Role of Donor Hemodynamic Trajectory in Determining Graft Survival in Liver Transplantation From Donation After Circulatory Death Donors

Daniel J. Firl,[1] Koji Hashimoto,[1] Colin O'Rourke,[2] Teresa Diago-Uso,[1] Masato Fujiki,[1] Federico N. Aucejo,[1] Cristiano Quintini,[1] Dympna M. Kelly,[1] Charles M. Miller,[1] John J. Fung,[1] and Bijan Eghtesad[1]

[1]Cleveland Clinic Lerner College of Medicine, Department of General Surgery, Digestive Disease Institute, and [2]Department of Quantitative Health Sciences, Cleveland Clinic, Cleveland, OH

Donation after circulatory death (DCD) donors show heterogeneous hemodynamic trajectories following withdrawal of life support. Impact of hemodynamics in DCD liver transplant is unclear, and objective measures of graft viability would ease transplant surgeon decision making and inform safe expansion of the donor organ pool. This retrospective study tested whether hemodynamic trajectories were associated with transplant outcomes in DCD liver transplantation (n = 87). Using longitudinal clustering statistical techniques, we phenotyped DCD donors based on hemodynamic trajectory for both mean arterial pressure (MAP) and peripheral oxygen saturation (SpO_2) following withdrawal of life support. Donors were categorized into 3 clusters: those who gradually decline after withdrawal of life support (cluster 1), those who maintain stable hemodynamics followed by rapid decline (cluster 2), and those who decline rapidly (cluster 3). Clustering outputs were used to compare characteristics and transplant outcomes. Cox proportional hazards modeling revealed hepatocellular carcinoma (hazard ratio [HR] = 2.53; $P = 0.047$), cold ischemia time (HR = 1.50 per hour; $P = 0.027$), and MAP cluster 1 were associated with increased risk of graft loss (HR = 3.13; $P = 0.021$), but not SpO_2 cluster ($P = 0.172$) or donor warm ischemia time (DWIT; $P = 0.154$). Despite longer DWIT, MAP and SpO_2 clusters 2 showed similar graft survival to MAP and SpO_2 clusters 3, respectively. In conclusion, despite heterogeneity in hemodynamic trajectories, DCD donors can be categorized into 3 clinically meaningful subgroups that help predict graft prognosis. Further studies should confirm the utility of liver grafts from cluster 2.

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The waiting list for liver transplantation continues to exceed organ availability.[1-3] With this persistent donor shortage, donation after circulatory death (DCD) donors have become an important source of lifesaving organs. However, the use of livers from DCD donors is generally characterized by increased rates of biliary complications and inferior survival compared with donation after brain death donors.[1-4] Although previous studies have demonstrated various risk factors for poor outcomes in DCD liver transplantation, the underlying mechanisms remain unclear.

Abbreviations: ALT, alanine aminotransferase; AST, aspartate aminotransferase; BMI, body mass index; CI, confidence interval; CIT, cold ischemia time; CVA, cerebrovascular accident; DCD, donation after circulatory death; DRI, donor risk index; DWIT, donor warm ischemia time; HCC, hepatocellular carcinoma; HCV, hepatitis C virus; HR, hazard ratio; IQR, interquartile range; ITBS, ischemic-type biliary stricture; MAP, mean arterial pressure; MELD, Model for End-Stage Liver Disease; RWIT, recipient warm ischemia time; SBP10, systolic blood pressure regression coefficient over first 10 minutes; SD, standard deviation; SpO_2, peripheral oxygen saturation; tPA, tissue plasminogen activator.

In DCD liver transplantation, all donors experience an unavoidable warm ischemic insult during the process of organ recovery after withdrawal of life support. An extended withdrawal period is uniformly viewed as detrimental to the viability of liver grafts due to poor perfusion and oxygenation, causing serious tissue damage that contributes to the apparent inferiority of DCD liver transplantation. Unsurprisingly, previous studies have shown that prolonged donor warm

ischemia time (DWIT; from extubation to cross clamp) is a strong indicator of increased biliary ischemia and poor graft survival.[5,6] However, several studies have failed to show significant impact of DWIT in transplant outcomes.[7,8] Therefore, there is no consensus on the magnitude of the effect of DWIT on graft viability.[9-12]

From withdrawal of life support to the determination of cardiopulmonary death, the hemodynamics of the donor can proceed via a variety of trajectories. According to our clinical experience, donors can be categorized into 3 major groups:

1. Those who gradually decline with a prolonged agonal phase.
2. Those who maintain stable hemodynamics for the initial phase after withdrawal and then decline rapidly to cardiac death.
3. Those who decline rapidly after withdrawal of life support.

However, characteristics of these phenotypes are unknown and impacts on transplant outcomes have never been studied. Accordingly, this study was conducted to evaluate whether the donor's hemodynamic trajectory during DWIT determines graft outcomes in DCD liver transplantation.

Patients and Methods

STUDY POPULATION

From 2007 to 2015, 98 recipients received DCD liver grafts at Cleveland Clinic. Donor and recipient data were retrieved from our prospectively collected transplant database. After excluding 11 recipients with incomplete data, 87 recipients were enrolled in this retrospective study. Recipient follow-up ranged from 20 to 2817 days (median, 1163 days). This study was approved by the Cleveland Clinic institutional review board.

The characteristics of donors used in this series are summarized in Supporting Table 1. Donors had a median age of 44 years (interquartile range [IQR], 28-51 years) and 44% were female. Donors' length of hospitalization before procurement was a median of 4 days (IQR, 3-6 days). The majority of donors (53%) suffered anoxic insult as the primary mechanism of brain injury. Most donors (59%) were receiving vasopressor support to maintain hemodynamic stability prior to withdrawal.

Recipient demographics are summarized in Supporting Table 2. They were predominantly white (90%) and male (76%) with a median age of 58 years (IQR, 54-64 years). Listing Model for End-Stage Liver Disease (MELD) score in this population was 22 (IQR, 18-22) with median laboratory MELD score of 16 (IQR, 10-20). Among the recipients, 35% had concomitant hepatocellular carcinoma (HCC) and 40% had hepatitis C virus (HCV)–positive serology. In the HCC cohort, 74% were within Milan criteria. Tumor burden was a median of 2 lesions (IQR, 1-3) with the largest lesion diameter a median of 2.5 cm (IQR, 2.2-3.8 cm) and a summed lesion diameter of 4.5 cm (IQR, 2.9-5.1 cm). The median time on the liver transplant waiting list was 80 days (IQR, 32-191 days).

DCD ORGAN RECOVERY

Organ recovery from DCD donors was described previously.[13,14] In brief, after the withdrawal of life support, vital signs (including blood pressure, pulse, and oxygen saturation) were recorded every minute and reported to the recovery surgeons every 5 minutes. DWIT was defined as time from withdrawal of life-sustaining measures to cross-clamp. For analysis purposes, hypoxia time was defined as minutes with peripheral oxygen saturation (SpO_2) < 70%, and hypoperfusion time was defined as minutes with mean arterial pressure (MAP) < 60 mm Hg as described in previous reports.[12]

Following cessation of circulation, declaration of death was made by an independent physician, after 2-5 minutes of mandatory observation to preclude autoresuscitation. Heparin was administered systemically prior to withdrawal of life support, if local policies permitted it. Otherwise, 30,000 units of heparin were mixed in the initial bag of cold preservation solution. Histidine tryptophan ketoglutarate solution (Custodiol, Essential Pharmaceuticals, PA) was our choice.[15] At incision,

Address reprint requests to Koji Hashimoto, M.D., Ph.D., Department of General Surgery, Digestive Disease Institute, Cleveland Clinic, 9500 Euclid Avenue, A100, Cleveland, OH 44195. Telephone: 216-445-0753; FAX: 216-444-9375; E-mail: hashimk@ccf.org

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TABLE 1. Transplant Characteristics

	Values	Min-Max
DRI	2.33 (2.03-2.63)	1.42-3.37
Allocation type		
Local	38% (33)	
Regional	45% (39)	
National	17% (15)	
Extubation to cessation of circulation, minutes	13 (10-17)	3-31
Cessation of circulation to cross-clamp, minutes	9 (8-10)	4-22
Extubation to cross-clamp, DWIT, minutes	23 (19-27)	13-38
CIT, minutes	374 (343-431)	266-588
RWIT, minutes	42 (36-49)	23-77
Intensive care unit stay, days	2 (1-4)	1-42
Postoperative length of stay, days	10 (7-15)	4-49
Peak liver function tests		
Total bilirubin, mg/dL	3.1 (1.8-5.9)	0.5-22.8
AST, IU/L	642 (413-1046)	159-3166
ALT, IU/L	468 (282-800)	69-2160

NOTE: Data are given as median (IQR) and percents and frequencies.

procurement proceeded via the super-rapid technique.[14,16] Tissue plasminogen activator (tPA) was injected into the hepatic artery on the back table, except for patients considered to have exceptional risk of post-reperfusion bleeding.[13] Cold ischemia time (CIT) was defined as the time from cross-clamp in the donor to graft reperfusion. Recipient warm ischemia time (RWIT) was defined as the time when a liver graft was taken out of cold preservation solution to the time when the graft was reperfused.

The details of organ recovery and transplantations are summarized in Table 1. Median time from withdrawal of life support to cessation of circulation was 13 minutes (IQR, 10-17 minutes). Time from cessation of circulation to incision and then cross-clamp was a median of 9 minutes (IQR, 8-10 minutes). Median DWIT was 23 minutes (IQR, 19-27 minutes). The median CIT was 374 minutes (IQR, 343-431 minutes) and RWIT was 42 minutes (IQR, 36-49 minutes).

GENERATION OF DONOR CLUSTERS AND STATISTICAL ANALYSIS

Analysis of the minutely collected hemodynamic data was done using longitudinal clustering techniques.[17,18] Data were transformed wide with 1 patient per row and each hemodynamic parameter having 1 variable per minute. Then clustering was performed with minute dimensions using the kmedians method in STATA with 10^6 iterations and random initialization. We chose $k = 3$ based on our clinical experience. Kmedians was chosen to minimize the effect of outliers because it is more robust in the L1-norm space than kmeans in L2.[19] Using these outputs, we generated clinical definitions (Fig. 1) for the sole purpose of reliably reproducing clusters on other data sets. Generalized linear models with random effects evaluated differences in hemodynamic course by cluster. Thus, we developed an unbiased, hybrid approach using both unsupervised techniques and clinical experience to phenotype DCD donors for the first time.

Graft loss was defined as patient death or retransplantation. Cox proportional hazards models (P value cutoffs < 0.20 for univariate, < 0.05 for multivariate) and the Kaplan-Meier method were used to assess association between graft survival and hemodynamic cluster scores and conventionally reported hemodynamic predictors. The Gehan-Breslow-Wilcoxon test was used instead of the log-rank for survival comparison because increased sensitivity to early graft loss was

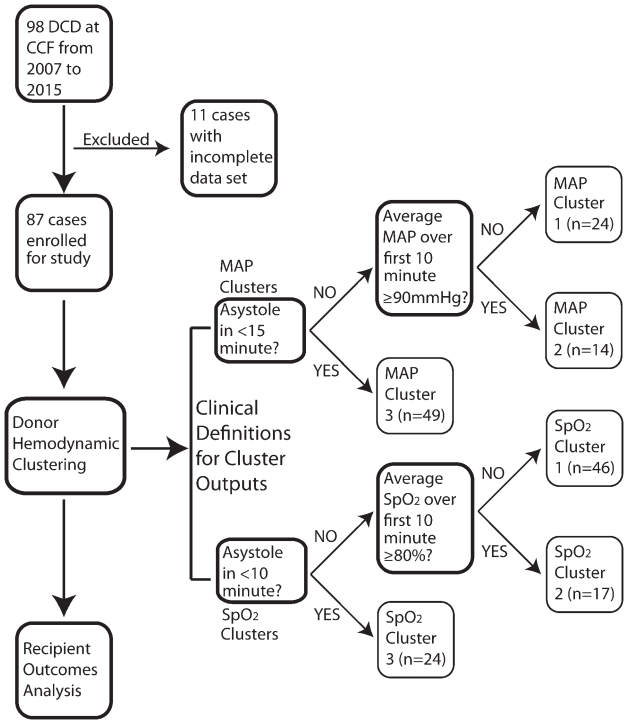

FIG. 1. Overview of study design and cluster approach. Eighty-seven DCD liver transplants were enrolled following inclusion/exclusion criteria assessment from 2007 to 2015. Hemodynamic data were used to cluster donors based on various phenotypes seen in clinical experience with DCD liver transplantation.

desired. The systolic blood pressure regression coefficient over first 10 minutes (SBP10) is the slope of systolic blood pressure over the first 10 minutes following withdrawal and was previously reported to be associated with graft survival.[9,20] Early allograft dysfunction was defined as aspartate aminotransferase (AST) or alanine aminotransferase (ALT) $\geq$ 2000 IU/L in the first 7 postoperative days, INR $\geq$ 1.6 at day 7, or bilirubin $\geq$ 10 mg/dL at day 7.[21] Categorical variables were summarized by counts and percentages and continuous variables by median (IQR). Categorical testing was done using Fisher's exact test. Continuous variables were tested using the 2-sample Wilcoxon rank sum (Mann-Whitney U) test or Kruskal-Wallis test (with posthoc Dunn's test) depending on the number of groups. All testing was 2-sided and used a 5% level of significance. All analyses were done using STATA 13 (STATA Corp., College Station, TX).

Results

CHARACTERIZING DCD LIVER TRANSPLANT BY DONOR MAP TRAJECTORY

Figure 2A demonstrates the heterogeneity of MAP trajectories in 87 donors following withdrawal of life support. Because donor hemodynamics were so heterogeneous, a clustering strategy was employed to maximize differences between groups (Fig. 1). Figure 2B shows the mean course for each MAP cluster. Donors were assigned as one of the following: donors with gradually declining MAP (MAP cluster 1, n = 24; Fig. 2C), donors whose MAP stayed nearly physiologic after withdrawal of life support and then rapidly declined (MAP cluster 2, n = 14; Fig. 2D), and donors who declined rapidly to asystole within 15 minutes after withdrawal (MAP cluster 3, n = 49; Fig. 2E). MAP clustering generated 3 groups with statistically and clinically meaningful differences ($P < 0.001$; Fig. 2B). Interestingly, the donors in MAP cluster 2 demonstrated a similar MAP trajectory to MAP cluster 3 once their MAP started declining (Fig. 2B).

In addition to characterizing the donors and their hemodynamics, clustering outputs were used to perform survival analysis. As shown in Fig. 2F, the 1-, 3-, and 5-year graft survival in MAP cluster 1 (73.5%, 62.0%, and 62.0%) tended to be worse than in cluster 2 (100.0%, 90.9%, and 90.9%) or cluster 3 (91.3%, 86.4%, and 71.7%), although not statistically significant ($P = 0.090$).

To further understand differences between donor clusters, we performed posthoc analysis of donor characteristics (Table 2). Donors in MAP cluster 2 were younger than 1 and 3 (29 versus 47 and 45 years, respectively; $P = 0.029$). However, consistent with our previous study,[14] donor age was not associated with graft survival (hazard ratio [HR] = 1.00 per year; $P = 0.937$). There was no difference in distributions of sex, race, donor body mass index (BMI), or causes of brain injury. There was a trend toward decreased vasopressor support in MAP cluster 2 compared with 1 and 3 (36% versus 75% and 57%, respectively; $P = 0.064$). Transplants across MAP cluster had similar CIT, donor risk index (DRI), RWIT, recipient age, HCC Milan status, and HCV serologies. There was a trend toward increased laboratory MELD scores in recipients who received MAP cluster 2 donors compared with 1 and 3 (19 versus 17 and 14, respectively; $P = 0.069$).

Surgical complications were examined by MAP cluster (Table 3), but no comparisons were statistically different. However, primary nonfunction and early allograft dysfunction tended to occur more often in MAP cluster 1 (8% and 25%) compared with 2 (0% and 7%) and 3 (2% and 16%), respectively ($P = 0.249$ and 0.424).

CHARACTERIZING DCD LIVER TRANSPLANT BY DONOR OXYGEN SATURATION TRAJECTORY

SpO_2 trajectories of the 87 donors are shown in Fig. 3A. Similarly to the MAP analysis, we clustered SpO_2 into 3 groups with differences in SpO_2 (Fig. 3B): a population of donors who had longer withdrawal time with low peripheral oxygenation (SpO_2 cluster 1, n = 46; Fig. 3C), donors whose SpO_2 stayed greater than 80% on average for the first 10 minutes (SpO_2 cluster 2, n = 17; Fig. 3D), and donors whose SpO_2 declined rapidly and who became asystolic within 10 minutes (SpO_2 cluster 3, n = 24; Fig. 3E).

As shown in Fig. 3F, although not statistically significant ($P = 0.117$), the 1-, 3-, and 5-year graft survival for SpO_2 cluster 1 (81.5%, 66.8%, and 66.8%) tended to be worse than cluster 2 (93.3%, 84.9%, and 74.2%) and cluster 3 (95.0%, 90.0%, and 81.0%).

Table 4 shows demographic comparisons by cluster. Contrary to the MAP analysis, donor age was similar between the SpO_2 clusters ($P = 0.547$). Donors between SpO_2 clusters also had similar distributions of

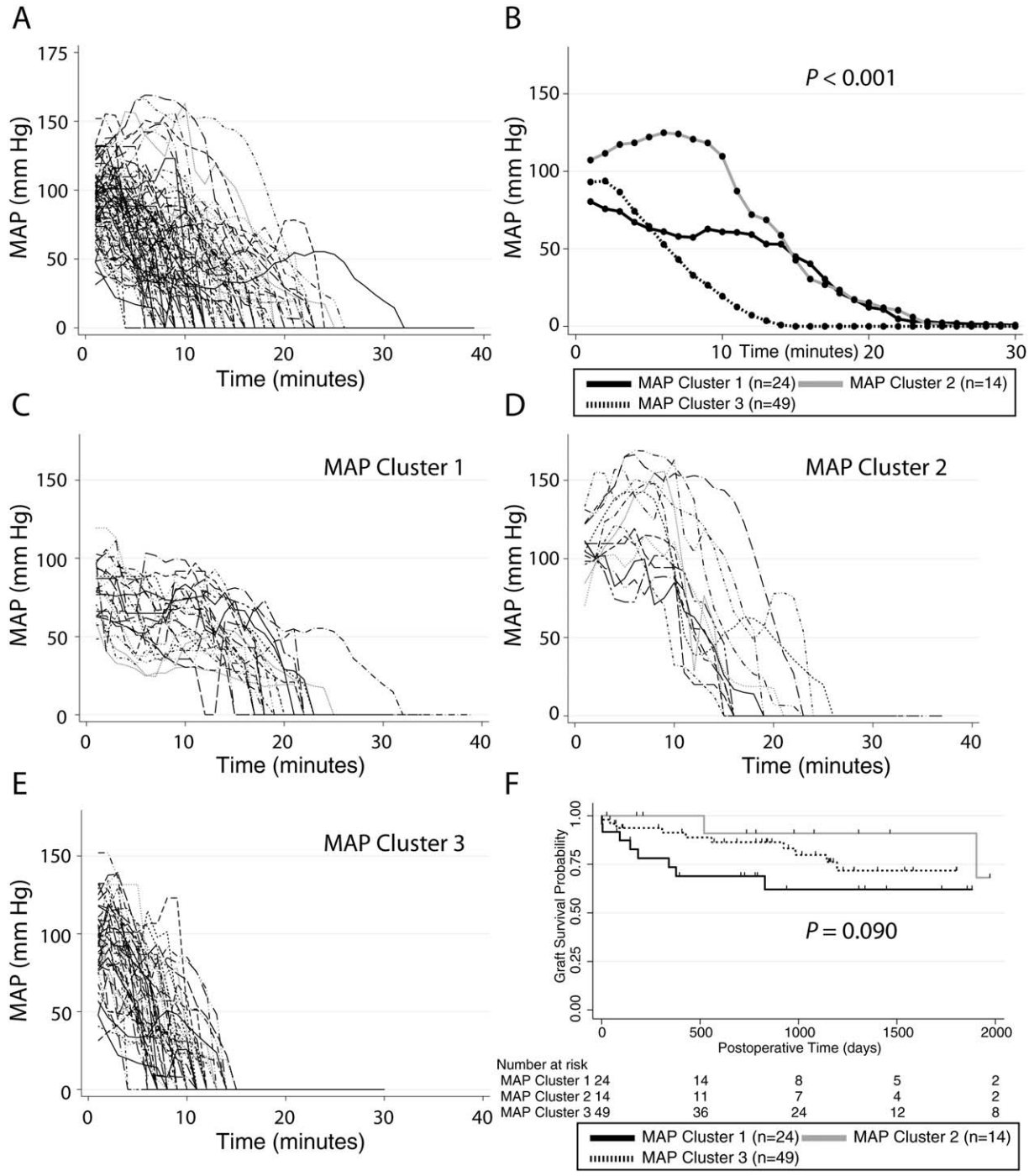

FIG. 2. Hemodynamic clustering by donor MAP trajectory. (A) Serial changes of minutely recorded MAP in 87 DCD donors from the time of withdrawal of life support to asystole and (B) mean values of MAP in 3 different clusters as determined by the criteria shown in Fig. 1 ($P < 0.001$). Individual donor MAP trajectories shown by (C) cluster 1 (n = 24), (D) cluster 2 (n = 14), and (E) cluster 3 (n = 49). (F) Kaplan-Meier estimates of graft survival by MAP cluster ($P = 0.090$).

sex, race, hospital stay prior to withdrawal, mechanism of brain injury, and rates of pharmacologic hemodynamic support. Transplants across SpO_2 clusters had similar CIT, RWIT, recipient age, HCC Milan status, and HCV serologies. Donors from SpO_2 cluster 1 had a statistically greater DRI than 2 and 3 (2.4 versus 2.1 and 2.2, respectively; $P = 0.039$). There was a trend toward increased laboratory MELD score in recipients of SpO_2

TABLE 2. Characterizing DCD Liver Transplant by MAP Cluster

	MAP Cluster 1 (n = 24)	MAP Cluster 2 (n = 14)	MAP Cluster 3 (n = 49)	*P* Value
Donor age, years	47 (33-52)	29 (20-44)†	45 (31-51)	0.029*
Donor sex, female	54% (13)	57% (8)	35% (17)	0.156‡
Donor race				0.513‡
White	88% (21)	100% (14)	94% (46)	
Black	8% (2)	0	6% (3)	
Other	4% (1)	0	0	
Donor BMI, kg/m^2	28.2 (22.4-33.8)	24.4 (23.5-30.5)	27.5 (23.8-32.1)	0.760*
Donor admission length, days	4 (3-7)	4 (3-6)	4 (3-6)	0.952*
Mechanism of injury				0.749‡
Anoxia	58% (14)	57% (8)	49% (24)	
Head trauma	17% (4)	14% (2)	29% (14)	
CVA/stroke	21% (5)	21% (3)	12% (6)	
Other	4% (1)	7% (1)	10% (5)	
Vasopressor use, yes	75% (18)	36% (5)	57% (28)	0.064‡
Number of pressors	1.0 (0.5-2.0)	0 (0-1.0)†	1.0 (0.0-2.0)	0.048*
Hypoxia time, $SpO_2 < 70\%$, minutes	22 (17-25)	22 (18-23)	16 (13-19)†	<0.001*
Hypoperfusion time, MAP < 60 mm Hg, minutes	17 (12-25)	15 (13-18)	13 (11-18)†	0.092*
SBP10, mm Hg/minute	−3.05 (–5.7 to –0.2)†	1.1 (–1.2-4.8)†	−13.9 (–19.4 to –7.0)†	<0.001*
DWIT, minutes	27 (24-30)	29 (25-32)	19 (16-22)†	<0.001*
CIT, minutes	358 (332-404)	372 (355-424)	376 (341-439)	0.420*
DRI	2.4 (2.1-2.7)	2.1 (2.0-2.3)	2.3 (2.0-2.6)	0.128*
RWIT, minutes	45 (38-50)	39 (35-47)	42 (36-47)	0.383*
Laboratory MELD	17 (13-21)	19 (12-25)	14 (9-18)	0.069*
tPA use, yes	58% (14)	50% (7)	59% (29)	0.870*
Early allograft dysfunction, yes	25% (6)	7% (1)	16% (8)	0.424‡
Recipient age, years	57 (55-62)	57 (50-67)	59 (54-64)	0.940*
Recipient HCV, positive serologies	29% (7)	50% (7)	43% (21)	0.409‡
HCC Milan criteria, within	67% (4/6)	83% (5/6)	67% (12/18)	>0.999‡
Follow-up, days	1193 (777-1733)	1192 (736-1925)	1159 (664-2072)	0.997*

Note: Data are given as median (IQR) and percents and frequencies.
*Denotes Kruskal-Wallis test with posthoc Dunn's test for multiple comparisons.
†Difference by multiple comparisons at the 0.05 level.
‡Fisher's exact test for count data.

cluster 2 compared with clusters 1 and 3 (17 versus 16 and 14, respectively; $P = 0.092$). In terms of surgical complications examined by SpO_2, similarly to the MAP cluster analysis, no comparisons were statistically significant although the relatively increased rate of primary nonfunction in cluster 1 persisted (Table 5).

GRAFT SURVIVAL BETWEEN THE SLOW DECLINER AND THE RAPID DECLINER

Hemodynamic trajectories for both MAP and SpO_2 cluster 3 were similar to those of cluster 2 once they

TABLE 3. Surgical Complications by MAP Cluster

Surgical Complications	MAP Cluster 1 (n = 24)	MAP Cluster 2 (n = 14)	MAP Cluster 3 (n = 49)	*P* Value‡
Primary nonfunction	8% (2)	0	2% (1)	0.249
Early allograft dysfunction	25% (6)	7% (1)	16% (8)	0.424
Exploratory laparotomy	13% (3)	0	10% (5)	0.528
Biliary anastomotic stricture	25% (6)	7% (1)	22% (11)	0.414
ITBS	4% (1)	7% (1)	4% (2)	0.802
Bile leak	0	0	2% (1)	>0.999
Hepatic artery thrombosis	0	0	2% (1)	>0.999
Portal vein thrombosis	0	7% (1)	2% (1)	0.371

Note: Data are given as percents and frequencies.
‡Denotes Fisher's exact test for count data.

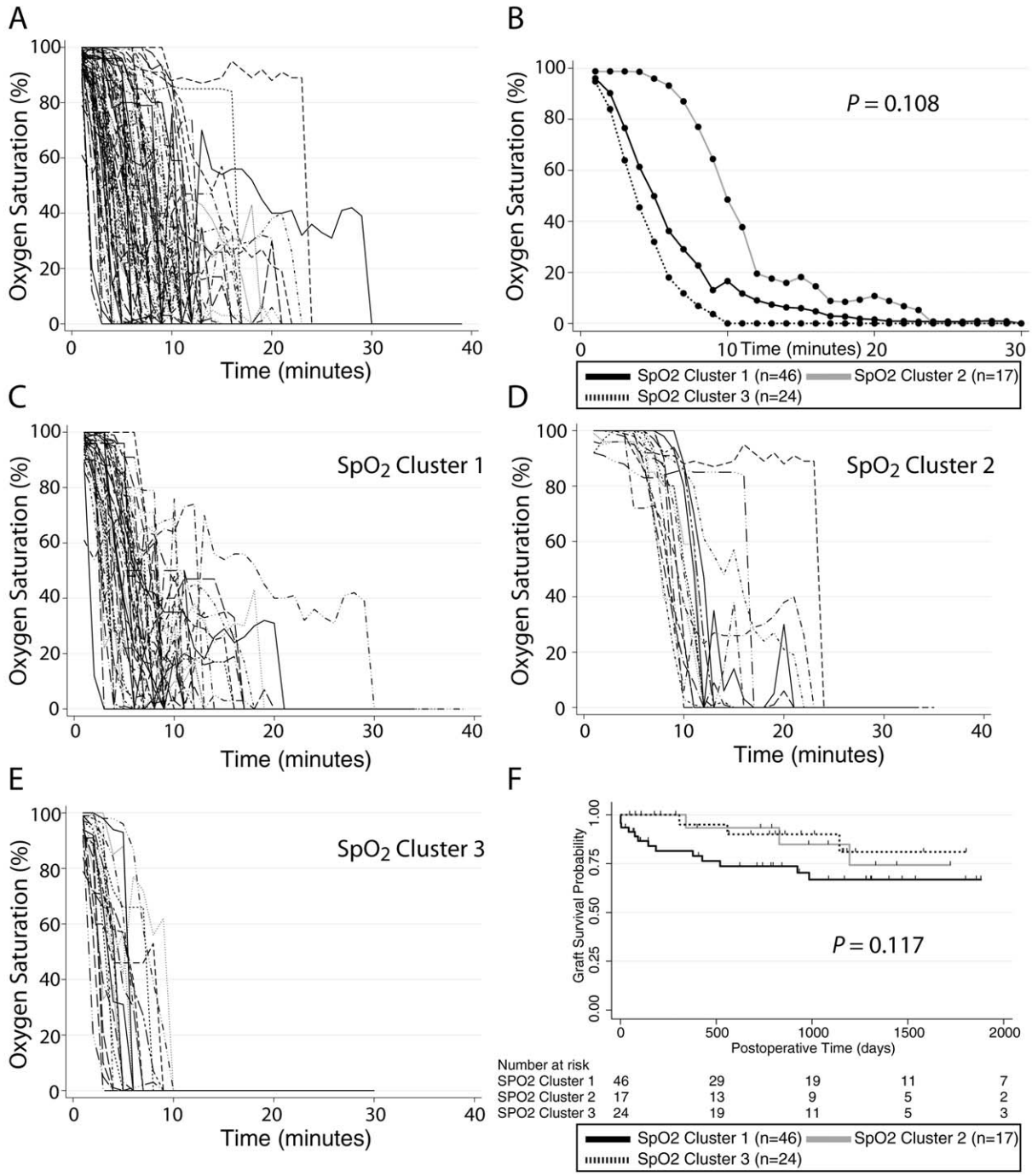

FIG. 3. Hemodynamic clustering by donor SpO_2 trajectory. Serial changes of minutely recorded SpO_2 in 87 DCD donors (A) from the time of withdrawal of life support to asystole, (B) mean values of SpO_2 in 3 different clusters as determined by the criteria shown in Fig. 1 ($P = 0.108$). Individual donor SpO_2 trajectories shown by (C) cluster 1 (n = 46), (D) cluster 2 (n = 17), and (E) cluster 3 (n = 24). (F) Kaplan-Meier estimates of graft survival by SpO_2 cluster ($P = 0.117$).

entered the agonal phase (Figs. 2B and 3B). Thus, we considered donors in cluster 2 as delayed rapid decliners and donors in cluster 3 as early rapid decliners. Although delayed rapid decliners had longer DWIT, they had nearly physiologic hemodynamics during the initial phase of DWIT after withdrawal of

TABLE 4. Characterizing DCD Liver Transplant by SpO_2 Cluster

	SpO_2 Cluster 1 (n = 46)	SpO_2 Cluster 2 (n = 17)	SpO_2 Cluster 3 (n = 24)	*P* Value
Donor age, years	45 (30-52)	41 (26-48)	44 (28-51)	0.547*
Donor sex, female	52% (24)	41% (7)	29% (7)	0.201†
Donor race				>0.999†
White	91% (42)	94% (16)	96% (23)	
Black	7% (3)	6% (1)	4% (1)	
Other	2% (1)	0	0	
Donor BMI, kg/m^2	28.2 (22.9-32.2)	24.3 (23.7-31.4)	27.2 (23.4-32.0)	0.792*
Donor admission length, days	4 (3-7)	4 (3-4)	5 (3-6)	0.280*
Mechanism of injury				0.738†
Anoxia	50% (23)	59% (10)	54% (13)	
Head trauma	20% (9)	24% (4)	29% (7)	
CVA/stroke	20% (9)	17% (3)	8% (2)	
Other	10% (5)	0	8% (2)	
Vasopressor use, yes	57% (26)	65% (11)	58% (14)	0.880†
Number of pressors	1.0 (0-2.0)	1.0 (0-1.0)‡	1.0 (0.0-2.0)	0.980*
Hypoxia time, $SpO_2 < 70\%$, minutes	21 (18-23)‡	16 (13-22)‡	13 (12-16)‡	<0.001*
Hypoperfusion time, MAP < 60 mm Hg, minutes	16 (13-23)‡	15 (11-17)	12 (11-14)	0.002*
SBP10, mm Hg/minute	−5.1 (–9.5 to –1.4)‡	0.0 (–2.1 to 1.7)‡	−19.4 (–21.7 to –14.0)‡	<0.001*
DWIT, minutes	24 (22-28)	26 (23-31)	16 (14-19)‡	<0.001*
CIT, minutes	379 (339-432)	374 (350-421)	367 (339-413)	0.673*
DRI	2.4 (2.1-2.7)‡	2.1 (2.0-2.4)	2.2 (2.0-2.4)	0.039*
RWIT, minutes	41 (36-49)	44 (34-46)	44 (36-54)	0.837*
Laboratory MELD	16 (12-20)	17 (11-24)	14 (9-18)	0.092*
tPA use, yes	61% (28)	47% (8)	58% (14)	0.701*
Early allograft dysfunction, yes	20% (9)	18% (3)	13% (3)	0.809†
Recipient age, years	57 (53-64)	56 (50-60)	61 (55-65)	0.274*
Recipient HCV, positive serologies, %	37% (17)	41% (7)	46% (11)	0.770†
HCC Milan criteria, within	69% (11/16)	75% (3/4)	70% (7/10)	>0.999†
Follow-up, days	1277 (707-1876)	1222 (784-1925)	978 (673-1688)	0.604*

Note: Data are given as median (IQR) and percents and frequencies.
*Denotes Kruskal-Wallis test with posthoc Dunn's test for multiple comparisons.
†Denotes Fisher's exact test for count data.
‡Denotes difference by multiple comparisons at the 0.05 level.

life support, which can only minimally affect graft viability. Thus, we hypothesized that clusters 2 and 3 as rapid decliners should have better graft survival than cluster 1 as slow decliners. For the MAP comparison, the 1-, 3-, and 5-year graft survival of the slow decliner (73.5%, 62.0%, and 62.0%) was significantly worse than that of the rapid decliner (93.2%, 82.2%, and 75.8%; $P = 0.038$; Fig. 4A). For the SpO_2 comparison, the 1-, 3-, and 5-year graft survival of the slow decliner (81.5%, 66.8%, and 66.8%) was significantly worse than the rapid decliner (94.3%, 87.7%, and 76.9%; $P = 0.039$; Fig. 4B).

We analyzed the correlation between donor phenotypes as clustered by MAP and SpO_2 (Table 6) and compared graft survival rates at 1 year. Confirming previous analysis, the greatest survival was found from

TABLE 5. Surgical Complications by SpO_2 Cluster

Complications	SpO_2 C1 (n = 46)	SpO_2 C2 (n = 17)	SpO_2 C3 (n = 24)	*P* Value*
Primary nonfunction	7% (3)	0	0	0.423
Early allograft dysfunction	20% (9)	18% (3)	13% (3)	0.809
Exploratory laparotomy	13% (6)	0	8% (2)	0.287
Biliary anastomotic stricture	17% (8)	24% (4)	25% (6)	0.725
ITBS	7% (3)	6% (1)	0	0.518
Bile leak	0	0	4% (1)	0.471
Hepatic artery thrombosis	0	0	4% (1)	0.471
Portal vein thrombosis	4% (2)	0	0	0.705

Note: Numbers after percents are frequencies.
*Denotes Fisher's exact test for count data.

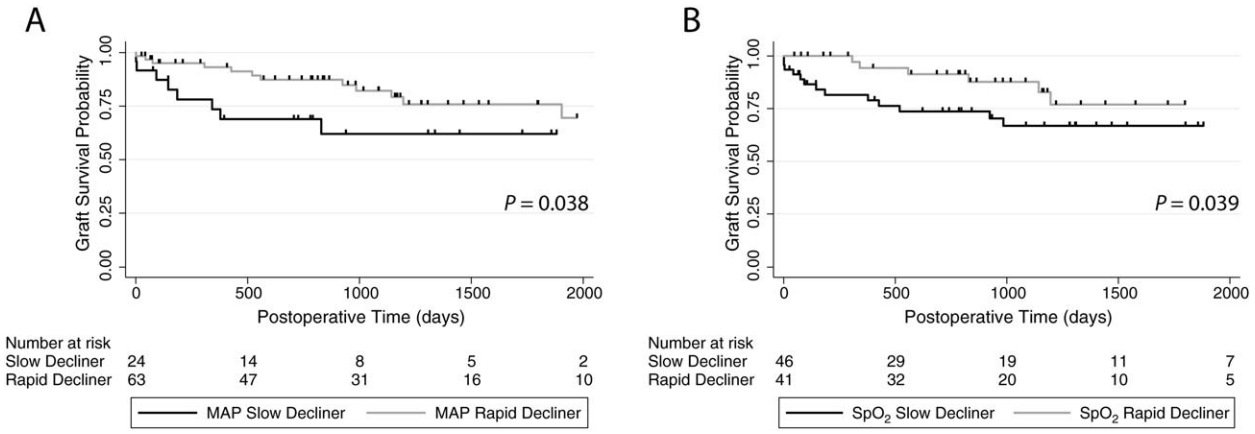

FIG. 4. Graft survival in the slow decliner and the rapid decliner. Kaplan-Meier estimates of graft survival in (A) the MAP slow decliner and rapid decliner ($P = 0.038$) and (B) the SpO_2 slow decliner and rapid decliner ($P = 0.039$).

donors exhibiting both MAP cluster 2 and SpO_2 cluster 2 (100.0%) and was comparable to MAP cluster 3 and SpO_2 cluster 3 (95.0%). Graft survival in these subgroups was significantly superior to the overlap of MAP cluster 1 and SpO_2 cluster 1 (68.3%; $P = 0.046$).

Additionally, causes of graft loss are described between the slow and rapid (early and late) decliner phenotypes in Table 7. Although differences were not statistically significant, there were trends toward decreased primary nonfunction and HCC recurrence in the rapid MAP decliner compared with slow decliner. For SpO_2 analysis, there was a trend toward decreased primary nonfunction in the rapid decliner compared with slow decliner, but there was no statistical significance. Ischemic-type biliary stricture (ITBS) and HCC recurrence occurred at similar rates between SpO_2 groups.

IMPACT OF DONOR HEMODYNAMIC PARAMETERS IN DETERMINING GRAFT SURVIVAL

Traditional intervals describing the donor course were assessed for association with graft survival in a univariate fashion (Table 8). The duration of DWIT was not significantly associated with graft survival (HR = 1.04; IQR, 0.96-1.12, per minute; $P = 0.308$). Similarly, there were no significant associations with graft survival in the time period from extubation to cessation of circulation (HR = 1.06; IQR, 0.98-1.16, per minute; $P = 0.158$) or the time from cessation of circulation to cross-clamp (HR = 0.96; IQR, 0.77-1.12, per minute; $P = 0.629$). The agonal phase defined by MAP < 60 mm Hg (HR = 1.00; IQR, 0.93-1.08, per minute; $P = 0.895$) or $SpO_2 < 70\%$ (HR = 1.05; IQR, 0.97-1.14, per minute; $P = 0.220$) was not significantly associated with graft loss. SBP10, a previously published indicator of the slope of the systolic blood pressure of the donor for the first 10 minutes following withdrawal of support,[21] was not associated with graft loss in our study (HR = 1.00; IQR, 0.95-1.06, per mm Hg/minute; $P = 0.911$).

Finally, a multivariate Cox proportional hazards model for graft failure was constructed (Table 9). The multivariate model showed that HCC (HR = 2.53; IQR, 1.01-6.32; $P = 0.047$) and CIT (HR = 1.50, per hour; IQR, 1.05-2.16; $P = 0.027$) were associated with

TABLE 6. Hemodynamic Cluster Associations and 1-Year Graft Survival

	SpO_2 Cluster 1	SpO_2 Cluster 2	SpO_2 Cluster 3	Total (n = 87)
MAP cluster 1	68.3% (n = 17)	85.7% (n = 7)	—*	68.9% (n = 24)
MAP cluster 2	80.0% (n = 6)	100.0% (n = 8)	—*	90.9% (n = 14)
MAP cluster 3	86.7% (n = 23)	50.0% (n = 2)	95.0% (n = 24)	91.3% (n = 49)
Total (n = 87)	81.5% (n = 46)	93.3% (n = 17)	95.0% (n = 24)	87.6% (n = 87)

Note: Graft survival percentages are actuarial graft survival at 1 year after transplant.
*Structural zeroes based on definitions in Fig. 1.

TABLE 7. Causes of Graft Loss Between Slow and Rapid Decliners

	MAP Cluster 1 (n = 24)	MAP Clusters 2/3 (n = 63)	*P* Value‡	SpO_2 Cluster 1 (n = 46)	SpO_2 Clusters 2/3 (n = 41)	*P* Value*
Graft loss cause:						
Total	33% (8)	17% (11)		28% (13)	15% (6)	
Primary nonfunction	8% (2)	2% (1)	0.183	7% (3)	0% (0)	0.244
ITBS	(0)	5% (3)	>0.999	4% (2)	2% (1)	0.619
HCC recurrence†	33% (2)	8% (2)	0.169	13% (2)	14% (2)	>0.999
Other (viral recurrence, cardiopulmonary, sepsis, etc.)	17% (4)	8% (5)	0.253	13% (6)	7% (3)	0.491

NOTE: Data are given as percents and frequency.
*Denotes Fisher's exact test for count data.
†HCC recurrence rate defined as number of recurrence leading to patient death/number of HCC recipients.

TABLE 8. Comparison of Hemodynamic Predictors

	Median	Mean ± SD	Range	HR (95% CI)	*P* Value
Extubation to cross-clamp, minutes	23.0	22.9 ± 5.9	13.0-38.0	1.04 (0.96-1.12)	0.308
Withdrawal to cessation of circulation, minutes	13.0	13.5 ± 5.5	3.0-31.0	1.06 (0.98-1.16)	0.158
Cessation of circulation to cross-clamp, minutes	9.0	9.3 ± 2.6	4.0-22.0	0.96 (0.77-1.12)	0.629
Hypoperfusion, MAP < 60 mm Hg, minutes	14.0	15.5 ± 5.8	5.0-33.0	1.00 (0.93-1.08)	0.895
Hypoxia, SpO_2 < 70%, minutes	18.0	18.2 ± 5.6	7.0-34.0	1.05 (0.97-1.14)	0.220
SBP10, mm Hg/minute	−6.2	−8.3 ± 9.1	−29.9 to 14.6	1.00 (0.95-1.06)	0.911

NOTE: *P* values and HR from univariate Cox proportional hazard models for graft loss.

graft failure. The MAP slow decliner was associated with a 3.13-fold increased risk of graft loss (HR = 3.13; IQR, 1.19-8.24; $P = 0.021$). However, the SpO_2 slow decliner was not significantly associated with graft loss in the multivariate model (HR = 1.64; IQR, 0.64-4.19; $P = 0.301$).

Discussion

The routine use of DCD grafts could provide more timely access to lifesaving liver transplantation. However, well-documented increased risks of graft failure and biliary complications preclude widespread acceptance of DCD grafts.[10,14,22,23] One of the major challenges in DCD liver transplantation is the lack of objective values predicting graft prognosis. If we can develop measures to accurately predict a priori graft viability, then transplant surgeon decision making would be easier and DCD liver transplantation could be more safely expanded.

Many studies have sought to identify the most useful clinical cut point for donor hemodynamics.[3,9,12,24] Early work hypothesized that, because DCD donors experience unavoidable ischemia prior to expiration, duration of DWIT might predict graft viability. de Vera et al.[23] reported that DWIT greater than 20 minutes was significantly associated with graft failure and increased biliary complication rates. Two additional studies using US national data identified 15- and 30-minute DWIT as cutoffs to avoid increased risk.[5,25] However, other researchers have failed to reproduce similar impacts of DWIT.[7,8] In the current

TABLE 9. Cox Proportional Hazards Modeling Graft Loss

	Unadjusted HR (95% CI)*	*P* Value*	Adjusted HR (95% CI)†	*P* Value†
HCC versus no HCC	1.94 (0.80-4.67)	0.140	2.53 (1.01-6.32)	0.047
CIT, per hour	1.36 (0.95-1.95)	0.091	1.50 (1.05-2.16)	0.027
MAP slow decliner (versus MAP rapid decliner)	2.10 (0.86-5.16)	0.105	3.13 (1.19-8.24)	0.021
SpO_2 slow decliner (versus SpO_2 rapid decliner)	1.88 (0.75-4.73)	0.177	—‡	—‡

*Univariate Cox proportional hazards models of graft loss.
†Multivariate Cox proportional hazards model of graft loss with the 3 indicated variables.
‡Insignificance ($P > 0.05$) and subsequent exclusion from the multivariate model.

practice, DWIT > 30 minutes is generally applied as a contraindication in DCD liver transplantation, but the real importance of DWIT remains controversial.[12] This appears to be partly attributable to the heterogeneity of donor hemodynamics during DWIT.

Abt et al.[9] recently reported on the importance of donor hemodynamics. Steeper values of SBP10 were associated with decreased risk of graft failure. In the present study, donors with steep SBP10 constitute MAP cluster 3 with the early rapid decliner phenotype. However, our study did not identify any association between SBP10 and graft survival (Table 8). Our study features a new category of donors based on hemodynamic trajectory (MAP cluster 2). Although MAP cluster 2 was characterized by longer DWIT, these donors initially showed stable hemodynamic values after withdrawal of life support; thus, they have greater values of SBP10. This period was followed by a sudden and rapid decline to asystole. Interestingly, the slope of terminal decline in MAP cluster 2 was almost identical to that of MAP cluster 3 (Fig. 2B). Congruity in the agonal phase trajectory just prior to circulatory death may explain the excellent prognosis in MAP cluster 2 and insignificant impact of SBP10 in our study. One possible explanation for lack of significance of hypoperfusion or hypoxia time (Table 8) is the presence of cluster 2 patients. We have characterized these patients extensively in Tables 2 and 4 and analysis reveals that although MAP cluster 3 has decreased hypoperfusion and hypoxia time, MAP cluster 2 has very similar durations to MAP cluster 1. In the case of hypoxia, this is a result of rapid desaturation during the course without corresponding hemodynamic collapse. However, MAP cluster 2 grafts proved quite viable. Similar hypoxia durations are associated with widely variable graft outcomes. Thus, if there would be an association between hypoxia or hypoperfusion duration and outcomes, they are less clear with small study populations and confounded by cluster 2 patients. Furthermore, the presence of this new category probably plays a role in diluting the significance of DWIT on graft outcomes. This is just 1 example where attempting to transform an entire hemodynamic course to a single continuous parameter inadequately segregates physiological heterogeneity.

Increased risk of biliary complications is the Achilles' heel of DCD liver transplantation. Taner et al.[10] reported that time from asystole to cross-clamp was most predictive of biliary complications in their large single-center experience. However, we found no association between ITBS and any of the intervals describing the donor hemodynamics. Interestingly, our study identified only 4 (5%) recipients with ITBS. Although the cluster analysis stratified the risk of graft failure, there was no significant association with the risk of biliary complications, likely because of the low event rate.

It has been shown that DCD recipients are at a relatively greater risk of HCC recurrence. A proposed mechanism involves increased ischemic insult, up-regulating adhesion molecules, and allowing better seeding of the donor graft by circulating tumor cells as compared to donation after brain death grafts.[24,26] Another multicenter study validated a role for both prolonged CIT and RWIT in increasing the rates of HCC recurrence.[27] In our series, 74% of HCC patients were within Milan criteria. We observed 13% of HCC patients undergoing DCD liver transplantation had recurrence leading to patient death at a median postoperative time of 907 days (range, 185-1196 days). Interestingly, the rate of graft loss from HCC recurrence was greater in MAP cluster 1 compared with 2 and 3 (33% versus 8%), although small event rate limits significance ($P = 0.169$).

As revealed in the multivariate analysis (Table 9), the use of DCD donors with the rapid declining MAP phenotype was associated with better graft survival. Conventionally, rapid decliners are donors with short DWIT and rapid deterioration of MAP (MAP cluster 3). In our study, we identified and characterized a new group of donors who have longer DWIT but who suddenly and rapidly declined following an initial period with stable hemodynamic values (MAP cluster 2). As shown in Fig. 2F, these 2 MAP clusters with different hemodynamic trajectories showed similar outcomes in terms of graft survival and exhibited a decreased risk of graft failure compared with the slow decliner (MAP cluster 1). During the initial phase of stable hemodynamics in MAP cluster 2 (Fig. 2D), the liver graft is well perfused until the donor enters the terminal decline. Such physiological hemodynamics may cause only minimal damage on the liver graft even with prolonged DWIT.

SpO_2 demonstrated a more rapid decline across all clusters as compared to MAP. We observed that many donors in SpO_2 cluster 1 had undetectable SpO_2 shortly after withdrawal of support which continued until asystole, showing a similar SpO_2 course as SpO_2 cluster 3 donors who had rapid desaturation and asystole, simultaneously. Such layered complexity contributed to statistically insignificant clustering of SpO_2 trajectories ($P = 0.108$; Fig. 2B). Although SpO_2 clustering seems to demonstrate clinically meaningful phenotypes, it was not a robust predictor of graft prognosis in the multivariate analysis (Table 8). This finding is despite the fact that the univariate survival analysis showed a clear trend (Figs. 3F and 4B). To

further determine the importance of SpO_2 in DCD donors, studies with a larger cohort are warranted.

There are important limitations to consider in the application of our findings. This study was carried out in a retrospective fashion, and thus, we are unable to infer definitive causal relationships. Additionally, there may be selection bias because surgeons may only accept organs that seem likely to survive; such decision processes might be associated with explanatory variables. Also, our center has been conducting a trial to evaluate the utility of tPA in DCD liver transplant,[13] a potential confounder. This clinical trial was underway during the generation of the data used in the present study. Importantly, the rate of tPA use was not significantly different by MAP ($P = 0.796$) or SpO_2 clusters ($P = 0.649$). Moreover, although Seal et al.[28] have provided evidence of dramatic effect of tPA, the Indiana group[29] and our own preliminary analysis (data not shown) do not support such results. Finally, it should be noted that no donors had DWIT longer than 38 minutes in our series. Therefore, applying our findings when the DWIT is far beyond this point may not be accurate.

In conclusion, despite physiological heterogeneity, we successfully categorized DCD donors into 3 clinically meaningful subgroups, particularly in the MAP clustering analysis. Our study revealed that donor hemodynamic trajectory was associated with graft survival. More importantly, this study is the first to describe and characterize a new category of donors who have initially stable and physiologic hemodynamics followed by sudden and rapid decline to asystole. Recipients who received this category of donor have better prognosis even with prolonged DWIT. This finding may explain the failure of our field to demonstrate consistent effects of DWIT, as well as inform the application of the 30-minute DWIT cutoff in predicting graft prognosis. Further studies will be needed to confirm the survival of this interesting new donor group and to identify additional variables to help determine graft viability and prognosis in DCD liver transplantation.

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