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Risk factors associated with blood transfusion in liver transplantation

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To explore preoperative and operative risk factors for red blood cell (RBC) transfusion requirements during liver transplantation (LT) and up to 24 h afterwards. We evaluated the associations between risk factors and units of RBC transfused in 176 LT patients using a log-binomial regression model. Relative risk was adjusted for age, sex, and the model for end-stage liver disease score (MELD) (adjustment 1) and baseline hemoglobin concentration (adjustment 2). Forty-six patients (26.14%) did not receive transfusion. Grafts from cardiac-death donors were used in 32.61% and 31.54% of non-transfused and transfused patients, respectively. The transfused group required more reoperation for bleeding ($P = 0.035$), longer mechanical ventilation after LT ($P < 0.001$), and longer ICU length of stay ($P < 0.001$). MELD and hemoglobin concentrations determined RBC requirements. For each unit of increase in the MELD score, 2% more RBC units were transfused, and non-transfusion was 0.83-fold less likely. For each 10-g/L higher hemoglobin concentration at baseline, 16% less RBC transfused, and non-transfusion was 1.95-fold more likely. Ascites was associated with 26% more RBC transfusions. With an increase of 2 mm from the baseline in the A10FIBTEM measurement of maximum clot firmness, non-transfusion was 1.14-fold more likely. A 10-min longer cold ischemia time was associated with 1% more RBC units transfused, and the presence of post-reperfusion syndrome with 45% more RBC units. We conclude that preoperative correction of anemia should be included in LT. An intervention to prevent severe hypotension and fibrinolysis during graft reperfusion should be explored.

***Trial register*: European Clinical Trials Database (EudraCT 2018–002,510-13) and ClinicalTrials.gov (NCT01539057).**

Keywords Blood component transfusion, Hemostasis, Liver transplantation, Mean hemoglobin concentration, Morbidity, Mortality, Thromboelastometry

Abbreviations

A10EXTEM	MCF amplitude at 10 min by EXTEM
A10FIBTEM	MCF amplitude at 10 min by FIBTEM
CIT	Cold ischemia time
EXTEM	Extrinsic thromboelastometry for fibrin tissue factor activation
FIBTEM	Thromboelastometry for fibrin tissue factor activation and platelet inhibition
INR	International normalized ratio
IRB	Institutional review board
LT	Liver transplantation
MCF	Maximum clot firmness
MELD	Model for End-Stage Liver Disease
NASH	Nonalcoholic steatohepatitis
PRS	Post-reperfusion syndrome
PT	Prothrombin time
PT/INR	INR of PT
PTT	Partial prothromboplastin time
RBCs	Packed red blood cells

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IRR	Incidence rate ratio
OR	Odds ratio

Although clear improvements have been made in liver transplantation (LT) techniques over time, the need for infusion of blood components or products and red blood cells (RBCs) remains high, influencing LT outcomes[1–3]. Moreover, as older and sicker patients began to enter waiting lists over the past decade[4], the infusion of blood components has even increased, and higher MELD (Model for End-Stage Liver Disease) scores have been reported[5]. A recent randomized controlled trial on fibrinogen administration and blood product requirements found that nearly 73% of patients required RBC correction during LT and in the following 24 h[6]. This figure was consistent with low baseline hemoglobin concentrations: around 94% of patients with baseline hemoglobin concentrations < 110 g/L required transfusions[6].

Thromboelastometry assesses the viscoelastic properties of whole blood by reflecting the function and interactions of plasma, blood cells, and platelets. In LT, coagulation management guided by thromboelastometry is widely accepted[7]. Although, the hemoglobin concentration is known to increase the risk of RBC transfusion[8], maximum clot firmness (MCF) measured by thromboelastmetry for fibrin tissue factor activation and platelet inhibition (Fibtem) has also been shown to predict blood product requirements[8]. However, little information is available on how hemoglobin and baseline coagulation status and intraoperative factors interact in driving RBC requirements.

We aimed to explore all modifiable preoperative and intraoperative risk factors associated with RBC transfusion during LT surgery and within 24 h afterwards. Data were collected prospectively for a multicenter series of liver recipients in a randomized controlled trial of two strategies for fibrinogen correction during LT[6].

Patients and methods

Data from a multicenter, hemoglobin-stratified, randomized controlled trial on fibrinogen administration and blood product requirements by our group[6] were used for a secondary analysis in the initial protocol approved by the Institutional Review Board (IRB) of the lead hospital (Hospital Universitari de Bellvitge, approval number AC033/18) and the IRBs of other participating centers (Hospital Universitario de Cruces and Hospital Clínic i Provincial de Barcelona). All research was performed in accordance with relevant guidelines/regulations. Patients were enrolled in the study if they provided written informed consent. No organs (livers) were procured from prisoners, all the institutions via which all organs were procured while taking care to did not violate the privacy of donors. This trial was registered in the European Clinical Trials Database (EudraCT 2018–002,510-13) and ClinicalTrials.gov (NCT01539057).. Patients and methods has been previously described in part in the former study[6].

Patients

All adults scheduled for LT were assessed for eligibility from August 2, 2019, to November 2, 2021. Low risk of intraoperative transfusion (preoperative hemoglobin concentration > 130 g/L) was the main exclusion criterion. Patients on aspirin, warfarin, or other anticoagulation therapy were also excluded; patients with complete portal vein thrombosis or a known history of thromboembolic events in the last 30 days or bleeding disorders; patients undergoing an acute retransplantation; patients whose indication for LT was familial polyneuropathy; or those receiving a graft from a living donor given the variability in surgical techniques in those settings.

Graft and anesthesia management, surgery, and transfusion protocols[6]

Liver allografts were preserved in University of Wisconsin solution. Organ recovery from controlled cardiac-death donors met criteria[9], which stipulate normothermic regional perfusion in the recovery of organs from non-living donors.

The anesthesia protocol was monitored to ensure consistency and compliance across all research centers. Vena cava preservation was attempted in all the patients. If such preservation was not feasible, a venovenous bypass or a complete caval clamp was used, and an additional portacaval shunt was used if the surgeon considered it necessary. At the end of surgery, all patients were mechanically ventilated and transferred to a postoperative intensive care unit.

The protocols for blood product and component transfusions were monitored to ensure consistency and compliance across the three hospitals. The infusion criteria were as follows: RBCs to maintain hemoglobin above 80 g/L and platelet concentrates if the count fell below 30 000/mm^3. Intravenous tranexamic acid boluses (500 mg) were administered if fibrinolysis (> 15% lysis at 60 min) was detected by extrinsic thromboelastometry for fibrin tissue factor activation (Extem). Cell-saver devices were not used in this study. Hemostatic management was guided by thromboelastometry. In case of massive bleeding (> 150 mL/min), we monitored both MCF amplitudes by Extem and Fibtem at 10 min (A10Fibtem). If we detected a value of < 15 mm by Extem or a clotting time > 300 s by Fibtem, we simultaneously infused 4 units of RBCs, 1 g of tranexamic acid, 2 g of fibrinogen concentrate, 1 unit of apheresis platelets, and 15 mL/kg of fresh frozen plasma.

Variables of interest

Variables considered as possible risk factors for the primary outcome, RBC transfusion, including recipient and donor characteristics, and intraoperative data related to LT. Recipient characteristics were age, sex, body mass index, diabetes mellitus, hypertension, cardiac disease, respiratory disease, indication for LT, MELD score, Child–Pugh score, and hospitalization when LT was scheduled. Additional patient characteristics studied were baseline hemoglobin, creatinine, glomerular filtration rate, sodium, plasma fibrinogen levels, partial thromboplastin time (PTT), international normalized ratio of prothrombin time (PT/INR), platelet count, and baseline thromboelastometry profile. Donor characteristics included donor type (after brain or cardiac death), donor age,

and cold ischemia time (CIT). Intraoperative data included surgical time, surgical techniques, warm ischemia time, infusions of blood components and products, fibrinogen concentrate, tranexamic acid, crystalloids, albumin, and the development of post-reperfusion syndrome (PRS).

Statistical analysis

Descriptive statistics for patients and surgeries are expressed, for discrete variables reported as counts (n) and percentages (%), and means and SD or medians (interquartile range [IQR] or range) for continuous variables.

A log-binomial regression model was used to evaluate the association between the risk factors and RBC transfusion. Associations were adjusted for age, sex, and MELD score (adjustment 1) based on their positive associations with the dependent outcome variable in most predictive models, indicating that there were substantial interactions between them and other modifiable variables. Given the clear influence of baseline hemoglobin, an additional adjustment including this variable was made (adjustment 2). Relative risk and adjusted relative risk were calculated with 95% CIs. We used the zero-inflated Poisson model, which generates a separate process for the expected number of RBC transfusions among those who received any (count model-IRR), and the possibility of having no RBC requirements (zero-inflated model-OR). The regression coefficients, standard errors, and constants were obtained. All analyses were performed using the statistical software package R, version 4.1.0 for Windows (http://www.R-project.org, The R Foundation).

The protocol, informed consent sheets, statistical analysis plan, case record forms, and datasets were stored by the IRB of the lead hospital and IDIBELL Foundation. This work was reported in line with the statement on Strengthening the Reporting of Observational Studies in Epidemiology (STROBE).

Ethical approval and consent to participate

The study protocol was approved by the Institutional Review Board (IRB) of the lead hospital (University Hospital of Bellvitge, approval number AC 033/18) and the IRBs of the other participating centers (University Hospital of Cruces and Clinic Hospital of Barcelona).

Informed consent

Patients were enrolled if they provided written informed consent and were approved by the Institutional Review Board of the lead hospital (University Hospital of Bellvitge, approval number AC 033/18).

Results

A total of 306 LTs were performed during the analysis period, and 93 of the operated patients were excluded because their baseline hemoglobin concentrations indicated that they would be at low risk for transfusion (> 130 g/L). Thirty-one additional patients were excluded for other reasons listed above, leaving a total of 182 patients enrolled[6]. After six procedures were cancelled, 176 patients were finally included in the analysis.

Forty-six patients (26.14%) did not receive RBC transfusions during or within 24 h after LT (non-transfused group), and 130 patients (73.86%) received at least one unit of RBCs (transfused group). Patient characteristics and baseline hemoglobin, coagulation, and fibrinogen values of both groups are shown in Table 1. There were more women in the non-transfused group, and the ages were similar in both the groups. Alcoholic cirrhosis was more common in the transfusion group, whereas diagnosis of hepatocarcinoma was more common in the non-transfusion group. Child-C scores and ascites were also more common in the transfused group, where the MELD scores, PTT, and PT/INR were higher. Preoperative acute kidney injury was more common in the transfusion group. Donor grafts after cardiac death were used in 15 patients (32.61%) in the non-transfused group and 41 patients (31.54%) in the transfused group. Vena cava preservation was used in nearly all patients (96%) in both groups, and a portacaval shunt was used in 32.40% of non-transfused patients and 38.46% of transfused patients.

There were no differences in median (IQR) findings between non-transfused and transfused patients for CIT 381.50 (268.75; 443.25) min vs. 362.50 (288.75; 444.25) min ($P = 0.861$) or warm ischemia time 40.00 (27.50; 47.00) minutes vs 35.00 (26.00; 50.00) min ($P = 0.61$). Post-reperfusion syndrome was present in 16 non-transfused patients (34.78%) and 66 transfused patients (50.77%) ($P = 0.090$) Median surgical times were 362.50 (301.25; 1438.50) minutes vs 410.00 (305.75; 1433.75) minutes in the two groups, respectively ($P = 0.477$).

Hemoglobin, platelet count, fibrinogen values, and Extem and Fibtem MCF measurements at each stage of LT are shown in Table 2 along with blood product, blood component, and fluid therapy requirements. Hemoglobin, hemostasis, and coagulation values were significantly lower in the transfused group at baseline (T1) and during all phases of the LT. Platelet counts and Extem MCF values increased during LT in non-transfused patients, even if they did not receive blood components. Plasma fibrinogen concentrations decreased during LT in both the groups. Infusions of fibrinogen concentrate were administered mainly after graft reperfusion (T3) in both groups, although the amounts were significantly higher in transfused patients.

Only one patient (2.17%) in the non-transfused group required reoperation related to non-bleeding complications, whereas 20 patients (15.38%) in the transfused group underwent reoperation (15 of them for bleeding complications) ($P = 0.035$). Thrombotic complications (hepatic artery, portal vein, and other systemic thromboses) developed in 8.70% of the non-transfused group and 3.08% of the transfused group; $P = 0.209$. Acute renal failure occurred in both groups, in four patients (8.70%) in the non-transfused group and in 23 (17.69%) in the transfused group ($P = 0.224$). The median (IQR) duration of mechanical ventilation after LT was significantly shorter in the non-transfused group, at 9.00 h (7.40; 12.30 h) than in the transfused group, at 13.95 h (10.00;19.65 h) in the transfused group ($P < 0.001$). The median (IQR) ICU length of stay was significantly shorter in the non-transfused group, 2.00 days (2.00; 3.00) than in the transfused group, 3.00 days (2.75;5.00) in the transfused group ($P < 0.001$). Nine patients were re-admitted in the ICU, all from the transfusion group.

	Non-transfused group (n = 46)	Transfused group (n = 130)
Age (years)*	57.80 (10.34)	58.58 (7.97)
Male	35 (76.09%)	104 (80.00%)
Female	11 (23.91%)	26 (20.00%)
Weight (kg)	76.23 (15.83)	78.87 (14.76)
Height (cm)	168.11 (9.88)	169.65 (8.57)
BMI (kg/m^2)	26.91 (4.84)	27.38 (4.64)
Alcoholic cirrhosis	20 (43.48%)	82 (63.08%)
NASH	3 (6.52%)	13 (10.00%)
Tumor	10 (21.74%)	7 (5.38%)
Biliary cirrhosis	5 (10.87%)	8 (6.15%)
Other	9 (19.57%)	21 (16.15%)
Prior abdominal surgery	12 (26.09%)	45 (34.62%)
Diabetes	13 (28.26%)	45 (34.62%)
Partial portal thrombosis	1 (2.17%)	11 (8.46%)
Altered echocardiogram	7 (15.22%)	22 (16.92%)
Pleural effusion + Ascites	12 (26.08%)	80 (61.54%)
Sodium (mEqu/L)†	138.00 [135.25;140.00]	134.00 [130.00;138.00]
Creatinine (mg/dL)†	0.80 [0.65;0.95]	1.03 [0.80;1.31]
Preoperative AKI	56 (44%)	7 (15.22%)
MELD score†	12.00 [9.00;19.00]	20.00 [16.00;25.00]
Child–Pugh score		
A	19 (42.22%)	8 (6.20%)
B	20 (44.44%)	39 (30.23%)
C	6 (13.33%)	82 (63.57%)
Hemoglobin (g/L)†	108 [104;118]	88.0 [81.0;99.0]
< 95 g/L	1 (2.17%)	68 (52.31%)
≥ 95 g/L	45 (97.83%)	62 (47.69%)
Platelet count (10^3/mm^3)†	84.00 [56.75;112.00]	71.00 [51.00;96.00]
PTT†	1.13 [1.02;1.23]	1.24 [1.10;1.40]
INR†	1.35 [1.21;1.52]	1.65 [1.41;1.91]
Fibrinogen (g/L)†	2.74 [1.83;3.60]	1.90 [1.30;2.66]
EXTEM coagulation time (s)†	63.00 [57.00;67.00]	66.00 [61.00;77.00]
EXTEM MCF (mm)†	56.50 [50.25;65.75]	49.00 [41.00;57.00]
FIBTEM MCF (mm)†	14.50 [10.00;20.50]	9.00 [6.00;14.00]

Table 1. Patient characteristics and hemoglobin, coagulation, and fibrinogen baseline values according to RBC transfusion group. Data are n (%), mean (SD)*,median [interquartile range]† AKI, acute kidney injury; EXTEM, extrinsic thromboelastometry for fibrin tissue factor activation; FIBTEM, thromboelastometry for fibrin tissue factor activation and platelet inhibition; INR, international normalized ratio; MCF, maximum clot firmness; MELD, Model for End-Stage Liver Disease; NASH, nonalcoholic steatohepatitis; PTT, partial thromboplastin time.

Retransplantation or death occurred in one patient (2.17%) in the non-transfusion group and seven (5.38%) in the transfused group ($P = 0.682$).

The IRR and ORR values for the expected units of packed RBCs transfused among those who received any RBC and the likelihood of having no RBC requirements before and after adjustments for age, sex, MELD, donor type, and hemoglobin are shown in Table 3. The main baseline factors associated with transfusion requirements (units of packed RBCs) and non-transfusions are shown in (Fig. 1).

For each unit increase in the MELD score, we observed a 2% increase in transfused RBCs units, and non-transfusion was 0.83-fold less likely. For each 10-g/L rise in hemoglobin concentration at baseline, 16% fewer units of RBCs were transfused, and non-transfusion was 1.95-fold more likely. The presence of ascites was associated with a 26% increase in RBC units transfused whereas each 0.5-g/L increase in baseline fibrinogen concentration was associated with a 5% decrease in RBC units. With an increase of 2 mm in the A10FIBTEM measurement from the baseline, non-transfusion was 1.14-fold more likely. A 10-min longer CIT was associated with 1% more RBC units transfused, and the presence of PRS with 45% more RBC units.

	Non-transfused group (n = 46)	Transfused group (n = 130)	*p* value
Hemoglobin (g/L)			
T1	108 [104;118]	88.0 [81.0;99.0]	< 0.001
T2	114 [105;122]	87.0 [77.0;99.0]	< 0.001
T3	106 [98.0;112]	82.0 [73.0;92.0]	< 0.001
T4	101 [92.2;106]	86.5 [80.0;92.0]	< 0.001
Platelet counts (10^3/mm^3)			
T1	84.0 [56.8;112]	71.0 [51.0;96.0]	0.066
T2	107 [76.0;140]	85.0 [63.5;118]	0.033
T3	103 [74.8;134]	77.0 [59.2;112]	0.003
T4	112 [77.0;153]	88.0 [63.5;121]	0.004
Plasma fibrinogen (g/L)			
T1	2.74 [1.83;3.60]	1.90 [1.30;2.66	0.001
T2	2.62 [2.21;3.35]	2.00 [1.50;2.60]	0.001
T3	2.18 [1.75;2.96]	1.61 [1.32;2.16]	0.001
T4	2.16 [1.70;2.77]	1.70 [1.38;2.16]	0.001
Extem MCF (mm)			
T1	56.5 [50.2;65.8]	49.0 [41.0;57.0]	< 0.001
T2	61.0 [54.0;67.8]	53.0 [48.0;60.0]	< 0.001
T3	57.0 [51.2;65.8]	50.5 [45.0;57.0]	< 0.001
T4	59.5 [53.0;66.8]	54.0 [48.0;59.0]	< 0.001
Fibtem MCF (mm)			
T1	15.5 [10.0;22.5]	10.0 [6.00;16.0]	< 0.001
T2	15.0 [11.0;20.8]	11.0 [8.00;14.0]	< 0.001
T3	13.5 [11.0;18.8]	10.0 [8.00;13.0]	< 0.001
T4	15.0 [12.0;18.0]	12.0 [9.00;15.0]	< 0.001
Fibrinogen administered (g)			
T1–T2	11 (23.9%), 2.00 [2.00;4.00]	71 (54.6%), 5.00 [2.00;6.00]	< 0.001*, 0.072†
T2–T3	14 (30.4%), 2.50 [2.00;4.00]	90 (69.2%), 5.00 [4.00;7.00]	< 0.001*, 0.012†
T3–T4	18 (39.13%), 3.50 [2.00;5.50]	101 (77.7%), [4.00;9.00]	< 0.001*, < 0.001†
During LT			
RBCs (units)	–	117 (90.00%), 3.00 [2.00;5.00]	
FFP (units)	1 (2.17%); 2.00 [2.00;2.00]	21 (16.15%), 2.00 [2.00;4.00]	0.027* 0.483†
Platelets (units)	1 (2.17%); 1.00 [1.00;1.00]	23 (17.69%), 1.00 [1.00;2.00]	0.017* 0.486†
Tranexamic acid	10 (21.74%)	57 (43.85%)	0.013
During and 24 h after LT			
RBCs (units)	NA	130 (100%), 4.00 [2.00;6.00]	
> 6 units RBCs (n)	NA	32 (24.62%)	
FFP (units)	1 (2.17%); 2.00 [2.00;2.00]	31 (23.85%), 2.00 [2.00;4.00]	0.002* 0.409†
Platelets (mL)	1 (2.17%); 1.00 [1.00;1.00]	40 (30.77%), 1.00 [1.00;2.00]	< 0.001* 0.397†
Fibrinogen(g)	18 (39.13%); 3.50 [2.00;5.50]	104 (80.00%), 7.00 [4.00;9.25]	< 0.001* < 0.001†
Fluid therapy (mL)	4500.00 [3650.00;6097.00]	5500.00 [4300.00;7701.00]	0.010

Table 2. Hemoglobin, platelet count, and fibrinogen values; Extem and Fibtem, MCF measurements at each stage of LT; and RBC units, blood component and fluid therapy requirements according to RBC transfusion group. Data are expressed as n (%) or median [IQR]. Extem, extrinsic thromboelastometry for fibrin tissue factor activation; FFP, fresh frozen plasma; Fibtem, thromboelastometry for fibrin tissue factor activation and platelet inhibition; IQR, interquartile range; LT, liver transplantation; MCF, maximum clot firmness; RBCs, red blood cell; T1, baseline; T2, 10 min after portal clamp; T3, 10 min after reperfusion of the liver graft; T4, end of procedure. *p-value referring to percentage comparison. †p-value referring to the median comparison.

Discussion

Factors associated with RBC requirements during LT and in the following 24 h in this study, consistent with previous studies[5,10–15], were high MELD scores and low baseline hemoglobin concentrations indicated by a median hemoglobin concentration of 88 g/L in our cohort. These two factors clearly influence blood transfusions. Altered laboratory parameters for hemostasis and coagulation, including thromboelastometry values, were also associated with RBC transfusion requirements. However, after adjusting for MELD score and hemoglobin concentration, baseline PT/INR, fibrinogen concentration, platelet count, and Extem values did not predict transfusion requirements. High baseline A10Fibtem values favored non-transfusion.

	IRR.ORR	a1 IRR.ORR	a2 IRR.ORR
Sex: Female vs Male †‡	0.62 [0.48;0.79] *	–	–
Sex: Female vs Male § ¶	1.09 [0.44;2.67]	_	_
Age†‡	0.97 [0.87;1.07]	–	–
Age §¶	0.89 [0.6;1.32]	–	–
Alcoholic Cirrhosis†‡	1.31 [0.99;1.72]	1.22 [0.94;1.6]	1.16 [0.89;1.52]
Alcoholic Cirrhosis§¶	0.46 [0.23;0.95]*	0.96 [0.24;1.32]	0.79 [0.26;2.08]
Hepatocarcinoma†‡	1.13 [0.6;2.12]	1.29 [0.65;2.32]	1.14 [0.61;2.12]
Hepatocarcinoma§¶	5.07 [1.77;14.57]*	3.61 [1.09;11.99] *	3.2 [0.68;15.04]
Prior abdominal surgery†‡	0.94 [0.72;1.24]	1.07 [0.81;1.4]	1.03 [0.8;1.34]
Prior abdominal surgery§¶	0.65 [0.29;1.43]	0.35 [0.13;0.9] *	0.35 [0.12;1.08]
Diabetes†‡	0.86 [0.64;1.17]	0.9 [0.68;1.2]	0.88 [0.67;1.16]
Diabetes§¶	0.71 [0.32;1.56]	0.42 [0.15;1.21]	0.41 [0.11;1.47]
Altered Echocardiogram †‡	1.08 [0.74;1.58]	1.04 [0.71;1.54]	1.08 [0.7;1.51]
Altered Echocardiogram§¶	0.89 [0.34;2.33]	0.71 [0.19;2.73]	0.57 [0.13;2.55]
Pleural Ascites effusion ‡†	1.42 [1.18;1.72] *	1.26 [1.03;1.54] *	1.09 [0.89;1.35]
Pleural Ascites effusion §¶	0.12 [0.05;0.29] *	0.2 [0.07;0.53] *	0.26 [0.08;0.82] *
Preoperative AKI ‡†	0.92 [0.63;1.34]	0.91 [0.66;1.25]	0.86 [0.63;1.19]
Preoperative AKI §¶	0.12 [0.01;0.9] *	0.16 [0.02;1.13]	0.21 [0.02;1.98]
Partial Portal thrombosis‡†	1.64 [1.07;2.52] *	1.61 [1.07;2.42] *	1.35 [0.86;2.04]
Partial Portal thrombosis§¶	0.25 [0.3;2.08]	0.28 [0.09;2.51]	0.84 [0.24;4.99]
Hemoglobin ‡†	0.83 [0.78;0.88] *	0.84 [0.79;0.89] *	–
Hemoglobin §¶	2.19 [1.65;2.91] *	1.95 [1.41;2.68] *	–
Platelets † ‡	0.98 [0.96;1]	0.99 [0.97;1.01]	1 [0.97;1.02]
Platelets § ¶	1.07 [1;1.14] *	1 [0.92;1.08]	0.99 [0.9;1.08]
Fibrinogen ‡†	0.93 [0.89;0.97] *	0.95 [0.91;1] *	0.96 [0.92;1.01]
Fibrinogen §¶	1.26 [1.09;1.45] *	1.11 [0.95;1.30]	1.11 [0.93;1.32]
PT†‡	1.27 [1.06;1.53] *	1.24 [0.95;1.6]	1.08 [0.82;1.41]
PT §¶	0.06 [0.01;0.27] *	0.36 [0.06;2.08]	1.14 [0.15;8.69]
CT Extem †‡	1.03 [1;1.06] *	1.01 [0.98;1.04]	1.02 [0.99;1.05]
CT Extem §¶	0.7 [0.52;0.95] *	0.84 [0.62;1.14]	0.81 [0.55;1.2]
A10Extem †‡	0.94 [0.88;1.01]	1.01 [0.93;1.09]	1.01 [0.94;1.1]
A10Extem §¶	1.72 [1.25;2.35] *	1.22 [0.84;1.77]	1.23 [0.81;1.88]
A10Fibtem †‡	0.96 [093;0.99] *	0.98 [0.95;1.01]	0.99 [0.96;1.02]
A10Fibtem §¶	1.23 [1.11;1.37] *	1.14 [1.02;1.28] *	1.13 [1.00;1.29] *
Creatinine †‡	1.05 [0.94;1.17]	0.99 [0.88;1.11]	0.98 [0.87;1.11]
Creatinine §¶	0.11 [0.03;0.42] *	0.12 [0.02;0.75] *	0.25 [0.04;1.59]
Sodium †‡	0.99 [0.97;1] *	0.99 [0.98;1.01]	1 [0.98;1.01]
Sodium §¶	1.16 [1.06;1.27] *	1.1 [1;1.21] *	1.06 [0.95;1.18]
MELD ‡†	1.02 [1.01;1.03] *	–	
MELD §¶	0.83 [0.78;0.9] *	–	
Child–Pugh score: C vs A†‡	1.35 [0.9;2.01]	1.11 [0.72;1.7]	1 [0.65;1.56]
Child–Pugh score: C vs A §¶	0.03 [0.01;0.1] *	0.07 [0.02;0.3] *	0.07 [0.01;0.42] *
Donor type: DCD vs DBD †‡	1 [0.84;1.19]	–	
Donor type: DCD vs DBD §¶	1.05 [0.5;2.21]	–	
CIT †‡	1.01 [1.01;1.02] *	1.01 [1;1.02] *	1.01 [1.01;1.02] *
CIT §¶	0.99 [0.96;1.03]	1.01 [0.96;1.05]	0.98 [0.94;1.03]
WIT ‡†	1.03 [0.99;1.07]	1.02 [0.98;1.06]	1.02 [0.98;1.06]
WIT §¶	0.99 [0.83;1.19]	1.08 [0.89;1.32]	1.04 [0.8;1.33]
Crystalloids + albumin, total †‡	1.01 [1.01;1.01]	1.01 [1.01;1.01]	1.01 [1;1.01]
Crystalloids + albumin total §¶	0.98 [0.96;1]	0.98 [0.96;1]	0.98 [0.95;1.01]
Tranexamic acid:† ‡	1.2 [1.01;1.41] *	1.19 [1.01;1.41] *	1.24 [1.05;1.47] *
Tranexamic acid: §¶	0.36 [0.16;0.8] *	0.46 [0.18;1.16]	0.3 [0.09;0.97] *
Post-reperfusion syndrome: † ‡	1.44 [1.22;1.7] *	1.45 [1.23;1.73] *	1.5 [1.27;1.79] *
Post-reperfusion syndrome: §¶	0.55 [0.27;1.12]	0.73 [0.31;1.71]	0.61 [0.22;1.64]
Surgical time †‡	1.01 [1.00;1.02]	1.01 [1.00;1.02]	1.01 [1.00;1.02]
Surgical time §¶	1 [0.96;1.04]	0.99 [0.95;1.41]	0.98 [0.93;1.03]
Continued			

	IRR.ORR	a1 IRR.ORR	a2 IRR.ORR
Temporary Portocaval shunt †‡	0.87 [0.67;1.13]	0.8 [0.63;1.09]	0.9 [0.71;1.15]
Temporary Portocaval shunt §¶	0.74 [0.35;1.58]	0.77 [0.32;1.86]	0.29 [0.08;1.04]

Table 3. Relative risk values for factors associated with RBC transfusion requirements († count model; ‡ incidence rate ratios) and with non-transfusion (§ zero-inflated model; ¶ odds ratio), once adjusted by sex, age and MELD (a1 IRR.ORR) and by sex, age, MELD and baseline hemoglobin concentration (a2 IRR. ORR). *Indicates a p value < 0.05. AKI, acute kidney injury; CIT, cold ischemia time; Extem, extrinsic thromboelastometry for fibrin tissue factor activation; FFP, fresh frozen plasma; Fibtem, thromboelastometry for fibrin tissue factor activation and platelet inhibition; MELD, Model for End-Stage Liver Disease; PT, prothrombin time; WIT, warm ischemia time.

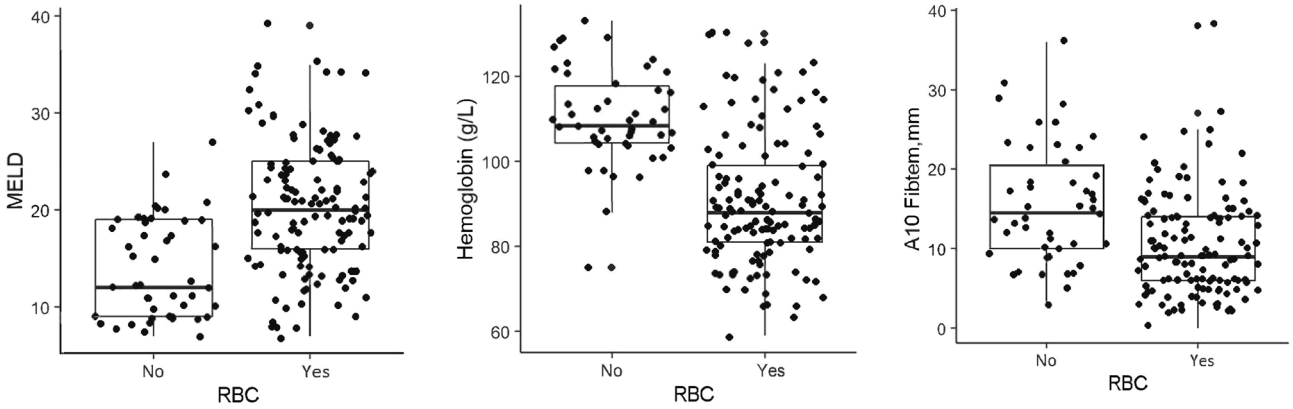

Figure 1. Main baseline factors associated with RBC transfusion. Box plots for ("yes") or with non- RBC transfusion ("no"). Horizontal lines indicate medians, boxes the interquartile range, and dots the individual patient data. A10Fibtem, the maximum clot firmness amplitude measured at 10 min by thromboelastometry for fibrin tissue factor activation and platelet inhibition. MELD, Model for End-Stage Liver Disease, RBC, packed red blood cells.

The presence of preoperative ascites was associated with higher transfusion requirements in our study. Bleeding during LT is associated with portal hypertension, surgical difficulties, and hemostatic failure. In one large series, the preoperative hepatic venous pressure gradient was associated with intraoperative bleeding, yet hemoglobin concentration remained the main factor associated with RBC requirements[16] Portal hypertension has nonetheless been reported to cause clinically significant ascites, which could induce systemic hyperfibrinolysis[17]. The only baseline factor favoring non-transfusion, other than hemoglobin concentration, was A10Fibtem. However, raising the A10Fibtem target to 11 mm failed to provide clinical benefits in the trial we ran[6].

Donor type was not relevant to either increasing RBC transfusion requirements or favoring nontransfusion. This is unsurprising given that normothermic and hypothermic oxygenation perfusion machines currently improve graft viability after procurement[18–20] longer duration of surgery and CIT was associated with RBC requirements, confirming the influence of a difficult surgical field[21,22].

Although one study found that machine learning managed to predict which patients are at risk for bleeding based on a large number of preoperative and intraoperative variables[23], the authors were unable to propose measures to reduce RBC requirements. In contrast, we used a regression model that generated separate predictions for the expected number of units of packed RBCs and the likelihood of non-transfusion, thus recognizing factors that can be modified to potentially improve transfusion outcomes. The clinical urgency of doing so is illustrated by the fact that 24% of transfused patients received more than six units of packed RBCs.

Even if baseline plasma fibrinogen concentration was not associated with RBC requirements after adjusting for cofactors, we observed a decrease in plasma fibrinogen concentration in both groups during LT. The decrease in plasma levels of fibrinogen at the reperfusion of the liver graft with respect to the values of the anhepatic phase was higher in the transfusion group than the non-transfusion group (median decrease, -0.44 vs -0.22 g/l) despite the fact that 77% of patients of the transfusion group received fibrinogen concentrate vs. 39% in the non-transfusion group. This fact was more pronounced in patients who presented PRS.

After graft reperfusion, several concurrent events occur: the return of normal splanchnic circulation, addition of the perfusate of the washed liver, and release of substances from the new graft[24]. PRS is caused by liver graft and recipient risk factors, the cold storage perfusion fluid of the liver graft produces the known effects of hypothermia, hyperkalemia and hypervolemia, events that are accentuated in large grafts. Together these events might result in major surgical bleeding and severe hemodynamic disturbances. In our series, PRS and bolus injection of tranexamic acid were linked; 47.56% of patients with PRS received tranexamic acid during reperfusion. Reasons that can account for this scenario include some degree of hyperfibrinolysis related to hypotension[25,26],

specifically continuous oozing bleeding or longer coagulation times (longer than 300 s at A10FIBTEM in extreme cases in our cohort), and MCF EXTEM < 15 mm.

Among the modifiable intraoperative risk factors were associated with larger transfusion requirements: the presence of PRS and fibrinolysis. The identification of these factors suggests that devising a protocol before reperfusion of the liver graft may improve outcomes. Therefore, the "timing" is important and performing this action before reperfusion of the liver graft has not been explored. For this protocol, we suggest preventing PRS with vasopressor support and infusing a bolus of 500 mg of tranexamic acid to prevent potential fibrinolysis. Such a strategy, although feasible, would require the support of data from randomized clinical trials.

For clinically compensated candidates whose medical condition before LT remains unchanged according to standard measures and MELD status, only hemoglobin optimization can clearly influence RBC requirements. Anemia, which is present in approximately 25% of cirrhotic patients, is multifactorial and involves iron and vitamin B deficiency, hypersplenism, hemolysis, gastrointestinal bleeding, bone marrow deficiency, and hepatitis C virus treatment[27,28]. In two retrospective series, anemia was linked to bleeding and major postoperative complications and one-year mortality[5,29]. In another series of cirrhotic patients, oral iron correction improved hemoglobin values[30]. In our cohort, the median for the highest quartile of patients without RBC administration was 118 g/L, this value suggests a target for a testable preoperative optimization strategy, even though optimization is clinically challenging in patients with liver disease[31]. Based on the studies we reviewed and the findings we report now, we believe that hemoglobin optimization merits further consideration as a correctable preoperative factor using intravenous administration of iron in candidates on waiting lists.

The main limitation of this study was related to the exclusion criteria of the trial. We excluded those patients who would have clearly influenced RBC requirements. The exclusion of 93 patients with hemoglobin concentrations > 130 g/L, in whom transfusion was not expected, conceivably changed the analysis. Another limitation is related to the low number of patients with massive transfusion, in comparison to old retrospective outcome studies[10,12]. In our series only 32 patients (18.2%) required more than 6 RBC's. So the degree of influence of these patients on major complications in the group of transfused patients is limited, especially if the incidence of major complications it is downloaded as it happens in our study. Nevertheless, the patients in our series correspond to the patients who are currently included in the waiting list in the European registries[32]. However, the conclusions we can draw would not necessarily change, although it is necessary to stress that our cohort represented patients not previously assumed to be at risk who might have benefited from an intervention to reduce transfusion requirements. We believe that this finding supports the clinical value of our results. The strengths of the study are the quality of the patient recruitment and data collection, monitored by an independent committee, available on request.

We conclude that the correction of anemia in LT candidates should be included in the preoperative patient blood management protocols. We also propose a randomized clinical trial testing a strategy for preventing PRS and fibrinolysis during graft reperfusion.

Data availability

The data that support the findings of this study are not openly available due to reasons of sensitivity and are available from the corresponding author upon reasonable request at http://idibell.cat/serveis/serveis-cientifico tecnics/bioestadistica/.

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References

1. Ramos, E. *et al.* Intraoperative red blood cell transfusion in liver transplantation: influence on patient outcome, prediction of requirements, and measures to reduce them. *Liver Transpl.* **9**, 1320–1327. https://doi.org/10.1016/jlts.2003.50204 (2003).
2. De Boer, M. T. *et al.* The impact of intraoperative transfusion of platelets and red blood cells on survival after liver transplantation. *Anesth. Analg.* **106**, 32–44. https://doi.org/10.1213/01.ane.0000289638.26666 (2008).
3. Viguera, L. *et al.* Baseline haemoglobin and thromboelastometry are predictive of red blood cell requirements and one-year mortality in liver transplantation. *Transfus Apher. Sci.*, https://doi.org/10.1016/j.transci.2021.103259 (2022).
4. Kwong, A. *et al.* OPTN/SRTR 2018 annual data report: Liver. Scientific register of transplant recipients. *Am. J. Transplant* **20**(1), 193–299. https://doi.org/10.1111/ajt.15674 (2020).
5. Collas, O., Robertson, F. P., Fuller, B. J. & Davidson, B. R. Anaemia in patients with chronic liver disease and its association with morbidity and mortality following liver transplantation. *Int. J. Surg.* **53**, 48–52. https://doi.org/10.1016/j.ijsu.2018.02.053 (2018).
6. Caballero, M. *et al.* Blood component requirements in liver transplantation: effect of two thromboelastometry-guided strategies for bolus fibrinogen infusion — the TROMBOFIB randomized trial. *J. Thromb. Haemost.* **21**, 37–46. https://doi.org/10.1016/j.jtha.2022.10.025 (2023).
7. U. Yoon, J. Bartoszko, D. Bezinover, Biancofiore G, Forkin KT, Rahman S, et al. Intraoperative transfusion management, antifibrinolytic therapy, coagulation monitoring and the impact on short-term outcomes after liver transplantation - A systematic review of the literature and expert panel recommendations. Clin Transplant 2022; https://doi.org/10.1111/ctr.14637
8. Sabate, A. *et al.* Assessment of rotational thromboelastometry for the prediction of red blood cell requirements in orthotopic liver transplantation. *Minerva Anestesiol.* **84**, 447–454. https://doi.org/10.23736/S0375-9393.17.12023-7 (2018).
9. Hessheimer AJ, Gastaca M, Minambres E, Colmenero J, Fondevila C, In representation of the SETH Working Group on DCDet al. Donation after circulatory death liver transplantation: consensus statements from the Spanish Liver Transplantation Society. *Transpl Int* 2020; 33: 902–916, https://doi.org/10.1111/TRI.13619.
10. Cywinski, J. B., Alster, J. M., Miller, C., Vogt, D. P. & Parker, B. M. Prediction of intraoperative transfusion requirements during orthotopic liver transplantation and the influence on postoperative patient survival. *Anesth. Analg.* **118**, 428–437. https://doi.org/10.1213/ANE.0b013e3182a76f19 (2014).
11. Xia, V. W. *et al.* Preoperative characteristics and intraoperative and vasopressor requirements in patients with low vs high MELD scores. *Liver Transpl.* **12**, 614–620 (2006).

12. McClusky, S. *et al.* Derivation of a risk index for the prediction of massive blood transfusion in liver transplantation. *Liver Transpl.* **12**, 584–593. https://doi.org/10.1002/lt.20868 (2006).
13. Cleland, S., Corredor, C., Ye, J. J., Srinivas, C. & McCluskey, S. A. Massive haemorrhage in liver transplantation: Consequences, prediction and management. *World J. Transpl.* **6**, 291–305. https://doi.org/10.5500/wjt.v6.i2.291 (2016).
14. Fernanes, D. S. *et al.* Pre-operative predictors of red blood cell transfusion in liver transplantation. *Blood Transfus.* **22**, 1–4 (2016).
15. Liu, C. H. *et al.* A quantitative model to predict blood use in adult orthotopic liver transplantation. *Transfus Apher Sci* **53**, 386–392. https://doi.org/10.1016/j.transci.2015.07.008 (2015).
16. Sanahuja, J. M. *et al.* Portal hypertension has no role in perioperative bleeding during liver transplantation with systematic porto-caval shunt. *HPB (Oxford).* **25**(4), 454–462. https://doi.org/10.1016/j.hpb.2023.01.009 (2023).
17. Thaler, J. *et al.* Intraperitoneal activation of coagulation and fibrinolysis in patients with cirrhosis and ascites. *Thromb. Haemost* **122**, 353–362. https://doi.org/10.1055/a-1515-9529 (2022).
18. Schlegel, A. *et al.* Outcomes of DCD liver transplantation using organs treated by hypothermic oxygenated perfusion before implantation. *J. Hepatol.* **70**, 50–57. https://doi.org/10.1016/j.jhep.2018.10.005 (2019).
19. Patrono, D. *et al.* Outcome of liver transplantation with grafts from brain-dead donors treated with dual hypothermic oxygenated machine perfusion, with particular reference to elderly donors. *Am. J. Transpl.* **22**, 1382–1395. https://doi.org/10.1111/ajt.16996 (2022).
20. Brüggenwirth, I. *et al.* Prolonged preservation by hypothermic machine perfusion facilitates logistics in liver transplantation: A European observational cohort study. *Am. J. Transpl.* **22**, 1842–1851. https://doi.org/10.1111/ajt.17037 (2022).
21. Azoulay, D. *et al.* Defining surgical difficulty of liver transplantation. *Ann. Surg.* **277**, 144–150. https://doi.org/10.1097/SLA.0000000000005017 (2023).
22. Ausania, F. *et al.* Development of a preoperative score to predict surgical difficulty in liver transplantation. *Surgery* **172**, 1529–1536. https://doi.org/10.1016/j.surg.2022.07.001accessonline (2022).
23. Park, S. *et al.* Development of machine learning models predicting estimated blood loss during liver transplant surgery. *J. Pers. Med.* **12**, 1028. https://doi.org/10.3390/jpm12071028 (2022).
24. Hilmi, I. *et al.* The impact of post-reperfusion syndrome on short-term patient and liver allograft outcome in patients undergoing orthotopic liver transplantation. *Liver Transpl.* **14**, 504–508. https://doi.org/10.1002/lt.21381 (2008).
25. Kim, E. H. *et al.* Evaluation of "flat-line" thromboelastography after reperfusion during liver transplantation. *Transpl. Proc.* **47**, 457–459. https://doi.org/10.1016/j.transproceed.2014.11.032 (2015).
26. Abuelkasem, I., Tanaka, K., Planinsic, R. & Sakai, T. Comparison between thrombelastography and thromboelastometry in hyper-fibrinolysis detection during adult liver trans- plantation. *Br. J. Anaesth.* **116**, 507–512. https://doi.org/10.1093/bja/aew023 (2016).
27. Sanvisens, A. *et al.* Gender and liver, nutritional and metabolic alterations of severe alcoholism: a study of 480 patients. *Med. Clin. (Barc).* **137**, 49–54. https://doi.org/10.1016/j.medcli.2010.11.030 (2011).
28. Patel YA, Kappus MR, Muir AJ. Anemia in liver disease. In: Management of Anemia. Springer, New York; 2018. p. 129–42.
29. Lichtenegger, A. *et al.* The association of pre-operative anaemia with survival after orthotopic liver transplantation. *Anaesthesia* **75**, 472–478. https://doi.org/10.1111/anae.149184 (2020).
30. Rashidi-Alavijeh, J. *et al.* Implications of anemia and response to anemia treatment on outcomes in patients with cirrhosis. *JHEP Rep.* **5**, 100688. https://doi.org/10.1016/j.jhepr.2023.100688 (2023).
31. Gkamprela, E., Deutsch, M. & Pectasides, D. Iron deficiency anemia in chronic liver disease: etiopathogenesis, diagnosis and treatment. *Ann. Gastroenterol.* **30**, 405–413. https://doi.org/10.20524/aog.2017.0152 (2017).
32. Müller, Ph., Kabacam, G., Vibert, E., Germani, G. & Petrowsky, H. Current status of liver transplantation in Europe. *Int. J. Surg.* **82S**, 22–29. https://doi.org/10.1016/j.ijsu.2020.05.062 (2020).

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Author contributions

A.S., L.P., A.B.: literature search, figures, study design, data collection, data analysis, data interpretation, funding acquisition, writing the original draft, and making subsequent revisions. M.C., R.P., R.G., S.L.l: literature search, data collection, data interpretation, and approval of the manuscript. P.H., J.P.: Monitoring, statistical analysis plan, data analysis, data interpretation. All authors approved this submission. All authors have provided their consent for publication.

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Competing interests

The authors declare no competing interests.

Additional information

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