Conclusions—Our novel grading system provides a simple, reproducible method for determining AIN. Preservation with UW is associated with less AIN than CS solution. Early biopsy ischemia is associated with primary graft dysfunction and right heart failure. AIN may have prognostic significance and its routine evaluation should be considered.

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Keywords

Heart transplantation; acute ischemic necrosis; heart preservative solution

INTRODUCTION

Orthotopic heart transplantation (OHT) remains the gold-standard for the treatment of end-stage heart failure. However, cardiac allograft failure continues to be a problem.¹ One cause of allograft dysfunction is suboptimal preservation, resulting in cardiac ischemia and subsequent cellular necrosis.^{1, 2} Although the foundation of contemporary allograft preservation is hypothermia, many heart preservative solutions (HPS) exist to supplement the protective effects of hypothermic storage.³ Despite substantial clinical and basic science research, no optimal HPS has been identified. In the United States alone, 167 different types of HPS have been utilized.⁴ Only 55% of these are standardized and some individual transplant centers use as many as three different HPS.⁴

Although many different HPS solutions are currently in use, transplant centers are increasingly using standardized, commercially available solutions. The two most commonly utilized HPS in the United States are the University of Wisconsin solution (UW; ViaSpan, DuPont Pharmaceuticals, Wilmington, DE) and the Celsior solution (CS; Genzyme Corporation, Cambridge, MA). Recently, in a large registry study, we reported that compared to CS, preservation with UW was associated with decreased 30-day and 1-year mortality in all OHT recipients and particularly in those receiving high-risk allografts.⁵ However that study was unable to provide significant insight into the physiological reason for the survival benefit associated with UW.

Acute ischemic necrosis (AIN) is often observed on routine early post-transplant biopsies. Although AIN on early post-transplant biopsies is thought to be related to the ischemic insult suffered during hypothermic ischemic preservation, its significance is not well understood.² We undertook this study to evaluate the impact of HPS on both the frequency and severity of AIN in early post-transplant biopsies. We developed a novel scoring system to provide a simple, reproducible method of grading the severity of AIN.

METHODS

Study Design

We conducted a retrospective review of our prospectively maintained OHT database. Our study included all adult (≥ 18 years) OHT from 1996–2010. All patients who received allografts preserved with either UW or CS were included. Pediatric recipients and combined heart-lung recipients were excluded. Our institutional review board approved this study.

We examined pertinent variables in our data set, including: recipient demographics and co-morbidities, measures of recipient acuity including hemodynamics and need for inotropic or mechanical circulatory support, donor demographics and co-morbidities, and transplant variables.

All donor hearts were procured using standard techniques, regardless of the HPS utilized. Diastolic arrest of the heart was accomplished with 30 mL/kg of HPS. Allografts were subsequently excised and placed in 2 liters of cold HPS for storage and transport. Throughout implantation, the hearts were kept in the pericardial well covered with saline slush. The allograft is not routinely re-flushed with HPS during the implantation procedure.

Histopathology

Post-transplant cardiac biopsies were evaluated by a single, blinded cardiac pathologist (MKH). Our institutional practice includes routine weekly post-transplant biopsies for the first month, as the patient's clinical status permits. As previous research has shown that the AIN caused by transplantation peaks around 16 days post-transplant, we evaluated the first three post-transplant biopsies to capture this peak.⁶

Since a grading system of AIN does not currently exist, a novel scale of ischemic severity was created (Table 1). The scale ranges from 0–3 with a score of 0 representing no AIN and a score of 3 representing severe AIN. Post-transplant biopsies were evaluated under 10X and 40X power and graded according to this scale. Representative pictures of each grade of AIN are depicted in Figure 1.

AIN was assessed based on myocyte injury and/or evidence of reparative processes. Mild AIN (Grade 1) was comprised of minor myocyte ischemic injury (~5 myocytes) showing nuclear loss, myocyte necrosis, and/or mild macrophage infiltration occurring on a single biopsy piece. Moderate AIN (Grade 2) was based on multifocal areas or a single large area of myocyte ischemic injury – macrophage infiltration and myocyte loss that was easily seen at low power but not extensive. Severe AIN (Grade 3) was based upon extensive injury to the myocardium with large areas across multiple biopsy pieces that had loss of cardiac myocytes, macrophage infiltration, and an active reparative process. Note that AIN is distinct from cellular (lymphocytic infiltration) and antibody-mediated (vascular injury) processes.

Each of the first three post-transplant biopsies was evaluated individually. Since cumulative AIN over the first three weeks post-transplant may be important, composite scores of all three biopsies including mean ischemic grade and maximum ischemic grade were also evaluated.

Outcomes

Impact of Preservative Solution—We stratified patients according to the HPS used. Primary clinical endpoints included 30-day, 90-day, and 1-year mortality. Secondary clinical endpoints included prolonged primary graft dysfunction (PGD) and right heart failure (RHF). To capture severe allograft dysfunction, PGD was defined as the need for post-transplant epinephrine support for ≥ 10 days or the need for post-transplant left ventricular assist device (LVAD) support. RHF was defined as the need for post-transplant right ventricular assist device (RVAD) support.

The impact of HPS on ischemic grade was evaluated at each of the three post-transplant biopsies and by the composite scores. Multivariable ordered logistic regression models were constructed to evaluate the impact of HPS and other covariates on ischemic grade.

Impact of Acute Ischemic Necrosis Score—Patients were stratified by their ischemic scores. The impact of ischemic score on 30-day, 90-day, and 1-year mortality was evaluated. Multivariable logistic regression models were constructed to determine the impact of ischemic grade on both PGD and RHF.

Statistical Analysis

We compared baseline characteristics among patients in the UW and CS cohorts by the t-test (continuous parametric variables), the Wilcoxon rank-sum test (continuous non-parametric variables), and the chi-square test or Fisher's exact test (categorical variables) as appropriate. Survival was estimated by the Kaplan-Meier method.

To construct multivariable models, all independent covariates were tested in univariate fashion. Variables associated with the outcome measured on exploratory analysis ($p < 0.20$), those with biological plausibility, and those previously reported in the literature to be significant were incorporated in a forward and backward stepwise fashion into the multivariable models. The likelihood ratio test and Akaike's information criterion were utilized in a nested model approach to identify which model had the greatest explanatory power.

For all analyses, values of $p < 0.05$ (2-tailed) were considered statistically significant. Mean values are displayed with standard deviations and median values are displayed with their interquartile ranges (IQR). Hazard ratios (HR) and odds ratios (OR) are presented with their 95% confidence intervals (CI). Statistical analysis was performed using STATA 11.2 (StataCorp, College Station, TX).

RESULTS

Cohort Statistics

From 1996–2010, 326 OHT were performed. We excluded pediatric transplants ($n=40$), combined heart-lung transplants ($n=9$), and allografts not preserved with either UW or CS ($n=103$). Our final cohort comprised 174 patients, 42 allografts preserved with UW and 132 preserved with CS.

The mean age of our final cohort was 49 ± 13 years and 123 (70.7%) were male. Forty-five (25.5%) patients were bridged to transplantation with an LVAD; 27 (15.5%) had a pulsatile device and 18 (10.3%) a continuous flow device. One hundred thirty-three (52.8%) involved a redo sternotomy and 26 (10.32%) were redo-OHT. The mean ischemic time was 3.0 ± 1.1 hours.

In our study cohort, 1-year mortality was 19.5%, 18.1% for primary OHT and 35.7% for redo-OHT. Postoperatively, 5 (2.9%) patients required LVAD support, 14 (8.0%) required RVAD support, and 40 (23.0%) met our definition of PGD.

Baseline Characteristics

When stratified by HPS, while most baseline characteristics were similar between the two cohorts, there were several differences (Table 2). Patients whose allograft was preserved with CS were more likely to be male and to have hypertension. Although the groups had similar ejection fractions, the CS group had lower pulmonary artery pressures, lower pulmonary capillary wedge pressures, was more likely to be bridged to transplant with a VAD, less likely to be hospitalized at the time of transplant, and tended to have a longer waitlist time. Importantly, the mean ischemic time was similar between the two groups.

Outcomes

Impact of Preservative Solution on Outcomes—When stratified by HPS, there was no difference between UW and CS in unadjusted survival at 30-days (92.9% [79.5–97.6] vs. 90.9% [84.6–94.7], $p=0.79$), 90-days (87.9% [73.3–94.8] vs. 85.6% [78.4–90.6], $p=0.79$), or at 1-year (79.5% [63.1–89.2] vs. 80.3% [72.4–86.1], $p=0.92$; Figure 2). On adjusted analysis, CS was associated with a trend towards an increased hazard of mortality at 90-days (HR: 1.78 [0.21–15.17], $p=0.60$), and 1-year (HR: 3.01 [0.37–24.37], $p=0.30$; Table 3). The adjusted hazard of mortality was similar at 30-days (HR: 1.05 [0.10–10.97], $p=0.97$).

Compared with the UW cohort, in patients receiving allografts preserved with CS there was a trend toward a greater incidence of PGD (5/42 (11.9%) vs. 35/132 (26.5%), $p=0.059$) and a

higher incidence of RHF (0/42(0.0%) vs. 14/132 (10.6%), $p=0.02$). Additionally, the two cohorts were not significantly different in regards to length of stay (12[10–17] vs. 15[9–24.5], $p=0.32$), need for reoperation for bleeding (8/42(19.1%) vs. 43/132(32.6%), $p=0.09$), and incidence of renal failure requiring dialysis (2/42(4.8%) vs. 13/132 (9.9%), $p=0.53$).

Impact of Preservative Solution on Ischemic Score—Overall, 431 post-transplant biopsies were evaluated. Eleven patients died before a biopsy could be performed. There was a trend towards more deaths in the CS cohort (1/42(2.4%) vs. 10/132(7.6%), $p=0.23$). While there is some variability in the exact timing of the biopsy, the median time to each of the first 3 biopsies was 8[7–10], 16[14–20], and 24[22–30] days respectively. The time to each biopsy was similar between the two cohorts ($p=0.42$, $p=0.16$, $p=0.12$ respectively).

Of the 431 biopsies, 123(28.5%) were scored as grade 0, 155(36.0%) grade 1, 96(22.3%) grade 2, and 57(13.2%) grade 3. When stratified by preservative solution, CS was associated with more AIN than UW. On the 1st post-transplant biopsy, CS was less likely to demonstrate no AIN (15/28(53.6%) vs. 35/116(30.2%), $p=0.02$) and more likely to demonstrate moderate AIN (1/28(3.6%) vs. 23/116(19.8%), $p=0.046$; Figure 3). Similarly, on the 2nd biopsy, CS was more likely to demonstrate severe AIN (0/30(0.0%) vs. 20/113(17.7%), $p=0.008$). Finally, on the 3rd biopsy, though no differences reached statistical significance, CS tended to be less likely to exhibit no AIN (11/30(36.7%) vs. 26/114(22.8%), $p=0.12$) and more likely to exhibit moderate AIN (3/30(10.0%) vs. 32/114(28.1%), $p=0.054$).

On unadjusted analysis, UW was associated with less median AIN on the 1st biopsy ($p=0.02$), the 2nd biopsy ($p=0.04$), and a trend toward less on the 3rd biopsy ($p=0.08$; Table 4). Additionally, when composite scores were generated to account for potential sampling error and the possibility of cumulative necrosis over time, UW was again associated with less AIN than CS as measured by the mean ischemic score ($p=0.01$) and the maximum ischemic score ($p=0.002$).

To adjust for potential confounders, a multivariable ordered logistic regression model was constructed. On multivariable analysis, preservation with CS was associated with an increased risk of AIN on the 1st biopsy (OR: 2.93[1.26–6.83], $p=0.01$; Table 5). Moreover, CS tended to increase the AIN associated with the 2nd (OR: 2.08[0.99–4.34], $p=0.052$), and 3rd (2.00[0.93–4.29], $p=0.08$) biopsies. When composite measures of AIN were analyzed, preservation with CS was associated with increased AIN as measured by the mean ischemic score (OR: 2.51[1.25–5.09], $p=0.01$) and by the maximum ischemic score (OR: 3.43[1.64–7.19], $p=0.001$).

Impact of Ischemic Necrosis Grade on Outcomes—When stratified by ischemic grade on the 1st biopsy, mean ischemic grade across all biopsies, or maximum ischemic grade across all biopsies, there were no differences in 30-day, 90-day, or 1-year survival. Patients who met our definition of PGD had higher AIN scores on their first biopsy (1[0–1] vs. 1[1–2], $p=0.03$) but similar scores on their 2nd ($p=0.38$) and 3rd ($p=0.89$). After adjusting for recipient age, donor age, and ischemic time, increasing ischemic grade on the 1st biopsy was associated with an increased incidence of PGD (OR: 1.59[1.02–2.47], $p=0.04$; Table 6). Although ischemic grades on the 2nd ($p=0.37$) and 3rd ($p=0.87$) biopsy were not predictive of PGD, there was a strong trend toward increased PGD as measured by mean ischemic grade (OR: 1.44[0.80–2.59], $p=0.22$) and maximum ischemic grade (OR: 1.57[0.97–2.53], $p=0.07$).

Postoperatively, 5 (2.9%) patients required LVAD support, 14 (8.0%) required RVAD support. There was a strong trend toward increased AIN in patients requiring LVAD (OR:

3.00[0.69–13.08], $p=0.15$) as measured by the first biopsy. After adjusting for recipient age, donor age, and ischemic time, this association strengthened (OR: 5.36[0.64–44.60], $p=0.12$). Patients who required a postop RVAD had higher AIN scores on their first biopsy (1[0–1] vs. 2[1–3], $p=0.02$) but not on their 2nd (0.57) or 3rd (0.54) biopsies. Therefore, there was significantly more AIN in those requiring RVAD support (OR: 2.54[1.25–5.18], $p=0.01$) as measured by the first biopsy score. This relationship persisted on multivariable analysis (OR: 2.45[1.14–5.27], $p=0.02$; Table 7). Ischemic grades on the 2nd ($p=0.59$) and 3rd ($p=0.54$) biopsies were not predictive of RHF. However, there was a strong trend toward increased RHF as measured by mean ischemic grade (OR: 2.49[0.87–7.11], $p=0.09$) and maximum ischemic grade (OR: 2.61[0.97–7.06], $p=0.06$).

DISCUSSION

In our study, although patients receiving allografts preserved with UW and CS have similar short-term mortality, patients in the CS cohort have an increased incidence of RHF requiring RVAD support and a trend toward an increased need for LVAD support. Moreover, allografts preserved with UW exhibited less AIN on early post-transplant biopsies than those preserved with CS as measured by our histopathological assessment. Finally, AIN as measured by our novel scoring system is predictive of both PGD and RHF.

Numerous HPS are currently used to augment hypothermic protection during explant, transport, and implantation of cardiac allografts. In the United States, UW and CS are the most commonly utilized HPS. UW, originally designed for kidney and liver transplantation, is classified as an intracellular solution for its high potassium concentration. Theoretical benefits of high potassium concentration include rapid cardiac mechanical arrest leading to less intracellular edema and less ionic flux.^{3, 4} Additionally, UW contains adenosine to maintain ATP-dependent ion gradients and free radical scavengers such as lactobionate and allopurinol to mitigate ischemia-reperfusion injury.⁷ Moreover, several large studies have shown superior outcomes of intracellular over extracellular solutions.^{4, 5} However, many clinicians and researchers are reticent to use such a high potassium solution, particularly with regard to its putative but controversial effect on coronary and pulmonary vascular endothelium.^{2, 3, 8}

Celsior, designed specifically for myocardial preservation, is classified as an extracellular solution for its relatively low potassium concentration. Theoretic benefits of CS include numerous specific additives, including mannitol, histidine, and glutathione, to mitigate ischemia-reperfusion injury, impermeants to reduce intra and extracellular swelling, glutamate to enhance energy production, a mild acidosis to prevent calcium overload, and the prevention of red blood cell aggregation.^{1, 7, 9, 10} However, several studies have demonstrated that more complex additives, such as glutathione, are unstable.^{11, 12} While both HPS contain glutathione, it is possible that CS, with its more numerous and complex additives, may not be as stable mandating optimal storage conditions of these solutions for favorable clinical outcomes.

Although there is no consensus on the optimal HPS at present, our group recently published a large registry study of almost 5,000 OHT demonstrating that patients who received an allograft preserved with UW had significantly greater short term survival than those who received an allograft preserved with CS.⁵ While one could speculate that this survival difference suggests that UW is a better preservative solution which more optimally supplements the effects of hypothermic storage, a large registry study precludes evaluation of preservation at the histopathological level. Therefore, we undertook the current study to better define the preservative ability of these two solutions at the cellular level.

To evaluate the impact of HPS on cardiac histopathology, we evaluated early post-transplant biopsies taking into account our previous finding of peak histologic injury occurring 16 days post-transplant.⁶ It is our institutional practice to obtain weekly cardiac biopsies for the first month after OHT; therefore, we evaluated the first 3 post-transplant biopsies in order to capture peak AIN. One prior study has evaluated the impact of HPS on AIN.² However, in their study, AIN was evaluated in binary fashion. Our experience suggests that AIN is not simply a binary phenomenon. Not only can ischemia be present or absent, but there is distinct variability in the severity of necrosis. Since this variable severity of ischemia may reflect the adequacy of preservation during OHT, we created a novel yet simple grading scale for AIN. To account for the possibility of sampling error and the potential delay between ischemic insult and pathologic evidence of ischemic necrosis, we also evaluated composite scores to account for AIN as it was observed over all 3 post-transplant biopsies.

On both unadjusted and adjusted analysis, allografts preserved with UW exhibited less AIN than those preserved with CS. The decreased AIN associated with UW combined with our previous finding that UW is associated with superior survival compared to CS has persuaded us to use UW exclusively for OHT. However, we recognize that this debate is likely to continue.

To further validate our novel grading system of AIN, the impact of ischemic grade on outcomes was analyzed. When stratified by ischemic grade on the first biopsy, although there was no difference in terms of short term mortality, we believe this was due to the relatively small sample size of the four groups. However, ischemic grade as measured at the 1st biopsy was strongly predictive of PGD and the need for post-transplant LVAD and RVAD support. While it is not surprising that increased ischemic necrosis is predictive of poor allograft function, the fact that our ischemic grading system predicts poor graft function serves to validate our novel ischemic score.

There is no consensus definition of PGD after OHT. Existing definitions encompass varying degrees of inotropic or mechanical circulatory support over a widely variable duration. In an attempt to validate our score, we wanted to restrict our definition of PGD to those allografts with severe dysfunction and therefore limited our definition to include implantation of an LVAD or prolonged epinephrine support (≥ 10 days). Many other definitions have been proposed, pointing to the need for a consensus definition.

Limitations

Our study utilizes a simple, novel schema for grading AIN. Since it is a novel system, it would require further validation in a different cohort before it could be widely applied. Our AIN scores were determined by a single cardiovascular pathologist blinded to the HPS used for preservation. This could be seen as a potential source of bias. However, the grading schema is simple and similar to currently accepted grading systems and could be reproduced at any center performing cardiac transplantation. Further research will be necessary to evaluate the generalizability of our results.

As a retrospective review, patients were not randomly assigned to HPS. Although HPS was ultimately according to surgeon preference, in general, HPS choice was time-dependent with CS utilization predominating from 2001–2008 and UW predominating before and since that time period. Despite attempts to guard against bias with multivariable analysis, we recognize this time-dependent utilization of HPS to be a potential confounder. Although we did not replicate the differential survival between HPS demonstrated in our previous research, we suspect this is due to the limited sample size of our cohort.

CONCLUSION

In conclusion, UW is associated with less AIN than CS as measured by a novel scoring system. Increasing ischemic scores are strongly associated with poor allograft function as measured by PGD and RHF. Additionally, increased AIN was associated with the need for post-transplant LVAD and RVAD support, further validating the prognostic ability of our scoring system. Our study suggests that UW is a better HPS than CS and that our novel method of ischemic scoring is a valid predictor of outcomes. These findings provide a histopathologic rationale to the superior clinical outcomes previously associated with UW. Coupled with these earlier findings, we believe that a renewed focus on the optimal HPS is warranted and routine evaluation of AIN on early post-transplant biopsies should be considered.

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Figure 1.
Representative light micrographs of post-transplant biopsies for each ischemic grade. Ischemic grade 0 (None) at 10X (A) and 40X (B) power. Ischemic grade 1 (Mild) at 10X (C) and 40X (D) power. Ischemic grade 2 (Moderate) at 10X (E) and 40X (F) power. Ischemic grade 3 (Severe) at 10X (G) and 40X (H) power.

Figure 2.
1-Year Kaplan-Meier survival estimates stratified by heart preservative solution.

Figure 3. Distribution of ischemic necrosis scores stratified by preservative solution on the 1st biopsy, 2nd biopsy, and 3rd biopsy.

Table 1

Novel Acute Ischemic Necrosis Scoring System

Ischemic Grade	Severity	Description
0	None	No evidence of ischemic necrosis.
1	Mild	Several focal myocytes with injury or at most 2 areas with approximately 5 myocytes injured.
2	Moderate	Multiple small foci of injury or a single large area of injury with the rest of the biopsy normal.
3	Severe	Marked injury across 1 or more biopsy.

Table 2

Baseline Characteristics Stratified by Heart Preservative Solution

Variable	UW (n=42)	Celsior (n=132)	P-value*
Demographics and Co-Morbidities			
Age (years)	48.45 $\pm$ 1.79	48.86 $\pm$ 1.18	0.86
Male	23/42 (54.76%)	100/132 (75.76%)	<0.01
White ethnicity	32/42 (76.19%)	92/132 (69.70%)	0.42
Black ethnicity	9/42 (21.43%)	30/132 (22.73%)	0.86
Diabetes	12/42 (28.57%)	36/132 (27.27%)	0.87
Hypertension	14/42 (33.33%)	78/132 (59.09%)	<0.01
Creatinine (mg/dL)	1.15 $\pm$ 0.06	1.34 $\pm$ 0.06	0.06
BMI (kg/m ²)	27.07 $\pm$ 0.65	27.16 $\pm$ 0.44	0.91
Ischemic CM	14/42 (33.33%)	42/132 (31.82%)	0.86
Acuity			
Ejection Fraction (%)	23.69 $\pm$ 3.16	20.42 $\pm$ 1.28	0.28
Cardiac output (L/min)	3.83 $\pm$ 0.27	4.38 $\pm$ 0.12	0.07
Cardiac index (L/min/m ²)	2.08 $\pm$ 0.13	2.25 $\pm$ 0.06	0.28
Mean PA pressures (mmHg)	31.64 $\pm$ 2.61	25.44 $\pm$ 0.98	0.01
PCWP (mmHg)	21.32 $\pm$ 1.70	16.40 $\pm$ 0.86	0.02
Preop Inotropes	16/42 (38.10%)	34/132 (25.76%)	0.12
Preop IABP	1/42 (2.38%)	2/132 (1.81%)	0.76
Preop VAD	5/42 (11.90%)	40/132 (30.30%)	0.02
Preop Vent	0/42 (0%)	1/132 (0.76%)	0.54
Hospitalized	14/42 (33.33%)	21/132 (15.91%)	0.01
ICU	6/42 (14.29%)	9/132 (6.82%)	0.13
Donor and Transplant Variables			
Age (years)	30.61 $\pm$ 2.11	33.14 $\pm$ 1.07	0.26
Male	20/41 (48.78%)	86/132 (65.15%)	0.06
Race matched	26/42 (61.90%)	72/132 (54.55%)	0.40
Sex matched	28/42 (58.33%)	96/132 (72.73%)	0.45
Cigarette Use	15/34 (44.12%)	51/123 (41.46%)	0.78
CMV mismatch	19/42 (45.24%)	59/132 (44.70%)	0.95
Redo sternotomy	20/42 (47.62%)	68/132 (51.52%)	0.87
Redo OHT	2/42 (4.76%)	12/132 (9.09%)	0.53
Time on waitlist (days)	181 [74–381]	400 [95–671]	0.02
Ischemic time (hours)	2.79 $\pm$ 1.06	3.10 $\pm$ 1.04	0.09

* P-Value based on results of Student's t-test (parametric continuous variables), Wilcoxon rank-sum test (non-parametric continuous variables) or the Chi square test (categorical variables)

Abbreviations: UW, University of Wisconsin; CS, Celsior; BMI, body mass index; CM, cardiomyopathy; PA, pulmonary artery; PCWP, pulmonary capillary wedge pressure; IABP, intra-aortic balloon pump; VAD, ventricular assist device; ICU, intensive care unit; CMV, cytomegalovirus.

Table 3**1-Year Mortality Cox Hazards Regression Model**

Variable	Univariate		Multivariable	
	HR [95% CI]	p-value ^a	HR [95% CI]	p-value ^b
Celsior	0.98 [0.44–2.18]	0.97	3.01 [0.37–24.37]	0.30
Recipient Variables				
Age (per year)	1.00 [0.98–1.02]	0.87	1.00 [0.97–1.03]	0.89
Creatinine (per mg/dL)	1.91 [1.34–2.72]	<0.001	1.81 [1.07–3.06]	0.03
CI (per L/min/m ²)	0.85 [0.46–1.58]	0.61	0.67 [0.27–1.63]	0.38
mPAP (per mmHg)	1.00 [0.97–1.04]	0.80	1.01 [0.96–1.06]	0.84
ICU support	2.14 [1.11–4.14]	0.02	3.66 [1.22–11.03]	0.02
No BTT	1 (Reference)		1 (Reference)	
BTT (pulsatile)	1.43 [0.72–2.81]	0.31	1.25 [0.34–4.64]	0.74
BTT (continuous flow)	1.68 [0.66–4.28]	0.28	1.74 [0.36–8.36]	0.49
Donor and Transplant Variables				
Donor age (per year)	1.03 [1.004–1.05]	0.02	1.01 [0.97–1.05]	0.66
Redo OHT	1.30 [0.55–3.06]	0.55	1.32 [0.29–6.05]	0.73
Same sex pairing	1.00 [0.56–1.79]	0.99	1.40 [0.47–4.16]	0.54
Ischemic time (per hour)	1.33 [1.003–1.76]	0.047	1.51 [0.92–2.49]	0.11

^aP-value based on univariate Cox proportional hazard analysis

^bP-value based on multivariable Cox proportional hazards analysis.

Abbreviations: HR, hazards ratio; CI, confidence interval; CI, cardiac index; mPAP, mean pulmonary artery pressure; ICU, intensive care unit; BTT, bridge to transplantation; OHT, orthotopic heart transplantation.

Table 4

Grade of Ischemic Necrosis Stratified by Preservative Solution

Biopsy	UW (n=42) Median [IQR]	CS (n=132) Median [IQR]	p-value^a
1 st biopsy	0.0 [0.0–1.0]	1.0 [0.0–2.0]	0.02
2 nd biopsy	1.0 [0.0–1.0]	1.0 [1.0–2.0]	0.04
3 rd biopsy	1.0 [0.0–2.0]	1.0 [1.0–2.0]	0.08
Composite Scores			
Mean ischemic score ^b	1.3 [0.7–2.3]	2.0 [1.3–3.3]	0.01
Max ischemic score ^c	1.0 [1.0–2.0]	2.0 [1.0–3.0]	0.002

^aP-value determined by the Wilcoxon rank-sum test^bScore reflects mean score across all 3 post-transplant biopsies^cScore reflects maximum score across all 3 post-transplant biopsies

Abbreviations: UW, University of Wisconsin; CS, Celsior; IQR, interquartile range;

Table 5Multivariable Ordered Logistic Regression Model of Ischemic Necrosis on the 1st Post-transplant Biopsy

Variable	Univariate		Multivariable	
	OR [95% CI]	p-value ^a	OR [95% CI]	p-value ^b
Celsior	2.70 [1.21–6.01]	0.02	2.93 [1.26–6.83]	0.01
Recipient Variables				
Age (per year)	0.99 [0.97–1.01]	0.39	0.99 [0.97–1.01]	0.41
White ethnicity	1 (Reference)		1 (Reference)	
Black ethnicity	1.37 [0.65–2.91]	0.41	1.40 [0.62–3.17]	0.41
Other ethnicity	0.81 [0.25–2.70]	0.74	0.60 [0.17–2.15]	0.43
Creatinine (per mg/dL)	0.54 [0.31–0.94]	0.03	0.47 [0.25–0.87]	0.02
Donor and Transplant Variables				
Donor age (per year)	1.00 [0.98–1.02]	0.98	1.00 [0.98–1.03]	0.76
Ischemic time (per hour)	1.24 [0.91–1.67]	0.17	1.31 [0.96–1.78]	0.093

^aP-value determined by univariate ordered logistic regression.^bP-value determined by multivariable ordered logistic regression model.

Abbreviations: OR, odds ratio; CI, confidence interval.

Table 6**Multivariable Logistic Regression Model of Primary Graft Dysfunction**

Variable	Univariate		Multivariable	
	OR [95% CI]	p-value ^a	OR [95% CI]	p-value ^b
1 st Biopsy Ischemic Grade (per point)	1.62 [1.05–2.50]	0.03	1.59 [1.02–2.47]	0.04
Recipient age (per year)	1.00 [0.97–1.03]	0.99	0.98 [0.96–1.01]	0.31
Donor age (per year)	1.01 [0.98–1.04]	0.56	1.01 [0.97–1.04]	0.75
Ischemic time (per hour)	1.26 [0.90–1.76]	0.17	0.95 [0.63–1.43]	0.80

^aP-value determined by univariate logistic regression.

^bP-value determined by multivariable logistic regression model.

Abbreviations: OR, odds ratio; CI, confidence interval.

Table 7**Multivariable Logistic Regression of Right Heart Failure**

Variable	Univariate		Multivariable	
	OR [95% CI]	p-value ^a	OR [95% CI]	p-value ^b
1 st Biopsy Ischemic Grade (per point)	2.54 [1.25–5.18]	0.01	2.45 [1.14–5.27]	0.02
Recipient age (per year)	1.04 [0.99–1.09]	0.15	1.01 [0.95–1.07]	0.78
Donor age (per year)	1.01 [0.97–1.06]	0.62	1.02 [0.96–1.08]	0.53
Ischemic time (per hour)	1.76 [1.07–2.91]	0.03	2.00 [1.01–3.96]	0.046

^aP-value determined by univariate logistic regression.

^bP-value determined by multivariable logistic regression model.

Abbreviations: OR, odds ratio; CI, confidence interval.