

# Early serum tacrolimus levels predict long-term chronic kidney disease after liver transplantation

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**OBJECTIVE:** To investigate risk factors for the development of chronic kidney disease and death two years post-liver transplantation.

**METHOD:** Associations between clinical and laboratory parameters and the development of chronic kidney disease and survival two years post-liver transplant were analyzed in a cohort of 148 adult patients with hepatic cirrhosis consecutively submitted to liver transplantation in a referral Brazilian center.

**RESULTS:** Median age at liver transplantation was 56 (range, 20-73) years, and 105 (70.9%) patients were males. The prevalence of chronic kidney disease at two years post-liver transplantation was: stage 1 or no chronic kidney disease, 27.5%; stage 2, 33.8%; stage 3, 34.6%; stages 4-5, 4.7%. Four variables were independently associated with the stage of chronic kidney disease two years after liver transplantation: (i) age (at liver transplantation), (ii) male gender, (iii) median tacrolimus levels in the first three months post-liver transplantation, and (iv) median of serum creatinine in the first six months post-liver transplantation. Two variables showed independent association with death in two years post-liver transplantation: (i) stay in Intensive Care Unit for three or more days after the liver transplantation surgery and (ii) median of serum creatinine levels in the first six months post-liver transplantation equal or higher than 1.3 mg/dL.

**CONCLUSIONS:** Administration of the lowest effective dose of tacrolimus and adoption of strategies to spare renal function are important measures to reduce the risk of late chronic kidney disease and death post-liver transplantation especially in high risk patients.

**KEYWORDS:** Chronic kidney disease; Kidney dysfunction; Liver transplantation; Tacrolimus.

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## INTRODUCTION

Liver transplantation (LT) is the main treatment for end-stage liver disease. Its results are becoming increasingly favorable. This has been attributed to the greater experience of the surgical and post-LT care teams, improved immunosuppressive therapy, and improvement of the selection criteria for transplant candidates.<sup>1</sup>

Chronic kidney disease (CKD) is a critical complication after LT. As the rate of survival of LT recipients increases, so does the incidence of renal disease. The risk of developing severe CKD has been reported to be close to 10%, 18% and 25% at two, five and ten years after LT, respectively.<sup>2,3</sup> Several potential causes are recognized, including hepatitis C virus (HCV)-related renal disease, hypertension, diabetes

mellitus, and other glomerular disorders such as IgA nephropathy.<sup>4-7</sup> Although there is a range of differential diagnosis for the etiology of CKD, several authors believe that the use of calcineurin inhibitors is a major contributor to kidney dysfunction in the context of LT.<sup>8,9</sup> LT recipients with end-stage kidney disease requiring chronic dialysis are at increased risk of death compared to those without this condition.<sup>10</sup>

Studies aimed to investigate the risk factors for the development of CKD among LT recipients show some conflicting results.<sup>11,12,13</sup> Additionally, the prevalence of CKD has been poorly investigated in the Brazilian liver transplant population.<sup>14,15</sup> Thus, the aims of this study were to investigate the prevalence of the different stages of CKD two years post-LT, the risk factors associated with its development, and factors that influence survival two years after LT in a referral center in Brazil.

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## ■ MATERIALS AND METHODS

This cohort study included adult patients (age  $\geq 18$  years) with liver cirrhosis who underwent LT consecutively at Hospital das Clínicas, Universidade Federal de Minas Gerais, Belo Horizonte, Brazil, between January 2006 and July 2009. Patients submitted to a liver-kidney double transplantation were excluded. This study was approved by the Research Ethics Committee of the Institution (ETIC 0242.0.203.000-10).

The associations between different clinical and laboratory parameters, and the development of CKD and survival at two years post-LT were investigated. Data were obtained by referring to the medical records. The pre-LT factors investigated were: (1) age at LT; (2) gender; (3) etiology of the liver disease; (4) presence of hepatocellular carcinoma; (5) presence of ascites (detected on clinical examination and/or ultrasound); (6) CKD stage based on the creatinine clearance, calculated by the Cockcroft Gault method:  $(140 - \text{age}) \times \text{weight} / \text{serum creatinine} \times 72$  for males, or multiplying the result by 0.85 for females;<sup>16</sup> (7) presence of systemic arterial hypertension and (8) diabetes mellitus (defined by fasting plasma glucose  $\geq 126$  mg/dL at two different occasions and/or glycohemoglobin (Hb A1C)  $\geq 6.5\%$  and/or presence of classic symptoms of hyperglycemia or hyperglycemic crisis associated with a random plasma glucose  $\geq 200$  mg/dL); (9) 24 hour-proteinuria (total protein  $\geq 300$  mg/dL in a 24-hour urine collection); (10) serum levels of sodium; (11) Model for End-Stage Liver Disease (MELD)-score (calculated according to the Organ Procurement and Transplantation Network [OPTN]  $9.6 \times \log$  serum creatinine mg/dL +  $3.8 \times \log$  bilirubin mg/dL +  $11.2 \times \log$  INR [International Normalized Ratio] + 6.4);<sup>17,18</sup> (12) the need for hemodialysis. The perioperative factors investigated were cold ischemia time, need for transfusion of packed red cells and number of units transfused. Regarding the donor, gender and age were the variables investigated. The following factors were considered after LT: length of stay in the Intensive Care Unit (ICU) and time on mechanic ventilation after the LT surgery, need for transfusion of packed red cells and number of units transfused, need for hemodialysis, use of amphotericin B, vancomycin, polymyxin E, acyclovir, and tacrolimus levels in the first three months and serum creatinine levels in the first six months post-LT.

### Immunosuppressive regimen

All patients received methylprednisolone during the surgery, followed by prednisone in progressively smaller doses until discontinuation three months after the transplant. Calcineurin inhibitors – tacrolimus or cyclosporine – were administered to 143 (96.6%) and five (3.4%) patients, respectively. Target trough serum levels of tacrolimus were as follows: 10-15 ng/mL in the first month post-LT, 8-10 ng/mL between the second and sixth months, and 3-7 ng/mL after six months of transplantation. In the patients receiving cyclosporine-based treatment, the trough level was maintained between 200 and 300 ng/mL during the first three months post-LT and then reduced to 100-200 ng/mL thereafter. Mycophenolate mofetil or mycophenolate sodium was administered to 27 (18.2%), azathioprine to two (1.4%); and daclizumab and sirolimus to only one patient (0.7%), each drug.

### Renal function evaluation

Based on the creatinine clearance calculated by the Cockcroft Gault method, patients were grouped according to the National Kidney Foundation Disease Outcomes Quality Initiative (NKF/KDOQI)<sup>TM</sup> guidelines, in stages 1 to 5: stage 1, glomerular filtration rate (GFR)  $> 90$  mL/min/ $1.73$  m<sup>2</sup>; stage 2, GFR = 89-60 mL/min; stage 3, GFR = 59-30 mL/min; stage 4, GFR = 29-15 mL/min; and stage 5, GFR  $< 15$  mL/min or dialysis. Each stage of CKD was defined in the presence of a sustained reduction of the GFR for at least three months.<sup>19</sup> As an abnormal urinalysis is required for the diagnosis of stage 1 CKD and was not routinely available, we grouped the patients with a GFR  $> 90$  mL/min as a single category, namely, no or mild CKD (stage 1). Stages 4 and 5 were also grouped because of the small number of patients.

To analyze the serum creatinine levels during the first six months after LT, a value per month was randomly selected for each patient and the median of these values was used; and to analyze the serum tacrolimus levels during the first three months post-LT, the median of all the measurements performed for each patient during the period was employed.

### Statistical analysis

Initially, the association between the different variables and the stage of CKD at two years post-LT was investigated by univariate analysis, using the chi-square test (linear trend) or the Kruskal-Wallis test, as appropriate. The variables that proved to be associated with the outcome on univariate analysis at the significance level of 0.25 were included in the logistic regression model. Since the dependent variable was ordinal (stages for CKD), multivariate ordinal logistic regression was performed using the partial proportional odds model.

Kaplan Meier curves and univariate Cox regression were employed in the comparative analysis of the independent variables and survival at two years post-LT. The Log-Rank test was used to compare the survival curves. The variables that proved to be associated with the outcome on univariate analysis at the significance level of 0.25 were selected for the multivariate Cox regression analysis. The model's adequacy was assessed by analysis of the residues.

The values of  $p \leq 0.05$  were deemed statistically significant. For statistical analysis, the SPSS version 19 software (SPSS INC., Chicago IL) was used, with the exception of the partial proportional odds model multivariate analysis, which was performed using the STATA<sup>®</sup> software (Copyright 1996-2012 StataCorp LP).

## ■ RESULTS

A total of 148 patients were included (105 [70.9%] males and 43 [29.1%] females) with a median age at LT of 56 years (range, 20-73 years). The indications for LT were: chronic HCV infection (26.4%), alcoholic liver disease (25%), cryptogenic cirrhosis (20.9%), autoimmune cirrhosis (6.8%), chronic hepatitis B virus (HBV) infection (5.4%), primary biliary cirrhosis (4.7%), secondary biliary cirrhosis (3.5%), primary sclerosing cholangitis (2.7%), and others (4.8%).

Tables 1 to 3 show the observed frequency of the pre- (Table 1), peri- (Table 2) and post-LT (Table 3) variables analyzed and the results of the univariate analysis on the comparison between those variables and CKD stage at two years post-LT.

**Table 1 - Frequency of pre-liver transplantation factors and their comparison with the chronic kidney disease stage at two years after liver transplantation (univariate analysis)**

Variables	Chronic kidney disease stage				P value
	1/no CKD	2	3	4-5	
Gender					0.001*
Male N (%)	30 (85.7)	35 (81.4)	22 (52.4)	4 (51.7)	
Female N (%)	5 (14.3)	8 (18.6)	20 (47.6)	3 (42.9)	
Age at LT (years; median; range)	45 (23-62)	55 (36-67)	59 (26-74)	63 (23-73)	<0.001**
Cirrhosis etiology					0.129*
Viral N (%)	13 (37.1)	9 (20.9)	16 (38.1)	5 (71.4)	
Criptogenic + alcoholic N (%)	11 (31.4)	26 (60.5)	16 (38.1)	1 (14.3)	
Autoimmune N (%)	5 (14.3)	7 (16.3)	7 (16.7)	1 (14.3)	
Others N (%)	6 (17.1)	1 (2.3)	3 (7.1)	0 (0.0)	
Ascites N (%)	23 (67.6)	30 (71.4)	31 (73.8)	5 (71.4)	0.688*
Diabetes mellitus N (%)	6 (17.1)	9 (21.4)	14 (33.3)	1 (14.3)	0.220*
Hepatocellular carcinoma N (%)	9 (25.7)	9 (20.9)	9 (21.4)	2 (28.6)	0.778*
Arterial hypertension N (%)	3 (8.8)	12 (28.6)	8 (19.0)	2 (28.6)	0.317*
24-hour proteinuria N (%)	9 (90.0)	17 (100.0)	16 (100.0)	1 (100.0)	0.142*
Pre-LT hemodialysis (%)	1 (2.9)	0 (0.0)	1 (2.4)	1 (14.3)	0.366*
CKD before LT					0.086*
Stage 1 or no CKD N (%)	25 (73.5)	23 (53.5)	8 (19.0)	0 (0.0)	
Stage 2 N (%)	4 (11.8)	10 (23.3)	15 (35.7)	2 (28.6)	
Stage 3 N (%)	4 (11.8)	9 (20.9)	17 (40.5)	3 (42.9)	
Stages 4 and 5 N (%)	1 (2.9)	1 (2.3)	2 (4.8)	2 (28.5)	
Pre-LT sodium (mEq/L; median; range)	137 (82-145)	137 (124-146)	137 (124-148)	136 (127-143)	0.946**
MELD-score (median; range)	18 (8-33)	17 (7-32)	17,5 (9-36)	13 (9-21)	0.502**

\*Chi-squared test.

\*\*Kruskal-Wallis test.

Abbreviations. CKD: chronic kidney disease; LT: liver transplantation; MELD: Model for End-Stage Liver Disease.

**Table 2 - Frequency of peri-liver transplantation factors and their comparison with the chronic kidney disease stage at two years after liver transplantation (univariate analysis)**

Variables	Chronic kidney disease stage				P value
	1/no CKD	2	3	4-5	
Peri-LT transfusion of PRC					0.285*
No transfusion N (%)	17 (48.6)	17 (39.5)	12 (29.3)	3 (42.9)	
1-2 units N (%)	9 (25.7)	12 (27.9)	14 (34.1)	1 (14.3)	
3-5 units N (%)	6 (17.1)	8 (18.6)	10 (24.4)	2 (28.6)	
6-10 units N (%)	1 (2.9)	6 (14.0)	4 (9.8)	1 (14.3)	
> 10 units N (%)	2 (5.7)	0 (0.0)	1 (2.4)	0 (0.0)	
Cold ischemia time					0.708*
< 12 hours N (%)	9 (69.2)	17 (85.0)	17 (73.9)	2 (100.0)	
Gender of donor					0.052*
Male N (%)	18 (51.4)	26 (60.5)	26 (61.9)	7 (100.0)	
Female N (%)	17 (48.6)	17 (39.5)	16 (38.1)	0 (0.0)	
Age of donor (years; median; range)	29 (17-58)	34 (8-62)	40 (6-58)	30 (22-46)	0.674**

\*Chi-squared test.

\*\*Kruskal-Wallis test.

Abbreviations. LT: liver transplantation; PRC: packed red cells.

Nineteen (12.8%) of the 148 patients initially included in the study were lost to follow-up or died; therefore, they were not included in the comparative analysis between clinical and laboratory data and two-year post-LT CKD stage. Two years after LT, 35 (27.5%) of the remaining 129 patients presented CKD stage 1 or did not show any evidence of kidney dysfunction, 43 (33.8%) showed evidence of stage 2, 44 (34.6%) of stage 3, and seven (4.7%) of stages 4-5.

Serum tacrolimus levels throughout the first three months post-LT exhibited a relatively steady median of 11.3 ng/mL and a wide range (1.7-30 ng/mL). Serum creatinine concentrations throughout the entire two-year post-LT period exhibited a similarly steady median of 1.29 mg/dL, with an equally wide range of variation (0.34-12.2 mg/dL).

**Evolution of CKD.** Thirty-four patients were in stage 3 CKD pre-LT; two years post-LT, the renal function improved

in 12 (35.3%), four to stage 1, eight to stage 2; in 18 (52.9%) patients, it remained at stage 3; in three (8.8%) subjects, kidney function worsened, two to stage 4 and one to stage 5. One patient died. Nine patients were in stage 4 CKD pre-LT; two years after the transplant, it improved to stage 1 in one patient, to stage 2 in two patients, and to stage 3 in one patient. It remained at stage 4 in two patients, whereas three died. Two subjects with stage 5 CKD before LT died.

In the multivariate analysis, the variables independently associated with post-LT CKD stage were: recipient's age at LT and gender, median of the tacrolimus concentrations in the first three months post-LT, and median of the serum creatinine levels in the first six months post-LT (Table 4). For each year increase in the recipient's age at LT, the risk of developing stages 4-5 CKD at two years post-LT was 1.22 times higher; for each unit increase in the median of the

**Table 3 - Frequency of post-liver transplantation factors and their comparison with the chronic kidney disease stage at two years after liver transplantation (univariate analysis)**

Variables	Chronic kidney disease stage				P value
	1/no CKD	2	3	4-5	
Packed red cells transfusion					0.082*
No transfusion N (%)	17 (48.6)	11(26.2)	11(27.5)	1 (16.7)	
1-2 units N (%)	5 (14.3)	7 (16.7)	8 (20.0)	2 (33.3)	
3-5 units N (%)	5 (14.3)	8 (19.0)	8 (20.0)	0 (0.0)	
6-10 units N (%)	6 (17.1)	6 (21.4)	6 (15.0)	2 (33.3)	
> 10 units N (%)	2 (5.7)	7 (16.7)	7 (17.5)	1 (16.7)	
Stay in ICU post-LT					0.760*
≤ 3 days N (%)	16 (45.7)	19 (46.3)	21 (52.5)	3 (50.0)	
3-6 days N (%)	14 (40.0)	10 (24.4)	11 (27.5)	1 (16.7)	
7-10 days N (%)	3 (8.6)	3 (7.3)	4 (10.0)	0 (0.0)	
> 10 days N (%)	2 (5.7)	9 (22.0)	4 (10.0)	2 (33.3)	
Time on mechanical ventilation					0.576*
≤ 24 hours N (%)	27 (79.4)	24 (58.5)	25 (61.0)	4 (66.7)	
2-7 days N (%)	3 (8.8)	8 (19.5)	11 (26.8)	2 (33.3)	
> 8 days N (%)	4 (11.8)	9 (22.0)	5 (12.2)	0 (0.0)	
Hemodialysis N (%)	2 (5.7)	4 (9.5)	6 (14.6)	2 (28.6)	0.078*
Use of vancomycin N (%)	6 (17.1)	15 (35.7)	12 (29.3)	4 (57.1)	0.078*
Use of acyclovir N (%)	6 (17.1)	5 (11.9)	14 (34.1)	3 (42.9)	0.039*
Use of polymyxin E N (%)	3 (8.6)	5 (11.9)	4 (9.8)	1 (14.3)	0.825*
Use of amphotericin B N (%)	1 (2.9)	3 (7.1)	4 (10.0)	1 (14.3)	0.165*
Serum tacrolimus (ng/mL; median; range)	9.55 (2.0-12.6)	10.95 (4.3-30.0)	11.80 (6.4-32.4)	10.85 (8.8-12.3)	0.012**
Serum creatinine (mg/dL; median; range)	1.12 (0.64-2.86)	1.32 (0.7-2.19)	1.38 (0.69-2.56)	1.6 (0.7-6.28)	0.016**

\*Chi-squared test.

\*\*Kruskal-Wallis test.

Abbreviations. CKD: chronic kidney disease; LT: liver transplantation; ICU: Intensive Care Unit; Serum tacrolimus: first three months post-LT; Serum creatinine: first six months post-LT.

serum tacrolimus concentrations in the first three months after LT, the risk of developing stages 4-5 CKD at two years after LT was 1.69 times higher; and for each unit increase in the median of the serum creatinine levels during the first six months post-LT, the risk of developing CKD (stages 2 to 4-5) at two years post-LT was 5.2 times higher (Table 4).

The results of the univariate analysis comparing the pre-, peri- and post-LT factors and survival after LT are shown in Table 5. In the multivariate analysis, two variables showed independent association with death in two years post-LT: stay in the ICU after the LT surgery for three or more days, and median of serum creatinine levels in the first six months post-LT  $\geq 1.3$  mg/dL. These cut-off values were selected because they were the median values of the variables "length of stay in the ICU" and "median serum creatinine levels during the first six months post-LT".

The risk of death in two years post-LT of the patients who stayed in the ICU for three days or more was 15.45 times the risk of the patients who stayed fewer than three days in the ICU (OR = 15.45; 95% CI = 2.01-118.76; p = 0.008) as seen in Figure 1. The risk of death in two years after LT of the patients with median serum creatinine concentrations in the

first six months post-LT  $\geq 1.3$  mg/dL was 4.26 times the risk of those with a median  $< 1.3$  mg/dL (OR = 4.26; 95% CI = 1.33-13.62; p = 0,015), as shown in Figure 2.

## DISCUSSION

CKD is a common complication after LT and is associated with increased morbidity and mortality.<sup>2-6</sup> Data from the Scientific Registry of Transplant Recipients demonstrate that the incidence of stages 4-5 CKD after LT at one, three, and five years is 8%, 14%, and 18%, respectively, increasing to approximately 25% by 10 years after the transplant.<sup>2</sup> The risk of CKD and the need for long-term hemodialysis will increase further, given the trend toward increasing longevity in the overall population of recipients of LT.<sup>2,7</sup> Considering the high incidence of chronic renal failure and the high risk of death associated with it, identifying risk factors for CKD becomes important in order to adopt preventive measures early.

In the present study, the risk factors independently associated with CKD stage, at two years post-LT, were recipient's age at LT and male gender, and serum levels of

**Table 4 - Multivariate analysis for chronic kidney disease stage after liver transplantation**

Covariables	Comparisons between chronic kidney disease stages					
	1 versus (2 + 3 + 4/5)		1 + 2 versus 3 + 4/5		1 + 2 + 3 versus 4/5	
	Coef1	OR1 (95% CI)	Coef 2	OR2 (95% CI)	Coef 3	OR3 (95% CI)
Male	1.855	6.40 (2.42-16.9)	1.855	6.40 (2.42-16.9)	1.855	6.40 (2.42-16.9)
Age at LT			0.082	1.09 (1.02-1.15)	0.199	1.22 (1.11-1.33)
FK Median			0.131	1.14 (1.00-1.30)	0.524	1.69 (1.33-2.14)
Cr Median	1.653	5.20 (2.26-12.0)	1.653	5.20 (2.26-12.0)	1.653	5.20 (2.26-12.0)

Abbreviations. Coef: ordinal logistical regression coefficient; OR: odds ratio; CI: confidence interval; LT: liver transplantation; FK Median: median serum tacrolimus levels during the first three months post-LT; Cr Median: median serum creatinine levels during the first six months post-LT.



**Table 5** - Comparison between pre-, peri- and post-liver transplantation factors and the occurrence of death at two years after liver transplantation (univariate analysis)

Variables	Kaplan Meier P value	OR	Cox regression (95% CI)	P value
Age at LT >56 years	0.703	1.176	(0.510-2.714)	0.703
Gender	0.389			
Male		1.460	(0.613-3.484)	0.392
Cirrhosis etiology	0.385			
Viral		1.000		0.413
Cryptogenic and ethanolic		0.592	(0.221-1.591)	0.299
Autoimmune hepatopathy		0.684	(0.185-2.527)	0.569
Other etiologies		1.758	(0.476-6.495)	0.398
Pre-LT hemodialysis	0.004	6.528	(1.514-28.159)	0.012
MELD-score >18	0.910	1.049	(0.453-2.429)	0.910
INR > 1.61	0.319	0.651	(0.278-1.524)	0.323
Total pre-LT bilirubin (>2.7mg/dL)	0.995	0.998	(0.432-2.301)	0.995
Age of donor >33 years	0.124	1.953	(0.819-4.656)	0.131
Gender of donor	0.765			
Male		1.142	(0.479-2.721)	0.765
Peri-LT PRC transfusion	0.095			
No transfusion		0.219	(0.024-1.959)	0.174
1-2 units		0.462	(0.056-3.844)	0.475
3-5 units		1.003	(0.125-8.028)	0.997
6-10 units		0.447	(0.040-4.932)	0.511
> 10 units		1.000		0.141
Days stay at ICU	0.003			
≥ 3 days		9.475	(2.197-40.86)	0.000
Time on mechanical ventilation	0.000			
1 day		0.049	(0.013-0.178)	0.000
2-7 days		0.438	(0.166-1.154)	0.095
≥ 8 days		1.000		0.000
Post-LT hemodialysis	0.000	8.893	(3.714-21.290)	0.000
Post-LT PRC transfusion	0.000			
No transfusion		0.117	(0.032-0.433)	0.001
1-2 units		0.067	(0.008-0.529)	0.010
3-5 units		0.146	(0.031-0.678)	0.014
6-10 units		0.446	(0.158-1.255)	0.126
> 10 units		1.000		0.002
FK median > 10.6ng/mL	0.206	1.750	(0.726-4.226)	0.212
Cr median > 1.3mg/dL	0.040	2.876	(0.999-8.279)	0.050
Pre-LT CKD stage	0.144			
Stage 1		1.000		0.174
Stage 2		1.618	(0.494-5.301)	0.427
Stage 3		2.944	(1.047-8.277)	0.041
Stages 4-5		3.378	(0.681-16.761)	0.136

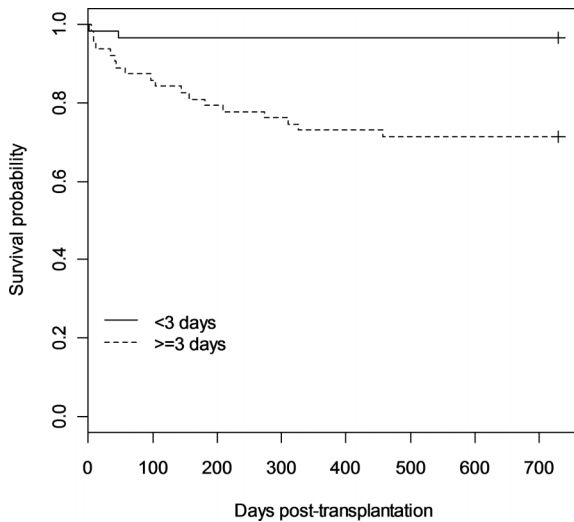
Abbreviations. OR: odds ratio; CI: confidence interval; LT: liver transplantation; MELD: Model for End-Stage Liver Disease; INR: International Normalized Ratio; PRC: packed red cells; ICU: Intensive Care Unit; FK Median: median serum tacrolimus levels during the first three months post-LT; Cr Median: median serum creatinine levels during the first six months post-LT; CKD: chronic kidney disease.

tacrolimus and creatinine in the first three and six months post-LT, respectively. Mortality in two years post-LT was associated with a stay in the ICU after the transplant surgery for a period equal or longer than three days, and with a median of serum creatinine levels in the first six months after LT ≥1.3 mg/dL.

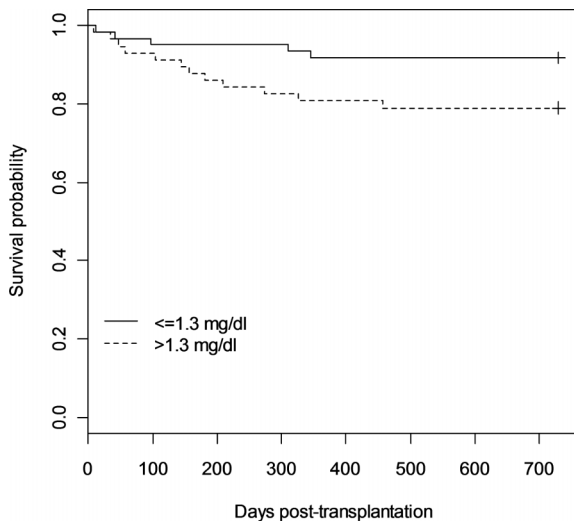
Advanced age of recipients has already been identified as a risk factor for the development of CKD after LT.<sup>2,19-21</sup> This finding was somewhat expected since renal function declines with increasing age, particularly over the age of 60 years.<sup>19</sup> On the other hand, the female gender has been identified by other authors as more commonly associated with CKD post-LT than males.<sup>2,20,22,23</sup> These non-modifiable factors should be used to help identify patients at higher risk of CKD in our population of post-LT patients.

Calcineurin inhibitors therapy has been implicated as a major cause of post-transplant renal dysfunction<sup>24,25</sup>, since

these agents may cause severe tubular atrophy, interstitial fibrosis, and focal hyalinosis of small renal arteries and arterioles.<sup>26-31</sup> In the present study, early tacrolimus levels were associated with CKD development after LT. Fisher et al.<sup>32</sup> showed that cyclosporine-A levels at one month and a cumulative dose of this agent at five years post-LT were associated with the development of severe CKD in LT recipients. In a Korean population of 431 patients submitted to LT, rapid progression of kidney disease was associated with tacrolimus levels above the target range at one, three and six months post-LT (target range: 8-13 ng/mL at one and three months, and 5-10 ng/mL at six months).<sup>16</sup> Calcineurin inhibitors levels minimizing protocols are expected to reduce CKD development in LT recipients, as well as creatinine levels in the first months after the transplant. This last condition was demonstrated to be associated with CKD and survival at two years post-LT in the present study. Most of



**Figure 1** - Two-year survival by length of stay in the Intensive Care Unit after liver transplantation.



**Figure 2** - Two-year survival by median of the serum creatinine levels during the first six months after liver transplantation.

our patients (81.8%) did not receive mycophenolate mofetil. The association of this agent with the calcineurin inhibitor could have allowed the prescription of lower doses of the last drug and, thus, lower serum tacrolimus levels.

Other studies have already demonstrated that early post-LT kidney dysfunction is associated with CKD development and mortality.<sup>32-34</sup> This finding suggests that all possible measures should be adopted early to spare renal function after LT, such as avoiding the use of nephrotoxic agents whenever possible, prevent hemodynamic insults to the kidneys and adopting calcineurin inhibitors levels minimizing protocols.

In the present study, we did not find any relationship between pre- and post-LT renal failure and neither between renal dysfunction before LT and mortality post-LT, in contrast to what was observed by others.<sup>2,3,7,11,23,24,28,35,36</sup> However, it should be pointed out that our results could

have been influenced by the small number of cases of CKD stages 4 and 5. On the other hand, renal dysfunction in the first six months post-LT was associated with CKD and mortality in the first two years after LT in this study. These findings emphasize the importance of early measures to preserve renal function after LT.

Because serum creatinine is a key component of the MELD-score and impaired pre-LT renal function is considered to be a poor prognostic indicator for both survival and post-LT CKD, a reduction in patient survival was expected after the implementation of this score for ordering patients in the waiting list for LT. However, different studies have shown that the MELD-score predicted neither the development of advanced CKD nor mortality more than one year after LT,<sup>5,12,33,37</sup> which is in agreement with our findings since, even in the univariate analysis, this variable was not associated with CKD stage or mortality, two years post-LT.

HCV infection has been identified as a risk factor for the development of CKD stages 4 and 5 after LT.<sup>3,7,33</sup> This association was not observed in this study, which may be explained, at least partially, by the small number of cases with the more advanced stages of CKD. In a recent study that compared patients transplanted for hepatitis C-related cirrhosis to other etiologies, with similar MELD-scores and mean pre-LT serum creatinine levels, the authors did not find any difference between the two groups regarding the estimated GFR, requirement for dialysis and death in three years post-LT.<sup>38</sup> The variables diabetes mellitus and hypertension were not identified as risk factors for CKD development after LT in our cohort. However, these conditions were associated with CKD post-LT in other studies.<sup>2,3,7,11,23,24,28</sup>

A stay in the ICU after the LT surgery for a period equal or longer than three days was associated with higher mortality in the first two years post-LT. This finding probably reflects the severity of patients who need intensive care for longer time. It is interesting to remark that higher mortality in patients requiring a longer stay in ICU occurred during the first year as shown in Figure 1.

This study was limited because it was retrospective, included a small numbers of patients with the more advanced stages of CKD, and GFR was calculated using a creatinine-based method, which often leads to overestimations of renal function in cirrhotic patients before LT, especially in those with poor nutritional status, low muscle mass, weight loss, and edema.<sup>31,33</sup>

## CONCLUSIONS

The judicious use of calcineurin inhibitors during the early post-LT period may help to preserve kidney function. Therefore, the administration of the lowest effective dose of these drugs and the association with other non-nephrotoxic immunosuppressive agents in the first months after LT may be strategies to spare renal function, avoiding late CKD and increasing long term survival, particularly in patients at high risk. Long term studies are necessary to confirm this hypothesis.

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Brandão VBA, Faria LC and Ferrari TCA: conceived and designed the study, and prepared the manuscript; Brandão VBA and Bicalho DM: collected data. Pereira FH performed statistical analysis. Brandão VBA, Faria LC, Bicalho

DM, Pereira FH and Ferrari TCA: analyzed and interpreted the data. Lima AS: provided intellectual content of critical importance to the study. All authors read and approved the final version of the manuscript.

## RESUMO

**OBJETIVO:** Investigar fatores de risco para o desenvolvimento de doença renal crônica e óbito dois anos após transplante hepático.

**MÉTODO:** Associações entre parâmetros clínicos e laboratoriais e o desenvolvimento de doença renal crônica e sobrevida dois anos pós-transplante hepático foram analisados em uma coorte de 148 pacientes adultos, com cirrose hepática, submetidos consecutivamente a transplante de fígado em centro de referência no Brasil.

**RESULTADOS:** A idade mediana ao transplante hepático foi 56 (limites, 20-73) anos, e 105 (70,9%) pacientes eram do gênero masculino. A prevalência de doença renal crônica dois anos pós-transplante hepático foi: estágio 1 ou ausência de doença renal, 27,5%; estágio 2, 33,8%; estágio 3, 34,6%; estágios 4-5, 4,7%. Quatro variáveis apresentaram associação independente com o estágio de doença renal crônica, dois anos pós-transplante hepático: (i) idade ao transplante hepático, (ii) gênero masculino, (iii) mediana dos níveis de tacrolimus nos primeiros três meses pós-transplante hepático e (iv) mediana das concentrações séricas de creatinina nos primeiros seis meses pós-transplante hepático. Duas variáveis apresentaram associação independente com óbito dois anos pós-transplante hepático: permanência em Unidade de Terapia Intensiva por três ou mais dias após a cirurgia de transplante, e mediana das concentrações séricas de creatinina nos primeiros seis e três meses após o transplante hepático igual ou superior a 1,3 mg/dL.

**CONCLUSÕES:** Administração da menor dose eficaz de tacrolimus e adoção de medidas para preservar a função renal são medidas importantes para se reduzir o risco de desenvolvimento de doença renal crônica, tardiamente, após o transplante de fígado, especialmente nos pacientes que apresentam fatores de risco.

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