Cardiovascular morbidity and mortality is increased post-liver transplantation even in recipients with no pre-existing risk factors

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Abbreviations: LT: liver transplantation, PTMS: post transplant metabolic syndrome, CVS: cardiovascular, MS: metabolic syndrome, BMI: body mass index, DM: diabetes mellitus, CAD:

coronary artery disease, DSE: dobutamine stress echo, IS: immunosuppression, RHC: right heart cathetirization

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Abstract:

Background/aims: Post-liver transplant (LT) metabolic syndrome (PTMS) and cardiovascular (CVS) mortality are becoming increasingly prevalent following sustained improvements in post-LT survival. We investigated the prevalence and predictors of PTMS and CVS complications in a cohort of consecutive LT recipients.

Methods: We reviewed prospectively collected data of patients (n=928) who underwent LT (1995-2013) and survived at least 1-year post-LT or died before that due to a major CVS complication.

Results: Median follow-up was 85 months (IQR=106). The prevalence of PTMS was 22.4% and it developed de novo in 183 recipients (19.7%). A total of 187 (20.2%) patients developed at least one CVS event post-LT within a median of 49 months (IQR=85). Overall mortality rate was 22.6% (n=210). Causes of death were CVS events (n=45, 21.4%), malignancies (21%), liver related deaths (20%) and infections (6.7%). Independent predictors of major CVS events were: documented CVS disease pre-LT **(Hazard Ratio (HR)=3.330; 95%CI=1.620-6.840)**, DM **(HR=1.120; 95%CI 1.030-1.220)**, **hypertension (HR=1.140; 95%CI 1.030-1.270), dyslipidaemia (HR=1.140; 95%CI 1.050-1.240) and** creatinine levels at 1 year **(HR=1.010; 95%CI=1.005-1.013)**. Among LT recipients without pre-LT CVS disease or MS components (n=432), 85 recipients developed ≥1 CVS events (19.7%) with independent predictors being DM **(HR=1.150; 95%CI=1.010-1.320)**, creatinine levels at 1 year **(HR=1.020; 95%CI=1.010-1.030)**, and **hypertension (HR=1.190; 95%CI=1.040-1.360).**

Conclusions: Post-LT patients are at increased risk of CVS morbidity even in the absence of pre-existing metabolic risk factors. Renal sparing immunosuppressive protocols might reduce CVS events post-LT.

Lay summary: Patients who have received a liver transplant are at high risk of developing cardiovascular events post transplantation, even in the absence of pre-existing metabolic risk

factors. Presence of diabetes **and hypertension**, **pre-existing cardiovascular disease** and higher creatinine levels at one year were predictive of cardiovascular events. The above data suggest that renal sparing immunosuppressive protocols might reduce such events post-liver transplantation.

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Introduction

Liver transplantation (LT) has altered the prognosis of patients with advanced liver disease, significantly improving the length and quality of life. Such patients usually died within months without LT (1), whereas LT recipients now have a 84% 1-year and 73% 5-year survival rates. (2-4) Survival following LT has been steadily improving over the last 2 decades, likely due to a greater surgical expertise, which reduced technical complications, a better selection of patients and improvements in the efficacy and tolerability of immunosuppressive therapy (IS), reducing graft loss from both acute and chronic rejection. (5) Indeed, graft loss due to rejection is becoming a relatively rare cause of morbidity and mortality post-LT. (6)

Although the main cause of death after the first year post LT is liver-related in up to 28% of patients, non-hepatic causes of death are increasing with malignancies (22%), cardiovascular disease (CVD) (11%), infection (9%), and renal failure (6%) being important causes. (7) The main causes of late mortality (>10 years) are hepatic in 40% and non-hepatic in 60% of patients, particularly de novo tumours and CVD. (8)

Metabolic syndrome (MS) is a common risk thread for each of these making the prevalence, prevention, and management of post-transplant MS (PTMS) and individual metabolic derangements of increasing interest and importance. MS currently affects 20-30% of the adult western world population. (9) It is an established risk factor for cardiovascular disease and its increasing prevalence correlates with an increasing incidence of CVD.

Liver transplant recipients have a high prevalence of risk factors for CVD, exceeding that of the general population, (10) and thus have a higher predicted risk of developing coronary heart disease (11-13). CVD hospitalizations post-LT have increased by 115% over the last decade (14). Recent studies from the US highlighted the high early cardiovascular morbidity and mortality following LT and have identified preoperative factors and liver disease aetiology as significant predictors (15, 16). However, there is a relevant paucity of data on the incidence and predictors of cardiovascular events following the perioperative period. Limitations of existing studies include inadequate sample

size, relevantly short follow-up, incomplete data on pre and post-LT metabolic comorbidities and lack of consensus regarding outcome definition (17).

The aim of our study was to investigate metabolic and cardiovascular complications in a well-defined cohort of consecutive LT recipients with longitudinal follow-up, identifying the predictors of PTMS and CV events after LT.

Patients and methods

We reviewed prospectively collected data of all patients (n=1175) who underwent LT between January 1995 and September 2013 at the Royal Free Hospital (London, UK). We included patients who survived longer than 1-year post-LT or died within the first year due to CV complications (defined as primary cause of death from arrhythmia, heart failure, myocardial infarction, primary cardiac arrest, and/or stroke). Patients who were re-transplanted were included if they met the inclusion criteria. We excluded patients who had multi-organ transplants. Each patient is represented only once in the analysis irrespective of the number of CV events he had.

Patients' medical records were reviewed from the time of their pre-transplant evaluation until their last post-transplant follow-up or death. We obtained routine demographic information, such as age at transplant, sex, race/ethnicity, aetiology of liver disease and data regarding pre-transplant clinical status including body mass index (BMI), smoking status, presence of diabetes mellitus (DM), hypertension and dyslipidaemia, and any cardiovascular morbidity pre-surgery. Data regarding the post-LT status included the development of metabolic derangements, their timing in relation to LT (as recorded for the first time in the medical records or based on clinical and laboratory findings) and their treatment. In addition, the primary immunosuppressive regimen used after LT, at 1 year, and at the end of follow up. Major CV events recorded after LT included acute coronary syndrome (defined as an ST-elevation myocardial infarction, a non-ST elevation myocardial infarction, or unstable angina), congestive heart failure, ischemic stroke, peripheral artery disease, stable angina and arrhythmias. The development of pulmonary embolism was not considered as a CV event. The post-LT cause of death was identified by the coding in the annual transplant audit forms that are submitted to NHS British Transplant. This study was part of an audit of post transplant mortality and ethical approval was therefore not required.

Metabolic Syndrome definition

MS was defined according to the 2001 guidelines of the National Cholesterol Education Program Adult Treatment Panel III (ATP III) and their revision in 2004 by the National Heart, Lung and Blood Institute and the American Heart Association. (18) Although BMI >30 kg/m^2 was not adopted by the ATP III in the definition of MS, being overweight clearly correlates with the metabolic risk factors.

Cardiovascular pre-transplant assessment

The cardiovascular pre-transplant assessment has been fairly consistent over the years and follows a multidisciplinary assessment protocol that includes full history and examination, a witnessed timed climb of 2 flights of stairs with pre- and post- O_2 saturations and heart rate recording, 12 lead ECG and trans-thoracic Echo (TTE) looking at left ventricular size and function, right ventricular size and function and evidence of pulmonary hypertension assessed from tricuspid regurgitation (TR). The main indication to perform a right heart catheterization is an estimated Right Ventricle Systolic Pressure >45mmHg (=peak TR Doppler jet >35mmHg) on TTE. RHC is also performed if there is a right bundle branch block or right axis deviation on ECG or evidence of enlarged pulmonary vessels on the postero-anterior chest-X Ray.

Dobutamine Stress Echo (DSE) is considered as a second-line investigation if there are any of the minor clinical predictors of coronary artery disease (CAD) such as history of stroke, systemic hypertension, history of smoking or obesity and/or a low functional capacity (as assessed by history and witnessed stair climb).

A DSE and/or further investigations are routinely performed to rule out CAD if there are intermediate clinical predictors such as angina with good exercise tolerance, previous myocardial infarction MI (revascularization > 5years), peripheral vascular disease and/or the presence of diabetes mellitus.

If any of the following major clinical predictors of CAD are present (angina with poor exercise tolerance, recent MI, unstable coronary syndromes, revascularization<5 years, impaired left

ventricular function, decompensated heart failure, significant symptomatic arrhythmias or second degree atrioventricular block, severe valvular heart disease), the patient is referred to a cardiologist for assessment and further invasive investigations.

Statistical Analysis

All data were analysed using the statistical package SPSS (version 22.0, IBM, New York, NY, USA) **or STATA (Release 12, College Station, TX: StataCorp LP).** Statistical analysis was performed using t-test, ANOVA, Mann-Whitney test or Kruskal-Wallis test for comparisons of continuous variables between or among groups, corrected chi-squared method or two-tailed Fisher's exact test for comparisons of qualitative data and Spearman's co-efficient for correlations of quantitative data, when appropriate. Multivariate analysis was performed using Cox regression (development of CV events/mortality) or binary logistic regression models (predictors of MS). **Time dependent covariate analysis was used for post-LT obesity, dyslipidaemia, diabetes and hypertension in the Cox regression models.** Only variables with a P value ≤0.10 at univariate analysis were entered in the multivariate models. Non-significant variables were excluded in a backward stepwise process. A two-tailed P-value <0.05 was considered statistically significant. The era of transplantation was included in all multivariate analyses to control for changes in practice.

Results

Among 1,175 patients who were transplanted during the study period, 928 fulfilled the inclusion criteria and were included in the analysis. The median follow up was 85 months (IQR=106). The baseline demographic characteristics are summarized in Table 1.

We performed a comparison regarding the rates of the constituents of MS before and after transplantation. As expected, the prevalence of obesity, hypertension, hypertriglyceridemia, DM and MS were significantly higher after LT **(Appendix).**

Metabolic complications after liver transplantation

The prevalence of the individual components of PTMS is shown in the appendix. Dyslipidaemia, obesity, diabetes mellitus and hypertension had a prevalence of 40.2% (n=373), 29% (n=269), 30.2% (n= 280) and 49.6% (n=460), respectively. The de novo development of MS components after LT was as follows: dyslipidaemia in 191 recipients (20.6%) after a median of 33 months (IQR=72), obesity in 147 recipients (15.8%), DM in 115 patients (12.4%) after a median of 32 months (IQR=90) and hypertension in 384 (41.4%) patients after a median time of 35 months (IQR=58). Regarding the treatment of the metabolic derangements during follow up, only 40.3% of the recipients with dyslipidaemia were treated, whilst the relevant treatment rates of DM and hypertension were 89.7% and 92.3% respectively.

The prevalence of PTMS was 22.4% (n=208) and it developed de novo in 183 recipients (19.7%) after a median of 48 months (IQR=84). The independent predictors for PTMS were: pre-LT DM (Odds Ratio (OR) = 6.602; 95% confidence interval (CI) 4.335-10.055), pre-LT obesity (OR=2.650; 95%CI=1.677-4.185), pre-LT hypertension (OR=1.996; 95%CI=1.123-3.548), alcohol aetiology (OR=2.843; 95%CI=1.698-4.759) and NASH/cryptogenic aetiology as compared with viral aetiology (OR=3.460; 95%CI=1.678-7.134), use of steroids at 1 year (OR= 5.612; 95%CI=1.973-15.962), and higher creatinine levels at 1 year after LT (OR=1.006; 95%CI=1.000-1012). The

univariate and multivariate binary logistic regression analysis is shown in table 2. Results did not significantly change when the presence of PTMS at 5 years was analysed as a binary outcome (data not shown).

Cardiovascular complications after liver transplantation

Two hundred-ten patients (22.6%) died after their first year post-LT in a median follow up of 56 months (IQR=80; range 0-194). Causes of death were CV events (n=45, 21.4%), malignancies (n=44, 21%), liver related deaths (n=42, 20%) and infections (n=14, 6.7%).

A total of 187 (20.2%) patients developed at least one CV event post-LT within a median of 49 months (IQR=85); of these patients, 51 (27.3%) developed more than one CV event during their follow-up post-LT. Acute coronary syndrome (ACS) occurred in 45 (24.1%), while 21 patients (11.2%) had a ST elevation myocardial infarct, 5 (2.7%) non-ST elevation myocardial infarct, 9 (4.8%) unstable angina and 10 (5.3%) had an unspecified diagnosis of ACS. Nineteen recipients (10.2%) were diagnosed with stable angina.

Congestive heart failure occurred in 47 recipients (25.1%) in a median follow up of 82 months (IQR=126). Ischaemic stroke occurred in 55 (29.4%) recipients in a median follow up of 40 months (IQR=82). Nineteen recipients (10.2%) were diagnosed with peripheral artery disease in a median follow up of 51 months (IQR=59).

Arrhythmias occurred in 66 patients (35.3%) after a median follow up of 62 months (IQR=124): among these patients, 34 (51.5%) were diagnosed with atrial fibrillation and 14 (21.2%) with episodes of paroxysmal supraventricular tachycardia.

Among the 187 recipients who had ≥1 CV events after LT, 27.3% had a major event within the first year post-LT, 28.3% between the 1st and the 5th year and the majority (44%) after the 5th year post-LT.

In the multivariate cox regression analysis, independent predictors for the development of a major CV event were: documented CVS disease pre-LT **(HR=3.330; 95%CI=1.620-6.840)**, DM

(HR=1.120; 95%CI 1.030-1.220), **hypertension (HR=1.140; 95%CI 1.030-1.270), dyslipidaemia (HR=1.140; 95%CI 1.050-1.240) and** creatinine levels at 1 year **(HR=1.010; 95%CI=1.005-1.013).** The results of the univariate and multivariate cox regression analysis are shown in table 3. Independent predictors of CV events at more than one year following LT were: documented CVD pre-LT (HR=2.9**66**;95%CI=1.1**99**-7.3**33**), higher creatinine levels at 1 year after surgery (HR=1.008, 95%CI=1.003-1.013), **post-LT diabetes (HR=1.773, 95%CI=1.232-2.552**), **post-LT dyslipidaemia (HR=1.867, 95%CI=1.291-2.669**) **and post-LT hypertension (HR=1.636, 95%CI=1.065-2.514**). Independent predictors of CV events at more than five years following LT were: **post-LT diabetes (HR=1.738, 95%CI=1.092-2.767**), post-LT hypertension (HR=2.4**71**, 95%CI=1.33**4**-4.5**80**) and higher creatinine levels at 1 year after surgery (HR=1.012, 95%CI=1.005-1.018). Documented CVD pre-LT was not associated with such events.

Cardiovascular complications in recipients with no pre-LT CV disease or MS components

In the subgroup of patients with no pre-existing CVD and none of the MS components pre-LT (n=432), 85 recipients developed ≥1 CV event (19.7%) as follows: 4.6% within 12 months from LT, 5.1% between 1 and 5 years after LT and 10% after the 5th year post-LT. Table 4 shows a comparison of such patients with and without CV events.

In these patients, independent predictors for the development of CV events were de novo post-LT DM **(HR=1.150; 95%CI=1.010-1.320)**, creatinine levels at 1 year **(HR=1.020; 95%CI=1.010-1.030)**, and **de novo post-LT hypertension (HR=1.190; 95%CI=1.040-1.360).**

These results are summarized in Table 5.

Discussion

In this study, we have confirmed the high incidence of the metabolic syndrome and CV morbidity and mortality post-LT. More importantly, we have shown that the incidence of post-LT CV events is alarmingly high even in recipients without pre-existing CVD or metabolic risk factors. Finally we have identified that pre-LT CVD, the development of post LT DM, **post LT hypertension** and an elevated creatinine at 1-year are predictive of post LT CVD.

In our unselected cohort, the prevalence of PTMS was 19.7%, while 20.2% of the patients developed at least one CV event. This is more than double than the prevalence of CVD in the general population in the United Kingdom, which is 8.2%. (19) Moreover, CVS was the leading cause of mortality in the post-LT population irrespective of the transplantation era. In the era of successful antiviral treatment for recurrent HCV infection, the rate of graft failure as a cause for post LT mortality is projected to decrease and hence the proportion of post-LT deaths attributable to CVD is likely to increase.

We have demonstrated that independent predictors for the development of CV events were post-LT DM and hypertension and higher creatinine levels at 1-year post LT. Although not all patients were screened with stress echo pre-LT, our standard assessment mirrors clinical practice in most transplant centres during the era we study. Therefore, we cannot answer the question on whether the increased incidence of CV events found in our study population was due to the presence of a subclinical asymptomatic CV disease pre-LT or to an accelerated atherogenesis and CVD following LT. However, we consider the latter hypothesis to be more likely, as we observed high rates of CV events in patients with no pre-existing metabolic risk factors.

Our analysis has revealed potentially modifiable factors that predict the occurrence of CV events that merit further attention. In particular, creatinine levels increase CV morbidity by 8-16% (whether you consider the whole population or only recipients without pre-existing CV and metabolic derangements). Renal impairment is an established risk factor for CV morbidity and mortality in non-cirrhotic individuals (20, 21) and in the LT setting the most studied renal

impairment, associated with post-LT mortality, is preoperative renal dysfunction. (22, 23) Our results therefore suggest that renal dysfunction is one of the predictors mostly associated with accelerated CVD in the LT recipient. Therefore early CNI minimization protocols should be universally advised among the LT population (6). Indeed we have recently shown that reduced tacrolimus trough concentrations (<10 ng/ml) within the first month after LT were associated with halved renal impairment rates at 1 year with no significant influence on acute rejection rates. (24) Moreover, tacrolimus levels between 7 and 10 ng/ml early after LT may be associated with longer graft survival. (6)

Dyslipidaemia was a risk factor for the development of post-LT CVD. **In our series, only 40% of patients with dyslipidaemia received statins and this could have influenced outcomes. We could not assess the effect of statin treatment due to collinearity with dyslipidaemia and cardiovascular events.** The undertreatment of dyslipidaemia is likely due to the fact that the liver transplant recipient is almost exclusively under the care of the liver specialist who seems to be, as per forma mentis, more focused on the liver related issues of the patient, overlooking the temporary or permanent metabolic complications that often occur in this special population.

The AASLD guidelines recommend the annual measurement of blood lipids for healthy LT recipients. An elevated LDL cholesterol level >100 mg/dL, with or without hypertriglyceridemia, requires therapy. If dietary interventions and lifestyle modification are not efficacious, the guidelines suggest to start statin treatment, with the addition of ezetimibe, if necessary (25). Although AASLD guidelines do not mention a specific safer or more efficacious statin in the LT patient, pravastatin is the most studied and used statin in solid organ-transplant recipients it is not metabolized via the P450 enzyme system and has no interaction with immunosuppressant medications like other statins. The EASL guidelines recommend the preferential use of hydrophilic statins such as pravastatin or fluvastatin due to the reasons above (26). The NICE guidelines suggest the use of a risk assessment tool (QRISK2) to identify and stratify people with high risk for developing CVD and point out that standard CVD risk scores underestimate risk in people who

have additional risk because of underlying medical conditions or treatment (autoimmune diseases, corticosteroids or immunosuppressant drugs). (27).

Therefore, the intriguing question is whether statins should be prescribed in all LT recipients irrespective of the presence of dyslipidaemia or increased cardiovascular risk scores. Indeed, it has been long established that statins have a pleiotropic effect over and above their lipid lowering properties. This could be even more pronounced in a cohort with accelerated atherogenesis and CVD such as post-LT patients. Such questions can only be answered in a context of a randomized controlled trial.

Our study is the largest report with individual patient data published to date, with also the longest available follow-up. The main overall causes of death were CV events (21.4%), followed by malignancies (21%), liver related deaths (20%) and infections (6.7%). These results align with the current literature that places CV events as well as other non-liver related deaths among the leading causes of mortality. Most of the literature has focused on short-term patient and graft survival while only few studies reported medium and long-term survival. (8, 28-29) Indeed, the largest study to date with 755 patients reported only 40 months of median follow-up, which is less than half of what we report in this paper (30).

A recent paper from VanWagner et al, which focused on early postoperative CVD mortality, provided the first multicenter prognostic model for the prediction of early post-LT CVD death, identified by the authors as the most common cause of early post-LT mortality. This study has been performed on 54,697 LT recipients, but it is an analysis of Organ Procurement and Transplantation Network (OPTN) database between February 2002 and December 2012 in order to assess the prevalence and predictors of early (30-day) CVD mortality (16). The same author, based on a population of 242 recipients, compared the incidence of CV events between patients transplanted for NASH and ETOH-induced cirrhosis. NASH patients were four times more likely to have a CV event <1 year after LT, compared to ETOH patients, even after controlling for various confounding factors. The majority (70%) of events occurred in the perioperative period, and the occurrence of a

CV event was associated with a 50% overall mortality. In our study, we focused on overall rather than early CV morbidity, and although NASH aetiology was not an independent predictor for CVD, we found multiple metabolic derangements associated with NAFLD as independent predictors (15). We also showed that the majority of CV events occur after the 5th year post-LT, focusing on the accelerated CV burden secondary to different factors probably linked to LT. This is of particular concern, as although post LT outcomes have improved over last 2 decades, this improvement has all been in the first 6-12 months and very little impact has been made on mortality rates beyond 12-months.

Our results would support the management of such patients in multidisciplinary clinics following the first year post-LT, in order to recognize and treat modifiable CV risk factors in a timely manner. Alternatively a CV comorbidity checklist including calculation of a risk assessment tool would be useful in reminding or alerting the transplant physicians on this particular aspect of management. One year mortality is currently the most commonly used quality measure for LT programs, however these results would argue for adding quality measures for longer term outcomes, such as statin treatment based on LDL levels or the QRISK2 score.

The strength of our study is that our clinical and data information have been collected across the clinical summaries and discharge letters of every individual recipient included in our study, from baseline throughout all the length of follow up (up to 234 months after LT).

Our study has some limitations. This is a single-center, retrospective study. We relied on chart review, therefore metabolic comorbidities, relevant medication or indeed some CV events could have been underreported. Moreover, the pre-LT cardiovascular workup has changed with time, which could also have introduced bias in the incidence of outcomes. Although limited by its retrospective design, it clearly points to the clinical significance of the metabolic derangements in the liver recipient, which lead to an increase in CV morbidity and mortality with a higher rate of CV complications than the general adult British population.

In summary, our study demonstrates that LT recipients, irrespective of the presence of metabolic risk factors or preexisting CVD, have an increased risk to develop CVD, likely due to an accelerated atherogenic process post-LT. Among risk factors and independent predictors of this post-LT CVD, renal dysfunction and **metabolic syndrome parameters** impact heavily on CV morbidity. A common thread could be a CNI minimization protocol early post-LT, given the known effect these drugs have both on metabolic derangements and renal dysfunction. Moreover, the importance of recognizing and treating the risk factors we identified is crucial to avoid CV morbidity and mortality. Transplant physicians should be better educated to promptly recognize potentially modifiable metabolic abnormalities and intervene early.

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Table 1. Baseline characteristics of liver transplant recipients (n=928)

Characteristics	**Value (N = 928)**
Age (years), median (IQR)	50 (14)
Male sex (n, %)	586 (63.1)
Ethnicity (n, %)	
Euro-Caucasoid	728 (78.4)
Asian-Caucasoid	127 (13.7)
Afro-Carribean	52 (5.6)
Indication for transplantation (n, %)	
Viral hepatitis*	307 (33.1)
Alcoholic liver disease	206 (22.2)
Primary biliary cholangitis	99 (10)
Primary sclerosing cholangitis	82 (9)
Acute liver failure	81 (8)
NASH+Cryptogenic cirrhosis	59 (6.4)
Rare metabolic liver diseases	40 (4)
Autoimmune hepatitis	27 (3)
Miscellaneous	27 (3)
Concomitant HCC	155 (16.7)
Follow up (months), median (IQR)	85 (106)
Initial immunosuppression (n, %)	
TAC based	758 (81.7)
CSA based	147 (15.8)
Steroids	635 (68.4)
MMF	246 (26.5)

AZA	362 (39)
Immunosuppression regimen (30 days post-LT) n, (%)	
Triple IS (CNI+MMF/AZA+steroids)	523 (56.4)
Double IS (CNI+steroids)	107 (11.5)
Single IS (CNI)	198 (21.3)
Double IS- no steroids (CNI+MMF/AZA)	80 (8.6)
Smoking (n, %)	143 (15.4)
Era OLT (n, %)	
< 2001	286 (30.8)
≥ 2001	642 (69.2)
Documented CVD pre-LT (n, %)	25 (2.7)

*viral included diagnosis of HCV and HBV cirrhosis.

All cases with HCC are under the primary aetiology of the chronic liver disease.

Abbreviations: NASH non alcoholic steatohepatitis; IQR interquartile range; TAC Tacrolimus; CSA Cyclosporine A; MMF mycophenolate mofetil; AZA azathioprine; IS immunosuppression; CNI calcineurin inhibitors, LT liver transplantation; CVD cardiovascular disease;

Table 2. Univariate and multivariate binary regression analysis for development of post transplant metabolic syndrome

Factors	PTMS (N=208)	No PTMS (N=720)	Univariate analysis P value	Multivariate analysis	
				P value	OR (95% CI)
Male sex, n (%)	156 (75)	430 (59.7)	<0.001	ns	
Age, median (IQR)	51 (14)	50 (14)	0.58	ns	
Donor BMI, median (IQR)	25 (4.8)	24.4 (4.1)	0.004	ns	
Aetiology of liver disease - Viral, n (%) - Alcohol, n (%) - NASH+Cryptogenic, n (%)	 71 (34.1) 69 (33.2) 40 (19.2)	 236 (32.8) 137 (19) 31 (4.3)	 0.304 <0.001 <0.001	 ns <0.001 0.001	 2.843 (1.698-4.759) 3.460 (1.678-7.134)
Pre-LT diabetes, n (%)	99 (47.6)	76 (10.6)	<0.001	<0.001	6.602 (4.335-10.055)
Pre-LT obesity, n (%)	62 (29.8)	82 (11.4)	<0.001	<0.001	2.650 (1.677-4.185)
Pre-LT hypertension, n	43 (20.7)	47 (6.5)	<0.001	0.019	1.996 (1.123-

(%)					3.548)
Pre-LT metabolic syndrome, n (%)	25 (12)	13 (1.8)	<0.001	ns	
Use of steroids (at 1 year), n (%)	7 (3.4)	88 (12.2)	<0.001	0.001	5.612 (1.973-15.962)
MMF, n (%)	74 (35.6)	172 (23.9)	0.004	ns	
Creatinine levels (at 1 year) μmol/L, median (IQR)	107 (29)	100 (28)	<0.001	0.038	1.006 (1.000-1012)

Abbreviations: PTMS (post-transplant metabolic syndrome), OR (odds ratio), CI (confidence interval), IQR (interquartile range), BMI (body mass index), NASH (non alcoholic steatohepatitis), LT (liver transplantation), MMF (mycophenolate mofetile), ns (non significant)

Table 3. Univariate and multivariate Cox regression analysis of predictors of cardiovascular events post liver transplantation.

Factors	CV event (N=187)	No CV event (N=741)	Univariate analysis P value	Multivariate analysis	
				P value	HR (95% CI)
Main effect					
Male sex, n (%)	135 (72.2)	450 (60.8)	0.001	0.08	1.38 (0.96-1.97)
Age, median (IQR)	50 (13)	50 (15)	0.292	ns	
Aetiology of liver disease - viral - alcohol - NASH/cryptogenic	 67 (35.8) 56 (29.9) 12 (6.4)	 239 (32.3) 150 (20.3) 47 (6.4)	 0.010 <0.001 0.090	ns	
CV diseases pre-LT	9 (4.8)	16 (2.2)	0.002	0.001	3.33 (1.62-6.84)
Use of steroids at 1 yr, n (%)	15 (8)	80 (10.8)	0.465	ns	
Creatinine levels at 1 yr μmol/L, median (IQR)	112 (32)	99 (28)	<0.001	<0.001	1.01 (1.005-1.013)
Era LT*, n (%) - LT < 2001 - LT ≥ 2001	 95 (50.8) 92 (49.2)	 191 (25.8) 549 (74.2)	 0.11	ns	

Time-dependent covariates					
Post-LT dyslipidaemia, n (%)	107 (57.2)	266 (35.9)	<0.001	0.003	1.14 (1.05-1.24)
Post-LT diabetes mellitus, n (%)	86 (46)	195 (26.4)	<0.001	0.009	1.12 (1.03-1.22)
Post-LT hypertension, n (%)	123 (65.8)	337 (45.5)	<0.001	0.01	1.14 (1.03-1.27)
Post-LT obesity, n (%)	69 (36.9)	200 (27)	0.001	ns	

Abbreviations: CV (cardiovascular), HR (Hazard ratio), CI (Confidence interval), IQR (interquartile range), NASH (nonalcoholic steatohepatitis), LT (liver transplant), ns (non significant)

*division between all LT performed before 2001 and after 2001.

Table 4. Characteristics of liver transplant recipients without pre-LT documented cardiovascular disease or components of the metabolic (n= 432) according to the development or not of post-LT CV events.

Factor	**Post-LT CV events (n= 85)**	**No Post-LT CV events (n= 347)**	**P value**
Male sex, n (%)	61 (71.8)	197 (56.8)	0.012
NASH aetiology, n(%)	2 (2.4)	14 (4)	ns
HCV aetiology, n (%)	27 (31.8)	80 (23.1)	ns
ETOH aetiology, n (%)	26 (30.6)	71 (20.5)	0.048
CSA based regimen IS, n (%)	23 (27.1)	67 (19.3)	ns
TAC based regimen IS, n (%)	60 (70.6)	276 (79.5)	ns
post-LT dyslipidemia, n (%)	45 (53)	120 (34.6)	0.002
Treatment of dyslipidemia, n (%)	21 (24.7)	30 (8.6)	<0.001
post-LT diabetes, n (%)	24 (28.2)	41 (11.8)	<0.001

post-LT hypertension, n (%)	54 (63.5)	135 (38.9)	<0.001
post-LT obesity, n (%)	22 (25.9)	51 (14.7)	0.004
PTMS, n (%)	25 (29.4)	51 (14.7)	<0.001
Cr at 1 yr post-LT, median (IQR)	115 (30)	97 (27)	<0.001
HDL at 1 yr post-LT, median (IQR)	1.3 (0.6)	1.4 (0.6)	ns
TGL at 1 yr post-LT, median (IQR)	1.55 (1.0)	1.3 (0.9)	ns

Abbreviations: CSA (cyclopsorin A), TAC (Tacrolimus), NASH (non-alcoholic steatohepatitis), HCV (hepatitis C virus), ETOH (ethanol), IS (immunosuppression), LT (liver transplantation), PTMS (post-trasnplant metabolic syndrome), Cr (creatinine), yr (year), HDL (High density lipoprotein cholesterol), TGL (triglycerides) , CV (cardiovascular), IQR (interquartile range)

Table 5. Predictors of CV events post-LT in univariate and multivariate cox regression analysis in analysis in recipients with no pre-LT cardiovascular disease or any of the individual components of the metabolic syndrome (n=432)

Factors	CV event (n= 85)	No CV event (n=347)	Univariate analysis P value	Multivariate analysis	
				P value	HR (95% CI)
Main effects					
Male sex, n (%)	61 (71.8)	197 (56.8)	0.02	0.08	1.64 (0.95-2.85)
HCV aetiology, n (%)	27 (31.8)	80 (23.1)	0.017	ns	
Use of steroids at 1yr after LT, n (%)	2 (2.4)	32 (9.2)	0.307	ns	
BMI at 1 yr after LT, median (IQR)	25 (4.9)	23.9 (5)	0.004	ns	
Creatinine levels at 1 yr μmol/L, median (IQR)	115 (30)	97 (27)	<0.001	<0.001	1.02 (1.01-1.03)
Time varying covariates					
Post-LT dyslipidaemia				0.15	1.09 (0.9-1.22)
Post-LT diabetes mellitus, n (%)	24 (28.2)	41 (11.8%)	<0.001	0.03	1.15 (1.01-1.32)

Post-LT hypertension, n (%)	54 (63.5)	135 (38.9)	0.001	0.01	1.19 (1.04-1.36)

Abbreviations: CV (cardiovascular), HR (Hazard ratio), CI (Confidence interval), IQR (interquartile range), LT (liver transplant), yr (year), BMI (body mass index), ns (non significant)