Intraoperative Verapamil Fails to Reduce Delayed Graft Function in Donation After Circulatory Death Renal Allografts

Jessica T. Lovett, BS,[1] Jeffrey Stern, MD,[2,3] Elaina P. Weldon, MSN, ACNP-BC,[2] Bonnie E. Lonze, MD, PHD,[2,3] and Zoe A. Stewart, MD, PhD[2,3]

Background. The shortage of transplantable organs has led to increased utilization of kidneys that may be particularly vulnerable to ischemia-reperfusion injury (IRI) and delayed graft function (DGF). Kidneys from donation after circulatory death (DCD) donors have additional IRI from donor procurement that results in increased risk of DGF. Verapamil may reduce IRI in kidney allografts when given at the time of organ reperfusion. This study sought to determine if intraoperative administration of verapamil (Ver) could reduce the risk of DGF in DCD kidney transplants. **Methods.** A single-center retrospective matched cohort study was performed of 93 Ver (–) kidney transplant recipients compared with 93 Ver (+) kidney transplant recipients, matched by donor age, Kidney Donor Profile Index, and DCD status. Covariates that could impact DGF risk were evaluated by univariate and multivariate logistic regression analyses. **Results.** The Ver (–) and Ver (+) matched cohorts did not have any significant differences in the demographic covariates. There was no difference in DGF rate between the Ver cohorts in either the overall study population or within the DCD subgroup. There was a trend toward reduced DGF in the Ver (+) cohort for cold ischemia time (CIT) ≤24 h, but this failed to achieve statistical significance. On multivariate analysis, only CIT was found to be independently associated with DGF. **Conclusions.** Intraoperative verapamil failed to reduce DGF risk in DCD kidney allografts. Limitations to this study include nonrandomization for the intraoperative administration of verapamil and the mean CIT >24 h in the study population. Only CIT was an independent prognosticator for DGF on multivariate analysis in a cohort matched for DCD status, consistent with prior studies.

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INTRODUCTION

The pervasive shortage of donor kidneys to meet the demand of the kidney transplant waiting list has led to increased utilization of donation after circulatory death (DCD) kidneys for transplant.[1] In the past decade, the number of DCD donors has tripled in the United States, and DCD donors now comprise 23% of the deceased donor pool.[2] DCD kidneys have increased vulnerability to ischemia-reperfusion injury (IRI) as a consequence of the procurement warm ischemia time (WIT). DCD donor status is a well-described risk factor for delayed graft function (DGF) after kidney transplantation.[3-5] Pulsatile preservation may reduce the risk of DGF in DCD kidneys, although this may be dependent on the total procurement WIT[4] and cold ischemia time (CIT).[3,6]

IRI has been extensively studied in experimental kidney transplant models.[7] Intracellular calcium signaling plays a critical role during IRI by impairing mitochondrial function, contributing to renal tubular apoptosis.[8-14] Several studies have examined the impact of perioperative verapamil on calcium-mediated IRI in kidney transplantation.[9,15-21] The addition of verapamil to the cold perfusate during kidney procurement resulted in improved renal function at 6 and 12 mo posttransplant, although there was no difference in the

risk of DGF between the Ver (+) and Ver (−) groups.[19] Other studies have used intraoperative renal artery verapamil injection immediately postreperfusion in a single-pass model in an attempt to mitigate calcium-mediated IRI.[16,18,21] Intra-arterial verapamil significantly reduced the incidence of DGF in early reports.[16,18] A more recent single-center study failed to demonstrate that intraoperative verapamil reduced DGF.[21] However, this latter study did observe that only 60% of the DCD Ver (+) group had DGF, in contrast to 80% in the DCD Ver (−) group.[21] Unfortunately, the small sample size and short CIT in the DCD groups limited the generalizability of this conclusion.

In this study, we sought to take advantage of our program's high-volume utilization of DCD kidney allografts in a practice model in which intraoperative verapamil administration was routinely performed in deceased donor transplants. The goal of this study was to determine if the intraoperative use of verapamil could reduce the risk of DGF in DCD kidney transplants. Herein, we report the outcomes of the largest DCD cohort (n = 54) of kidney transplants examined for the impact of intraoperative verapamil therapy on DGF.

MATERIALS AND METHODS

A single-center retrospective matched cohort study was performed of 93 Ver (−) kidney transplant recipients compared with 93 Ver (+) kidney transplant recipients, matched by donor age, Kidney Donor Profile Index (KDPI), and DCD status. The study population comprised deceased donor kidney transplants performed at the New York University Langone Health Transplant Institute from January 1, 2018, until July 31, 2020. Exclusion criteria included recipients <18 y of age, pediatric en bloc transplants, and multiorgan transplants. Approval for this study was obtained from the New York University Grossman School of Medicine Institutional Review Board. The study adheres to the Declaration of Helsinki. The clinical and research activities being reported are consistent with the Principles of the Declaration of Istanbul as outlined in the "Declaration of Istanbul on Organ Trafficking and Transplant Tourism."

Deceased donor transplant procedures were conducted in accordance with standard protocols. At our center, 1 surgeon routinely performs intraoperative injection of 5 mg of verapamil immediately after renal reperfusion. The verapamil is injected directly into the external iliac artery after release of the proximal artery clamp. The distal arterial clamp is maintained for a minimum of 1 min to allow for first pass circulation of the verapamil within the transplanted kidney. The operation and perioperative care are otherwise standardized within the surgical practice. Induction immunosuppression was methylprednisolone, mycophenolate mofetil, and either rabbit antithymocyte globulin (cumulative dose 3–6 mg/kg) or basiliximab (20 mg doses intraoperative and on postoperative day 4). Maintenance immunosuppression was prednisone, mycophenolate mofetil, and tacrolimus (target trough 8–10 ng/mL). DGF was defined as the need for hemodialysis within 1 wk of transplantation. Renal function posttransplant was evaluated by the glomerular filtration rate based on serum creatinine using the Chronic Kidney Disease Epidemiology Collaboration creatinine equation.[22]

Recipient, donor, and transplant demographics were compared between the Ver (+) and Ver (−) cohorts for the overall study population and stratified by donor type. Statistical analysis was performed in Stata Version 16.0 (StataCorp, College Station, TX). All statistical tests were 2-sided with statistical significance set at $\alpha = 0.05$. Comparisons of demographic variables between the 2 groups are reported using unpaired 2-tailed t tests for continuous covariates and chi-square tests of independence for categorical variables.

Risk factors for DGF were determined using logistic regression. Multivariate models were adjusted for the following clinically relevant covariates: donor hypertension, CIT, pulsatile preservation, and verapamil administration. Odds ratios (ORs) with 95% confidence intervals and 2-tailed P values were reported.

RESULTS

A total of 281 patients undergoing deceased donor kidney transplant at our institution met inclusion criteria during the study period. Intraoperative verapamil (Ver [+]) was used for 93 patients during their kidney transplant. Matching was performed for donor age, KDPI, and DCD status to identify the Ver (−) matched cohort. There were no statistically significant differences between the Ver (+) and Ver (−) matched cohorts in recipient, donor, or transplant demographics (Table 1). Stratification of the study population by donor type found that DCD Ver (−) recipients were more likely to be hypertensive ($P = 0.003$), obese ($P = 0.01$), and sensitized ($P = 0.02$) as compared to the DCD Ver (+) recipients (Table 2). DCD Ver (−) kidneys were also more likely to have had pulsatile preservation ($P = 0.04$). The 2 DCD cohorts were otherwise similar in the key demographics examined, with both exhibiting >50% rate of DGF (Table 2). To evaluate if the prolonged mean CIT of the study population (mean CIT 28 h) was a potential confounder blocking the impact of verapamil on DGF, the DGF incidence between the Ver (−) and Ver (+) groups was examined after stratification by CIT (Table 3). However,

TABLE 1.

Study population demographics

	Ver (−) (n = 93)	Ver (+) (n = 93)	P
Recipient			
Age (y)	56.5	56.0	0.41
Female (%)	34.4	26.9	0.27
HTN (%)	68.8	64.5	0.53
Diabetes (%)	51.6	39.8	0.11
BMI ≥30 (%)	31.2	31.2	1.00
PRA ≥30 (%)	18.6	11.8	0.15
Donor			
Age (y)	45.0	45.0	0.47
HTN (%)	38.7	36.6	0.76
Diabetes (%)	3.2	6.5	0.11
DCD (%)	29.0	29.0	1.00
DCD WIT (min)	39	41	0.34
KDPI	61	63	0.22
Terminal Cr (mg/dL)	2.5	2.6	0.36
COD anoxia (%)	57.0	50.5	0.38
Hemodialysis (%)	4.3	3.2	0.41
Transplant			
CIT (h)	29	28	0.14
Anastomotic WIT (min)	41	41	0.27
Pulsatile preservation (%)	87.0	80.6	0.17
DGF (%)	58.1	49.5	0.24

Age, WIT, KDPI, Cr, CIT are reported as means; all other parameters are reported as proportions. BMI, body mass index; CIT, cold ischemia time; COD, cause of death; Cr, creatinine; DCD, donation after circulatory death; DGF, delayed graft function; HTN, hypertension; KDPI, Kidney Donor Profile Index; PRA, panel reactive antibody; Ver, verapamil; WIT, warm ischemia time.

TABLE 2.

DCD demographics by verapamil status

	Ver (−) (n = 27)	Ver (+) (n = 27)	P
Recipient			
Age (y)	60.3	62.6	0.11
Female (%)	48.1	29.6	0.16
HTN (%)	**88.9**	**51.9**	**0.003**
Diabetes (%)	62.9	44.4	0.17
BMI ≥30 (%)	**40.7**	**11.1**	**0.01**
PRA ≥30 (%)	**25.9**	**3.7**	**0.02**
Donor			
Age (y)	50.1	50.7	0.41
HTN (%)	37.0	33.3	0.76
Diabetes (%)	3.7	0	0.31
DCD WIT (min)	41	40	0.31
KDPI	71	70	0.41
Terminal Cr (mg/dL)	1.1	1.1	0.50
COD anoxia (%)	55.6	48.1	0.59
Transplant			
CIT (h)	31	29	0.19
Anastomotic WIT (min)	41	40	0.31
Pulsatile preservation (%)	**96.2**	**77.8**	**0.04**
DGF (%)	62.9	55.6	0.58

Age, WIT, KDPI, Cr, CIT are reported as means; all other parameters are reported as proportions. Bold values correspond to $P < 0.05$.
BMI, body mass index; CIT, cold ischemia time; COD, cause of death; Cr, creatinine; DCD, donation after circulatory death; DGF, delayed graft function; HTN, hypertension; KDPI, Kidney Donor Profile Index; min, minutes; PRA, panel reactive antibody; Ver, verapamil; WIT, warm ischemia time.

TABLE 3.

Impact of CIT on DGF

Full study population	Ver (−) (n = 93)	Ver (+) (n = 93)	P
CIT ≤ 12 h	0% (3)	33.3% (3)	0.30
CIT 12 ≤ 24 h	59.3% (27)	37.5% (24)	
CIT >24 h	60.3% (63)	54.5% (66)	

DCD study population	Ver (−) (n = 27)	Ver (+) (n = 27)	P
CIT ≤12 h	0% (0)	0% (0)	0.54
CIT 12 ≤24 h	71.4% (7)	42.9% (7)	
CIT >24 h	60.0% (20)	60.0% (20)	

Reported as percentage with DGF, total number of patients in each group shown in parentheses.
CIT, cold ischemia time; DCD, donation after circulatory death; DGF, delayed graft function; Ver, verapamil.

although there was a trend toward a higher incidence of DGF in the CIT 12 ≤ 24 h Ver (−) patients, it did not achieve statistical significance for either the overall study population ($P = 0.30$) or the DCD subgroup ($P = 0.54$) (Table 3).

The estimated glomerular filtration rate (eGFR) was recorded posttransplant (day 7, 15, 30, 60, 120, 150, 180, and 365). The eGFR (mL/min) was compared between the Ver (−) and Ver (+) cohorts stratified by donor type. There were no significant differences in the eGFR at any time point posttransplant between the Ver (−) and Ver (+) cohorts for either donation after brain death (DBD) (Figure 1A) or DCD (Figure 1B) donors.

Increasing allograft CIT (OR, 1.05; 95% CI, 1.01-1.09, $P = 0.006$) was associated with a significantly increased risk of DGF on univariate analysis (Table 4). After adjusting for donor, recipient, and transplant factors impacting DGF, CIT remained an independent predictor of DGF (OR, 1.06; 95% CI, 1.02-1.10, $P = 0.005$) (Table 4).
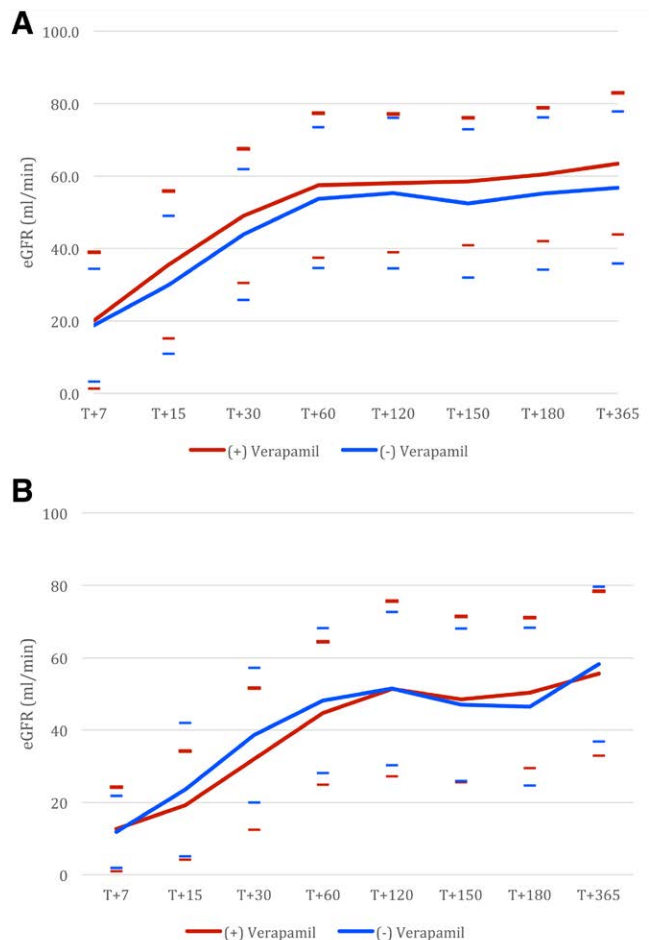

FIGURE 1. Intraoperative verapamil does not impact posttransplant GFR. Data plotted as eGFR (mL/min) versus time (days posttransplant). A, DBD. B, DCD. Data represented as mean ± SD. DBD, donation after brain death; DCD, donation after circulatory death; eGFR, estimated glomerular filtration rate; GFR, glomerular filtration rate.

TABLE 4.

Intraoperative verapamil fails to reduce risk of delayed graft function

	Univariate model			Multivariate model		
Variable	OR	95% CI	P	OR	95% CI	P
Intraoperative verapamil	0.71	0.40-1.26	0.24	0.73	0.40-1.32	0.30
Pulsatile preservation	1.02	0.47-2.23	0.96	0.83	0.37-1.86	0.64
CIT (h)	**1.05**	**1.01-1.09**	**0.006**	**1.06**	**1.02-1.10**	**0.005**
Donor hypertension	0.86	0.47-1.56	0.62	1.75	0.40-1.39	0.36

Bold values correspond to $P < 0.05$.
CI, confidence interval; CIT, cold ischemia time; OR, odds ratio.

DISCUSSION

Calcium homeostasis plays a critical role in the IRI molecular signaling pathways during kidney allograft reperfusion.[7] DCD kidneys have a dual ischemic insult secondary to the procurement WIT that directly impacts the risk for DGF.[4,5] Prior studies found a reduced incidence of DGF with intraoperative verapamil administration in small case series.[16,18] However, kidney transplants in the current era use more expanded criteria donors (eg, older, DCD, longer CIT, elevated creatinine). A larger contemporaneous series comprised mainly living donor (39%) and DBD (58%) transplants failed

to demonstrate a significant benefit with the use intraoperative verapamil.[21] In the latter report, the small DCD Ver (+) subgroup did have a lower incidence of DGF that did not achieve statistical significance because of sample size limitations.[21] Our single-center matched cohort study included a large Ver (+) DCD cohort (n = 27); however, we also did not demonstrate a significant reduction in DGF versus the Ver (–) DCD cohort. In our cohort matched for donor age, KDPI, and DCD status, the only covariate independently associated with an increased risk of DGF on multivariate logistic regression analysis was CIT ($P = 0.005$).

Complex interactions occur between donor variables and transplant parameters that likely impact the risk for DGF. CIT is a well-recognized independent predictor of DGF in DCD allografts.[3,6,23] It is possible that the mean CIT >24 h of our DCD population may preclude the ability to observe a benefit from intraoperative verapamil administration. Although there was a trend in the CIT 12 ≤24 h Ver (+) patients to have reduced incidence of DGF, our sample size remains inadequately powered to definitively address this question. Gupta et al[21] had a much shorter mean CIT in their DCD cohorts; Ver (+) = 7 h and Ver (–) = 10 h. The ability of CIT to override a protective therapy for DGF risk reduction has been demonstrated previously with pulsatile preservation. A majority of DCD allografts in the United States have pulsatile preservation,[3] with the intent to reduce DGF.[24] A national registry analysis found that for DCD kidneys with CIT 6 to 24 h, pulsatile preservation reduced DGF.[3] However, once CIT exceeded 24 h, pulsatile preservation no longer lowered the risk of DGF.[3] Of note, other studies have reported no benefit to pulsatile preservation on the DGF risk for DCD allografts.[25] Clearly, there are many potential confounding variables to consider, including the procurement WIT,[4,5] preservation solution,[26] organ extraction time,[27] and duration of pulsatile preservation.

Of these latter potential confounders, our study did evaluate procurement WIT. A recent trial reported that for procurement WIT >30 min, there was a 5.8-fold increased risk of DGF.[4] Similarly, another study found that procurement WITs from 20 to 40 min had an increased risk of DGF.[5] Our study population had procurement WITs of 36 to 40 min that correlates with the WIT reported as increased risk for DGF.[4,5] Thus, the procurement WIT of our cohort could be another factor that prevented any benefit from intraoperative verapamil.

The strength of this study is the DCD sample size. This report includes the largest DCD cohort to examine intraoperative verapamil administration. Furthermore, unlike the prior studies of intraoperative verapamil that had short mean CIT <10 h,[16,18,21] our study population had a mean CIT >24 h, which is more reflective of the current practice in the United States, with wider geographic allocation of organs leading to longer CIT. The major limitation to this study is the nonrandomization of verapamil utilization.

The deficit of organs for transplantation has led to increased utilization of DCD organs to help patients on the waiting list. DCD kidneys have higher rates of DGF as compared to DBD kidneys. Herein, we report our institutional experience with intraoperative verapamil administration, including a large DCD cohort. In conclusion, intraoperative verapamil fails to reduce the risk of DGF when CIT extends beyond 24 h in DCD allografts. Further data are needed to determine if DGF risk can be mitigated by the use of intraoperative verapamil in DCD kidneys with shorter CIT.

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