Survival Predictors in Liver Transplantation: Time-Varying Effect of Red Blood Cell Transfusion

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ABSTRACT

Background. Many attempts have been undertaken to better predict outcome after liver transplantation. The aim of this study was to identify the pre- and intraoperative variables that may influence the survival after liver transplantation, at a single institution.

Methods. Anesthetic records from 543 consecutive patients who underwent liver transplantation from June 2006 to June 2014 were reviewed in this retrospective study. Patients undergoing retransplantation were excluded from the analysis, as were patients with familial amyloid polyneuropathy. Preoperative variables studied were age, sex, Model for End-Stage Liver Disease score, primary diagnosis, cold ischemia time, preoperative international normalized ratio, serum albumin, and and hemoglobin levels. Intraoperative variables included were norepinephrine consumption, blood loss, red blood cell transfusion, and surgical time. Variables significant in the univariate analysis with a P value of $<.2$ were included in a multivariate Cox regression model.

Results. Only red blood cell transfusion (hazard ratio [HR], 1.16; 95% confidence interval [CI], 1.04–1.29) and female sex (HR, 1.71; 95% CI, 1.10–2.65) were identified as significant independent predictors for survival after liver transplantation. Because of proportionality assumption violation, the multivariate Cox regression model was subsequently upgraded by adding a time-varying interaction between red blood cell transfusion and time since liver transplantation. As a result, we found that at 3 months after liver transplantation, the rate of dying increased 14% (95% CI, 2%–26%) for each unit transfused, and at 6 months it increased 12% (95% CI, 0.3%–24%).

Conclusions. Red blood cell transfusion ceased to influence survival from 1 year onward.

LIVER transplantation (LT) is a standard of care for treatment of end-stage liver disease. LT is a highly complex procedure involving significant human and economic resources. Many efforts have been made to identify risk factors that could influence patient survival after LT. The knowledge and anticipation of these variables have allowed improvement in graft and patient survivals [1].

Our center has had an LT program since 1995 with ~1,100 liver transplants performed. In 2006 we started a clinical registry to audit and compare our practices. After 7 years of prospectively collecting data we decided to review it and retrospectively identify factors that could improve our clinical practice.

The aim of the present study was to identify the pre- and intraoperative variables that could influence survival after LT at our institution.

METHODS

After Investigation Review Board approval (003-DEFI-NA-CES), a retrospective analysis of anesthetic records from 543 consecutive adult LTs performed from June 2006 through June 2014 was conducted. Retransplantation ($n = 86$) and LT for familial amyloid polyneuropathy (FAP; $n = 116$) were excluded from the analysis because these were considered to be very different entities, leaving a final sample of 341 patients.

A team of 6 surgeons and 7 anesthesiologists remained steady throughout the study period. The anesthetic protocol was the same

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Table 1. Characteristics of the Study Population ($n = 341$)

Variable	Median (IQR) or *n* (%)
Age (y)	53 (49–59)
Sex	
Male	231 (67.7)
Female	110 (32.3)
Primary diagnosis	
Alcoholic cirrhosis	155 (45.45)
Fulminant hepatic failure	32 (9.38)
Hepatocarcinoma	51 (14.96)
Hepatitis B/C–related cirrhosis	42 (12.32)
Cholestatic liver disease	26 (7.62)
Miscellaneous	35 (10.26)
MELD score	13.5 (10–18)
Preoperative hemoglobin (mg/dL)	12 (10–13.5)
Preoperative INR	1.3 (1.1–1.6)
Preoperative serum albumin (mg/dL)	3.3 (2.9–3.8)
Cold ischemia time (min)	450 (340–600)
Intraoperative RBC (units)	2 (0–5)
Intraoperative blood loss (mL)	3,000 (1,700–5,300)
Intraoperative norepinephrine (mg)	1.73 (0.67–3.88)
Surgical time (min)	240 (219.5–278)

Abbreviations: IQR, interquartile range; MELD, Model for End-Stage Liver Disease; INR, international normalized ratio; RBC, red blood cells.

during the study period, as was the surgical technique. Patient follow-up for this study ended in August 2014.

All transplantations analyzed in this study ($n = 341$) were performed with donation after brain death grafts ($n = 246$) or with domino donors ($n = 95$). Domino donors are patients with FAP transplanted with cadaveric donors, whose liver is structurally normal and can be transplanted in recipients with other diagnoses. From the anesthetic records we analyzed the following preoperative variables: age, sex, Model for End-Stage Liver Disease (MELD) score, primary diagnosis, cold ischemia time, preoperative international normalized ratio, and serum albumin and hemoglobin levels. Intraoperative variables included norepinephrine consumption, blood loss, red blood cell (RBC) transfusion, and surgical time. MELD score was calculated with the use of the standard formula with no points added for special conditions [2].

Continuous variables are presented as median (interquartile range) and categoric variables as *n* (%). Influence on patient survival for each variable was first assessed with the use of univariate Cox regressions. Variables with statistical significance at $P < .20$ were selected for inclusion in a multivariate Cox regression model. In this final model, a statistical significance level of $P < .05$ was used. Linearity assumption in the log hazard for continuous variables and proportional hazards assumption were checked.

RESULTS

Study population characteristics for the sample of 341 patients are summarized in Table 1. The results of the univariate Cox regressions are summarized in Table 2. Only 5 variables were associated with patient survival (female sex, norepinephrine consumption, blood loss, RBC transfusion, and surgical time). Despite a *P* value of .20, age was also included in the multivariate model because of its expected natural importance to patient survival.

Table 2. Univariate Cox Regressions

Variable	HR	95% CI	*P* Value
Age (y)	1.01	0.99–1.03	.200
Female sex	1.37	0.92–2.04	.130
Primary diagnosis			.451
Alcoholic hepatic cirrhosis	1.00 (ref)		
Fulminant hepatic failure	1.55	0.82–2.93	.182
Hepatocellular carcinoma	1.57	0.88–2.81	.128
Cholestatic liver disease	1.48	0.72–3.05	.283
Hepatitis B/C–related cirrhosis	1.67	0.89–3.13	.109
Miscellaneous	1.19	0.60–2.36	.626
MELD score	1.00	0.98–1.04	.623
Preoperative hemoglobin (mg/dL)	0.95	0.87–1.03	.208
Preoperative INR	1.11	0.91–1.36	.287
Preoperative serum albumin	1.02	0.76–1.38	.882
Cold ischemia time (h)	0.99	0.93–1.06	.814
Intraoperative RBC (units)*	1.09	1.05–1.12	.000
Intraoperative blood loss (L)*	1.10	1.06–1.14	.000
Intraoperative norepinephrine (mg)*	1.07	1.03–1.10	.000
Surgical time (h)*	1.26	1.05–1.51	.013

Abbreviations: HR, hazard ratio; CI, confidence interval; others as in Table 1.
*$P < .2$.

After including those variables in a multivariate Cox regression model, only RBC transfusion (hazard ratio [HR], 1.16; 95% confidence interval [CI], 1.04–1.26) and female sex (HR, 1.71; 95% CI, 1.10–2.65) were identified as significant independent predictors of survival after LT (Table 3).

When checking the proportional hazards assumption for RBC transfusion, we found that the estimated HR could depend on time since LT. Addressing this issue, we improved the multivariate Cox regression model by adding a time-varying interaction between RBC transfusion and time since LT (Table 4).

According to these results, female patients were dying at a rate 66% higher than male transplant recipients and the 95% CI indicated that this increase in the rate of dying could be as much as 156% or as little as 7%.

Regarding RBC transfusion, because its effect on survival depended on time since LT, we calculated the HRs for 3 and 6 months as well as 1, 2, and 3 years since LT (Table 5). For each RBC unit transfused, we found that at 3 months after LT, the rate of dying increased 14% (95% CI, 2%–26%) and at 6 months increased 12% (95% CI, 0.3%–24%). From 1 year onward, RBC transfusion ceased to influence survival.

DISCUSSION

Unlike other published papers that dichotomized RBC transfusion according to median sample or massive transfusion definition, in the present study we analyzed RBC consumption as a continuous variable after confirming the linearity assumption. This way we could assess the influence on survival for each RBC unit transfused. For example, in our sample, 3 months after LT the hazard of mortality increased 14% (95% CI, 2%–26%) for each RBC unit transfused.

Table 3. Multivariate Cox Regression (Time-Varying Interaction With RBC Transfusion Not Included)

Variable	HR	95% CI	*P* Value
Age (y)	1.02	1.00–1.04	.061
Female sex*	1.71	1.10–2.65	.016
Intraoperative RBC (units)	1.16	1.04–1.29	.005
Intraoperative blood loss (L)	0.90	0.79–1.03	.135
Intraoperative norepinephrine (mg)	1.02	0.97–1.07	.508
Surgical time (h)	1.19	0.95–1.48	.125

Abbreviations as in Tables 1 and 2.
*$P < .05$.

Our results are in agreement with previously published studies demonstrating the association between intraoperative transfusion of RBC and adverse outcome after orthotopic LT [3–9]. The association between LT and major blood loss is well known. Nevertheless, advances made over the years in both surgical and anesthetic techniques, as well as better management of risk factors of massive blood loss, have resulted in a steady reduction in intraoperative hemorrhage and transfusion requirements. Ramos et al [5] and Massicotte et al [3] reported, respectively, that in 34% and 32% of the patients submitted to LT, RBC transfusion was not necessary. The percentage of LT without RBC transfusion in our sample was 30.5%.

Common complications of massive transfusions are immunologic adverse effects, metabolic derangements, infectious exposure with increased septic episodes, and acute lung injury. Moreover, blood transfusions have also been considered to be a surrogate marker for sicker patients and technically more difficult surgery, potentially confounding its role in outcome [10]. The exact mechanism linking transfusion and poor outcomes after LT remains otherwise unknown.

Reduction of the risk of transfusion can be attained if the principles of patient blood management are put into practice. These include preoperative recognition and treatment of anemia, reduction of perioperative blood loss, and the use of restrictive hemoglobin-based transfusion triggers [11]. Nevertheless, evidence that restrictive RBC transfusion is associated with better outcomes than a liberal approach is still lacking [12]. A remarkable variability between institutions in what concerns transfusion triggers as well as surgical techniques continues to be observed, and this can influence study results such that no conclusions can be drawn. As mentioned before, at our institution we adopt a restrictive transfusion policy. Transfusion therapy guided by blood patient management principles is the current practice at our hospital. The decision to transfuse in our center is an individual one, based on the stage of transplantation surgery, patient comorbidities, and physiologic triggers.

Table 4. Multivariate Cox Regression Including the Time-Varying Interaction With RBC Transfusion

Variable	HR	95% CI	*P* Value
Age (y)	1.02	1.00–1.04	.077
Female sex	1.66	1.07–2.56	.024
Intraoperative RBC (units)	1.25	1.12–1.40	.000
Intraoperative blood loss (L)	0.91	0.80–1.03	.147
Intraoperative norepinephrine (mg)	1.01	0.96–1.06	.803
Surgical time (h)	1.20	0.96–1.49	.105
Time-varying interaction with intraoperative RBC transfusion	0.98	0.97–0.99	.001

Abbreviations: as in Tables 1 and 2.

Table 5. Time-Varying Effect of RBC Transfusion on Patient Survival

Time Since LT	HR	95% CI	*P* Value
3 mo	1.14	1.020–1.257	.015
6 mo	1.12	1.003–1.240	.033
1 y	1.11	0.986–1.225	.070
2 y	1.09	0.968–1.210	.132
3 y	1.08	0.958–1.202	.183

Abbreviations: LT, liver transplantation; others as in Tables 1 and 2.

In Cox regression, the proportional hazard assumption must always be checked, otherwise a single HR for a given variable could misrepresent the effect on survival throughout the follow-up period [13]. Given the fact that the proportionality assumption did not hold in our sample, we decided to address this issue by adding a time-varying interaction term to the Cox model.

In the present study, the influence of RBC transfusion on survival was statistically significant for only the 1st 6 months after LT and not for the remaining survival time (Table 5). In fact, most adverse outcomes associated with transfusion are temporally close to its administration. In a literature review, we did not find studies discussing a time-varying effect of RBC transfusion in liver transplant patients. However, in other clinical domains, such as cardiac surgery and major trauma, investigators consistently described a time-varying effect of RBC transfusion on survival [14]. Some of those articles showed a biphasic pattern with an early phase (1st 6 months of follow-up) of diminished survival associated with RBC transfusion and a late phase with less or no significant association [15,16].

Regarding the association between female sex and decreased survival found in our sample, other authors did not find sex to be an independent risk factor for survival in LT [6–8]. The association between sex and survival has been studied in other specialties, namely, cardiac and orthopedic surgeries. According to those studies, women submitted to cardiac surgery had poor survival [17–21], whereas in orthopedic surgery better outcomes for women were observed [22–25]. In their discussions, the authors of these papers point out very different explanations for their results. Regarding our sample, this result could reflect confounding involving variables not included in our study, such as donor/recipient sex or liver size mismatch. Given this uncertainty, we consider any explanation for this result to be speculative.

In our population, the MELD score was relatively low, with a median of 13.5, which could translate to low risk of mortality. However, as previously mentioned, exception points were not included. Also, our patient waiting list for

LT is managed according to variables other than just MELD (uncontrollable ascites, encephalopathy, etc), differentiating our center from others.

FAP is an autosomal hereditary systemic disease characterized by hepatic production of an abnormal protein that causes systemic amyloidosis. These patients were excluded from the analysis because hepatic functions and structure are preserved, thus being associated with a less demanding surgery and a reduced amount of intraoperative blood loss (median RBC transfusion in our center is 0 units; unpublished data).

The present study was retrospective in nature and the database restricted to the clinical data in the anesthesia records, explaining why some of the potential important variables are missing from the analysis, such as donor characteristics. This represents an important limitation in this study that should be addressed in future analysis regarding this topic. Nevertheless, the time-varying effect of RBC transfusion is not mentioned in earlier studies concerning the same subject. So we consider this to be a starting point for the development of other clinical studies that focus on the effect of RBC transfusion on survival of patients submitted to LT and the way that this could be influenced by the time factor. On the other hand, given the low rate of transfusion of other blood products (29% for fresh-frozen plasma, 15% for platelets), this analysis was not pursued.

In conclusion, we found RBC transfusion to be an independent risk factor for survival in LT patients at our institution, its effect on survival being limited to the early phase of follow-up (<1 y).

REFERENCES

[1] Smith CM, Davies DB, McBride MA. Liver transplantation in the United States: a report from the UNOS Liver Transplant Registry. Clin Transpl 1999;1:23–34.

[2] Freeman Jr RB, Wiesner RH, Harper A, McDiarmid SV, Lake J, Edwards E, et al. The new liver allocation system: moving toward evidence-based transplantation policy. Liver Transpl 2002;8:851–8.

[3] Massicotte L, Sassine MP, Lenis S, Seal R, Roy A. Survival rate changes with transfusion of blood products during liver transplantation. Can J Anesth 2005;52:148–55.

[4] Boin I, Leonardi M, Luzo A, Cardoso AR, Caruy CA, Leonardi LS. Intraoperative massive transfusion decreases survival after liver transplantation. Transplant Proc 2008;40:789–91.

[5] Ramos E, Dalmau A, Sabate A, Lama C, Llado L, Figueras J, et al. Intraoperative red blood cell transfusion in liver transplantation: influence on patient outcome, prediction of requirements, and measures to reduce them. Liver Transpl 2003;9:1320–7.

[6] Wiederkehr JC, Igreja MR, Nogara MS, Gonçalves NL, Wiederkehr BA, Nascimento VB, et al. Analysis of survival after primary liver transplantation: multivariate analysis of 155 cases in a single center. Transplant Proc 2010;42:511–2.

[7] Cywinski JB, Alster JM, Miller C, Vogt DP, Parker BM. Prediction of intraoperative transfusion requirements during orthotopic liver transplantation and the influence on postoperative patient survival. Anesth Analg 2014;118:428–37.

[8] de Boer MT, Christensen MCA, van der Hilst CS, Hendriks HGD, Slooff MJH, Porte RJ. The impact of intraoperative transfusion of platelets and red blood cells on survival after liver transplantation. Anesth Analg 2008;106:32–44.

[9] Rana A, Petrowsky H, Hong JC, Agopian VG, Kaldas FM, Farmer D, et al. Blood transfusion requirement during liver transplantation is an important risk factor for mortality. J Am Coll Surg 2013;216:902–7.

[10] de Boer MT, Molenaar IQ, Hendriks HG, Slooff MJ, Porte RJ. Minimizing blood loss in liver transplantation: progress through research and evolution of techniques. Dig Surg 2005;2:265–75.

[11] Clevenger B, Mallet SV. Transfusion and coagulation management in liver transplantation. World J Gastroenterol 2014;20:6146–58.

[12] Holst LB, Petersen MW, Haase N, Perner A, Wetterslev J. Restrictive versus liberal transfusion strategy for red blood cell transfusion: systematic review of randomised trials with meta-analysis and trial sequential analysis. BMJ 2015;350:h1354.

[13] Bellera C, MacGrogan G, Debled M, de Lara CT, Brouste V, Mathoulin-Pélissier S. Variables with time-varying effects and the cox model: some statistical concepts illustrated with a prognostic factor study in breast cancer. BMC Med Res Methodol 2010;10:20.

[14] Holcomb JB, Junco DJ, Fox EE, Wade CE, Cohen MJ, Schreiber MA, et al. The Prospective, Observational, Multicenter, Major Trauma Transfusion (PROMMTT) study: comparative effectiveness of a time-varying treatment with competing risks. JAMA Surg 2013;148:127–36.

[15] Koch CG, Li L, Duncan AI, Mihaljevic T, Loop FD, Starr NJ, et al. Transfusion in coronary artery bypass grafting is associated with reduced long-term survival. Ann Thorac Surg 2006;81:1650–7.

[16] Surgenor SD, Kramer RS, Olmstead EM, Ross CS, Sellke FW, Likosky DS, et al. The association of perioperative red blood cell transfusions and decreased long-term survival after cardiac surgery. Anesth Analg 2009;108:1741–6.

[17] Mandegar MH, Marzban M, Lebaschi AH, Ghaboussi P, Alamooti ARA, Ardalan A. Gender influence on hospital mortality after coronary artery bypass surgery. Asian Cardiovasc Thorac Ann 2008;16:231–5.

[18] Miśkowiec DL, Walczak A, Jaszewski R, Marcinkiewicz A, Ostrowski S. Independent predictors of early mortality after CABG in single centre experience—does the gender matter? Kardiol Pol 2015;73:109–17.

[19] Kim C, Redberg RF, Pavlic T, Eagle KA. A systematic review of gender differences in mortality after coronary artery bypass graft surgery and percutaneous coronary interventions. Clin Cardiol 2007;30:491–5.

[20] Lehmkuhl E, Kendel F, Gelbrich G, Dunkel A, Oertelt-Prigione S, Babitsch B, et al. Gender-specific predictors of early mortality after coronary artery bypass graft surgery. Clin Res Cardiol 2012;101:745–51.

[21] Becker ER, Rahimi A. Disparities in race/ethnicity and gender in in-hospital mortality rates for coronary-artery bypass surgery patients. J Natl Med Assoc 2006;98:1729–39.

[22] Librero J, Peiró S, Leutscher E, Merlo J, Bernal-Delgado E, Ridao M, et al. Timing of surgery for hip fracture and in-hospital mortality: a retrospective population-based cohort study in the Spanish National Health System. BMC Health Serv Res 2012;12:15.

[23] Grewal K, Wijeysundera DN, Carroll J, Tait G, Beattie WS. Gender differences in mortality following noncardiovascular surgery: an observational study. Can J Anesth 2012;59:255–62.

[24] Shoda N, Yasunaga H, Horiguchi H, Matsuda S, Ohe K, Kadono Y, et al. Risk factors affecting in-hospital mortality after hip fracture: retrospective analysis using the Japanese Diagnosis Procedure Combination Database. BMJ Open 2012;2:e000416.

[25] Singh JA, Kwoh CK, Richardson D, Chen W, Ibrahim SA. Gender and surgical outcomes and mortality after primary total knee arthroplasty: a risk adjusted analysis. Arthritis Care Res (Hoboken) 2013;65:1095–102.