Diploma Thesis

Single-center experience with low-dose anti-thymocyte globulin induction therapy in patients undergoing liver transplantation

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Graz, am 10.11.2019 *Kerstin Führlinger eh*

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Abbreviations

AAT	Alpha-1 Antitrypsin deficiency
ALG	Anti Lymphocyte Globulin
ARDS	Acute Respiratory Distress Syndrome
ATG	Anti-Thymocyte Globulin, Thymoglobulin©
BMI	Body Mass index
CMV	Cytomegalo Virus
CMV-PCR	Cytomegalo Virus – Polymerase chain reaction
CNI	Calcineurin Inhibitor
CyA	Cyclosporine A
D/R	Donor/ Recipient CMV status
eGFR	estimated Glomerular Filtration Rate
HCC	Hepatocellular Carcinoma
HCV	Hepatitis C Virus
INR	International Normalized Ratio
IS	Immunosuppression
LT	Liver transplantation
MMF	Mycophenolate Mofetil
MELD	Model of End-Stage liver Disease
NHL	Non-Hodgkin Lymphoma
PCR	Polymerase-chain reaction
PELD	Pediatric End-Stage Liver Disease
PTLD	Post-transplant lymphoproliferative disorder
TAC	Tacrolimus
Tx	Transplantation
UNOS	United Network for Organ Sharing
KDIGO	Kidney Disease Improving Global Outcome

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Zusammenfassung

Einleitung: Thymoglobulin© (ATG) als niedrig dosierte Induktionstherapie nach Lebertransplantation wird kontrovers betrachtet und die Datenlage dazu ist rar. Ziel dieser Studie ist es potenzielle Risikofaktoren zu identifizieren und ein Patientenkollektiv zu definieren, welches von Thymoglobulin© profitieren könnte.

Material und Methoden: Untersucht wurden alle PatientInnen, welche zwischen 01.01.2007 und 31.12.2018 (n=217) an der Klinischen Abteilung für Transplantationschirurgie der Universitätsklinik für Chirurgie, Graz eine Erst-Lebertransplantation erhielten. PatientInnen mit fehlenden Daten oder/und additiver Nierentransplantation (n=6) wurden von der Studie ausgeschlossen. In der dadurch entstandenen 1. Studienkohorte (n=211) wurde das Patientenüberleben, das Transplantatüberleben, das abstoßungsfreie Transplantatüberleben, das malignomfreie Überleben, sowie die Rate an CMV-PCR positiven Episoden, die Entstehung von bronchopulmonalen und/ oder Harnwegsinfektionen innerhalb des ersten Monats nach der Transplantation mittels multivariabler Cox Regression sowie logistischer Regression untersucht. Nach weiterem Ausschluss wegen Mangel an Daten (n=31) zur Nierenfunktion 6 Monate postoperativ wurde in einer 2. Studienkohorte der Einfluss von Thymoglobulin© auf eine KDIGO-Stadien Verbesserung 6 Monate nach der Transplantation mittels multivariabler logistischer Regressionsanalyse durchgeführt. Durch ROC-Kurven Analysen (AUROC) wurden potenziell prognostische Modelle erstellt.

Ergebnisse: Die Induktionstherapie mit Thymoglobulin© zeigte keinen signifikanten Einfluss auf das Patientenüberleben, das Transplantatüberleben, das abstoßungsfreie Intervall nach der Transplantation sowie das malignomfreie Überleben. Weiters hatte Thymoglobulin© keinen signifikanten Einfluss auf nicht-virale Infektionen innerhalb des ersten Monats postoperativ. Es zeigte sich, dass Thymoglobulin© ein unabhängiger, signifikanter Risikofaktor für die Entstehung von CMV-PCR positiven Episoden innerhalb des ersten Monats und der ersten 6 Monate nach Transplantation ist. Diese CMV-PCR positiven Episoden innerhalb des ersten Monats zeigten einen signifikanten Einfluss auf das Patientenüberleben. Thymoglobulin© und Empfänger mit einem BMI > 25kg/m² zeigten einen unabhängigen, signifikanten Einfluss auf die Nierenfunktionsverbesserung

angegeben in KDIGO Stadien innerhalb von 6 Monaten nach Transplantation. Daraus ergab sich ein potenziell nutzbares prädiktives Modell (AUROC = 0.916).

Diskussion: PatientInnen mit prätransplant höheren KDIGO-Stadien und einem BMI ≤ 25 kg/m² profitieren am Meisten von einer Induktionstherapie mit Thymoglobulin© im Sinne einer Nierenfunktionsverbesserung innerhalb der ersten 6 Monate nach der Transplantation. PatientInnen, welche eine Induktionstherapie durch Thymoglobulin© erhalten, sollten eine CMV-Prophylaxe bekommen, da die Rate an frühen CMV-PCR positiven Episoden nach der Transplantation durch die Gabe von Thymoglobulin© signifikant erhöht wird.

Abstract

Introduction: Thymoglobulin© induction therapy after liver transplantation is considered highly controversial. Studies about Thymoglobulin© induction therapy are rare. The aim of this study is to identify potential risk factors and patients who may profit from Thymoglobulin© induction therapy.

Patients and Methods: Patients receiving primary liver transplants at the Clinical Department of Transplantation surgery of the University Hospital for Surgery, Graz between 01.01.2007 and 31.12.2018 (n=217) were included and patients with lack of data and/or additional kidney transplantation were excluded from analysis (n=6) defining Study Cohort 1 (n=211). Study Cohort 1 was used to determine independent influences of Thymoglobulin© induction on patient survival, graft survival, rejection-free graft survival, cancer-free survival as well as CMV infections, bronchopulmonary and/ or urinary tract infections within the first month deploying multivariable Cox regression and logistic regression modelling, respectively.

Further exclusion of patients with lack of follow-up data on kidney function 6 months after transplantation (n=31) defined Study Cohort 2 (n=180) which was used to determine the influence of ATG on KDIGO-stage improvement 6 months after transplantation using multivariable logistic regression analysis. ROC-curve analysis (AUROC) was used to evaluate potential prognostic models.

Results: Induction therapy with Thymoglobulin© did not influence patient survival, graft survival, rejection-free graft survival, and cancer-free survival significantly. Furthermore, Thymoglobulin© had no significant influence on non-viral infections within the first month after transplantation. Thymoglobulin© was revealed as an independently significant risk factor for CMV-PCR positivity within the first month and within the first 6 months after transplantation. Furthermore, CMV-PCR positive events within the first month after transplantation demonstrated a significant influence on patient survival. Thymoglobulin© and a recipients BMI > 25 kg/m^2 independently and significantly influenced the KDIGO stage of kidney function at 6 months after transplantation suggesting a potentially useful predictive model (AUROC = 0,916).

Discussion: Patients with compromised pre-transplant renal function with higher KDIGO stages and a BMI ≤ 25 kg/m^2 profit most from Thymoglobulin© induction therapy resulting in the improvement of kidney function 6 months after

transplantation. Patients with Thymoglobulin© induction therapy should receive CMV-prophylaxis due to the increased associated risk for early CMV-PCR positive events.

1 Introduction

Liver transplantation (LT) is state of the art for end stage liver disease, including chronic liver disease, acute liver failure or hepatocellular carcinoma (HCC) within Mazzaferro criteria (1). Since the introduction of liver transplant surgery the survival rate of patients following acute liver failure has increased massively, from 10-20% to 75-80% after one year (2).

The usage of immunosuppressive drugs after LT is the most important factor for preventing graft rejections and consecutive graft loss after transplantation (3). This is noticeable in an improvement of patient survival and graft survival after the introduction of immunosuppressive agents such as Calcineurin Inhibitors (CNI's) which were first introduced after kidney transplantation (4,5).

Thus, while patient survival is constantly increasing, long term outcome and long-term side effects of immunosuppression are nowadays of increasing significance. All patients with expected survival rates less than one year without transplantation and with a markedly predicted improvement of quality of life after transplantation should be evaluated for transplantation (2).

To determine the importance of liver transplantation and the severity of liver disease the Model for End stage Liver Disease (MELD) was introduced to list patients according to priority. This system was established in 2002 in the United States. It is based on objectively measured parameters such as bilirubin, creatinine and international normalized ratio (INR) (6). The MELD-score is a model for predicting 3-month mortality in patients with end-stage liver diseases. All patients with a score ≥15 are recommended for being listed for transplantation. A prediction of mortality following liver transplantation is only possible with a MELD higher than 35 (7). For some indications the MELD system cannot be considered as an adequate scoring tool. In those cases, other parameters have to be considered for evaluating and prioritizing patients. These exceptional indications contain pulmonary complications of cirrhosis, amyloidosis, primary hyperoxaluria, hepatic encephalopathy and patients with hepatocellular carcinoma. Patients with hepatocellular carcinoma need to receive priority on the waiting list as long as they are within the Mazzaferro criteria. To increase the rating score additional, so called match-MELD points, can be added according to tumor size, number of nodules, waiting time, response to downstaging

therapies and tumor marker level of alpha fetoprotein which is specifically depending on national allocation rules (2,8).

Induction therapy after liver transplantation was introduced to decrease the side effects of other nephrotoxic immunosuppressive agents including CNI's. The idea was to start with lower doses of CNIs and with later onset after transplant. To bridge the time between transplantation and initiation of immunosuppression with nephrotoxic drugs, that require blood level monitoring, new medication needed to be developed, without setting patients at risk for early graft loss or acute rejection episodes.

As a result, in addition to basic immunosuppression after LT, an antibody-based induction therapy has been established. The goal of this induction therapy was to decrease rejection episodes after transplantation while being able to delay the initiation of immunosuppression with CNI's, antimetabolites and Steroids (9–11).

The first use of polyclonal antithymocyte globulin (ATG) which is a rabbit-derived antibody, after organ transplantation was first mentioned in the 1960's.(12). The main indications were the prevention and treatment of early graft rejections after transplantation (13–17).

One of the main side effects caused by immunosuppressive agents like CNI's is the impairment of renal function especially during the first days after transplantation. In many cases kidney function is compromised due to hepatorenal syndrome emerging from end-stage liver disease. Furthermore, surgery and the administered medication during and after an operation affects renal function too (18–23).

1.1 ATG/ Thymoglobulin©

Thymoglobulin© is a T-cell depleting polyclonal antithymocyte antibody. The production of Thymoglobulin© is based on the immunization of rabbits with human thymocytes. Thymoglobulin© does not focus on a special T-cell line, and according to its wide spectrum affecting T-cells and other leukocytes it is also used in the therapy of graft versus host diseases in allogeneic hematopoietic stem cell transplantation. The depletion of T-cells is based on various mechanisms like complement-dependent and cell-mediated functions as well as induced apoptosis (24). In this study all patients received as ATG medication Thymoglobulin©

The initiation of induction therapy with ATG is seen highly controversial in liver transplantation (11). The main worry about the delayed initiation of basic immunosuppression is not to set patients at risk for experiencing early acute rejection episodes leading to increased graft failure and higher mortality (25).

A trend towards a higher incidence of bacterial and fungal infections in patients with ATG induction therapy was apparent in a study investigating ATG induction administered intraoperatively during liver transplantation. Due to small case numbers the results of this study have to be seen critically (26).
Based on Thymoglobulin's© unspecific effects, many different cell lines are affected. The most important side effects mentioned in a paper referring on a retrospective analysis of intraoperative administration of ATG (26) include cytokine release syndromes and hemolytic anemia which may occur during the intraoperative administration of ATG. These effects lead to higher intraoperative instability with an increased need to transfuse blood products and fresh frozen plasma (26). The subsequent hypotension and hyperthermia which were mainly detected in the ATG-group needed to be treated by crystalloids and catecholamine infusions (26).
Because Thymoglobulin© also causes platelet and leukocyte side effects it additionally leads to thrombocytopenia and neutropenia what increases the risks for fever, sepsis and serum sickness (9,13,27).
In a small trial the effect of ATG induction therapy versus no ATG induction therapy in combination with Steroids and Tacrolimus showed a decrease in alanine aminotransferase one day after liver transplantation in the ATG group. A histologic improvement was also seen in the ATG induction group (28).
High dose ATG induction therapy has been suspected to induce post-transplantation lymphoproliferative diseases (PTLD). According to a more recent article which analyzed PTLD development after low dose ATG induction therapy it was shown to be unlikely that Thymoglobulin© increases the risk for PTLD significantly (29). Nevertheless, it is currently still difficult to explore the exact impact of low dose ATG induction therapy on the post-transplant PTLD risk due to the lack of long-term follow-up data in prospective observational trials and the fact that there is always a multidrug immunosuppression administered (29).
Due to the above shown potential side effects Thymoglobulin© induction therapy after liver transplantation is currently regarded as highly controversial.

1.2 Maintenance immunosuppression

1.2.1 Steroids

Steroids are commonly used early after transplantation in high dose regimens with the goal to avoid early graft rejection. The side effects of Steroids are manifold containing steroid-induced diabetes, hypertension, hyperlipidemia and consecutive cardiovascular events, increased risk of infections as well as bone loss (30).

Therefore, ATG induction therapy has been considered to allow steroid sparing or steroid-free immunosuppressive regimens during the first days after transplantation (30). Steroid-free immunosuppression with ATG induction and Tacrolimus monotherapy was performed in a few trials. The results of these trials demonstrated a decrease in typical steroid-induced side effects like post-transplant diabetes, postoperative Cytomegalo Virus (CMV) infections and a lower viral load of Hepatitis C Virus (HCV) in HCV infected patients. The incidence of steroid-requiring rejections was also lower while there were no significant disadvantages in the steroid-free therapy regimens apparent (31,32).

1.2.2 Calcineurin Inhibitors

The first introduction of Calcineurin Inhibitors in combination with Steroids showed a great improvement of liver transplantation results (33). After the initiation of Calcineurin Inhibitors the number of liver transplantations and the success in regard to patient survival was continuously rising (34). After the Calcineurin Inhibitor Cyclosporine a new CNI called Tacrolimus (TAC) was introduced in 1989. Advantages of Tacrolimus in relation to acute steroid-resistant and chronic rejections could be shown in clinical trials while no significant differences in patient and graft survival could be observed. However, these trials also demonstrated increased drug toxicity in regard to the nephro-, and neurotoxicity and diabetogenic side effects of Tacrolimus (35,36). One of the main adverse effects of CNI's is their nephrotoxicity which may lead to renal failure which is associated with higher mortality after liver transplantation (18,36–39).

1.2.3 Antimetabolites

Azathioprin and Mycophenolate Mofetil (MMF) are the two drugs used in liver transplantation patients for immunosuppression. Antimetabolites reduce purine synthesis affecting T- and B-cell proliferation. Nowadays Azathioprin is rarely used after liver transplantation and the standard antimetabolite in clinical practice is

Mycophenolate Mofetil (MMF), which was first established in the late 1990's as an immunosuppressive agent without nephro-, neuro-, and hepatotoxicity and which was first implemented after kidney transplantation. In trials it was shown that MMF should not be used for primary immunosuppression, while it can help to reduce rejection rates when deployed in addition to lower dosed CNIs. Nowadays MMF is used routinely in combination with reduced dosages of Steroids and reduced dosages of Calcineurin Inhibitors (3,40,41).

1.2.4 m-Tor Inhibitors

Everolimus and Sirolimus belong to the group of m-Tor Inhibitors. They affect T- and B-Cell proliferation by blockade of IL-2 and IL-15 pathways. Sirolimus was first used after renal transplantation and according to studies after liver transplantation it is suspected to increase the rate of early hepatic artery thrombosis post-transplantation with consecutive higher mortality and higher graft loss after transplantation (42). M-Tor Inhibitors are used for preventing graft rejection and are considered as an alternative to CNIs. Reported dose dependent adverse effects include hyperlipidemia, thrombocytopenia, anemia and leukopenia due to reduced proliferation of B- and T-Lymphocytes (43–46). The absence of neurotoxicity and nephrotoxicity as well as no diabetogenesis makes mTOR inhibitors a favorable alternative to Steroids and/or CNIs (43–46).

1.3 Aim of this Study

There exist only few studies which investigate ATG induction therapy after liver transplantation and its effects on patient survival, graft survival and cancer-free survival as well as infective complications and the development of post-transplant kidney function.

At the General, Visceral and Transplant Surgery Department, Transplant Center Graz at the Medical University of Graz a low dose ATG induction therapy was used after liver transplantation in many patients until April 2017. The controversial nature of ATG induction therapy and the lack of published data on ATG induction therapy after liver transplantation prompted this study.

The aim of this retrospective survey is to investigate the benefits and risks of low dose ATG induction therapy after liver transplantation.

2 Patients and Methods

2.1 Setting

The setting of this study was provided by the General, Visceral and Transplant Surgery Department, Transplant Center Graz at the Medical University of Graz which belongs to the Eurotransplant community. A retrospective data set with patient data from 1.1.2007 to 31.12.2018 was examined. The study has been approved by the ethics committee of Medical University of Graz.

2.2 Inclusion and Exclusion Criteria

Figure 1 demonstrates the flow of patients through the study guided by predefined inclusion and exclusion criteria. All patients who have received primary liver transplantation between 1.1.2007 and 31.12.2018 were included (n=217). Patients with additional kidney transplants within the first year and patients with lack of data on ATG induction were excluded (n=6, 2.76%). Defining Study Cohort 1 (n = 211) which was used to investigate independent influences of ATG induction on patient survival, graft survival, cancer-free patient survival and rejection-free graft survival as well as CMV infections, urinary tract infections and bronchopulmonary infections within the first month after transplantation (n=211). For further analysis all patients with lack of data for post-operative KDIGO-stages at 6 months after transplant due to death or loss of follow up were excluded (n=31, 14.69%) leading to Study Cohort 2 (n = 180). Study Cohort 2 (n=180) was used to determine the correlation between pre-transplant KDIGO-stages and post-transplant KDIGO-stages at 6 months after transplant and to investigate the independent influence of ATG induction on KDIGO stage improvement.

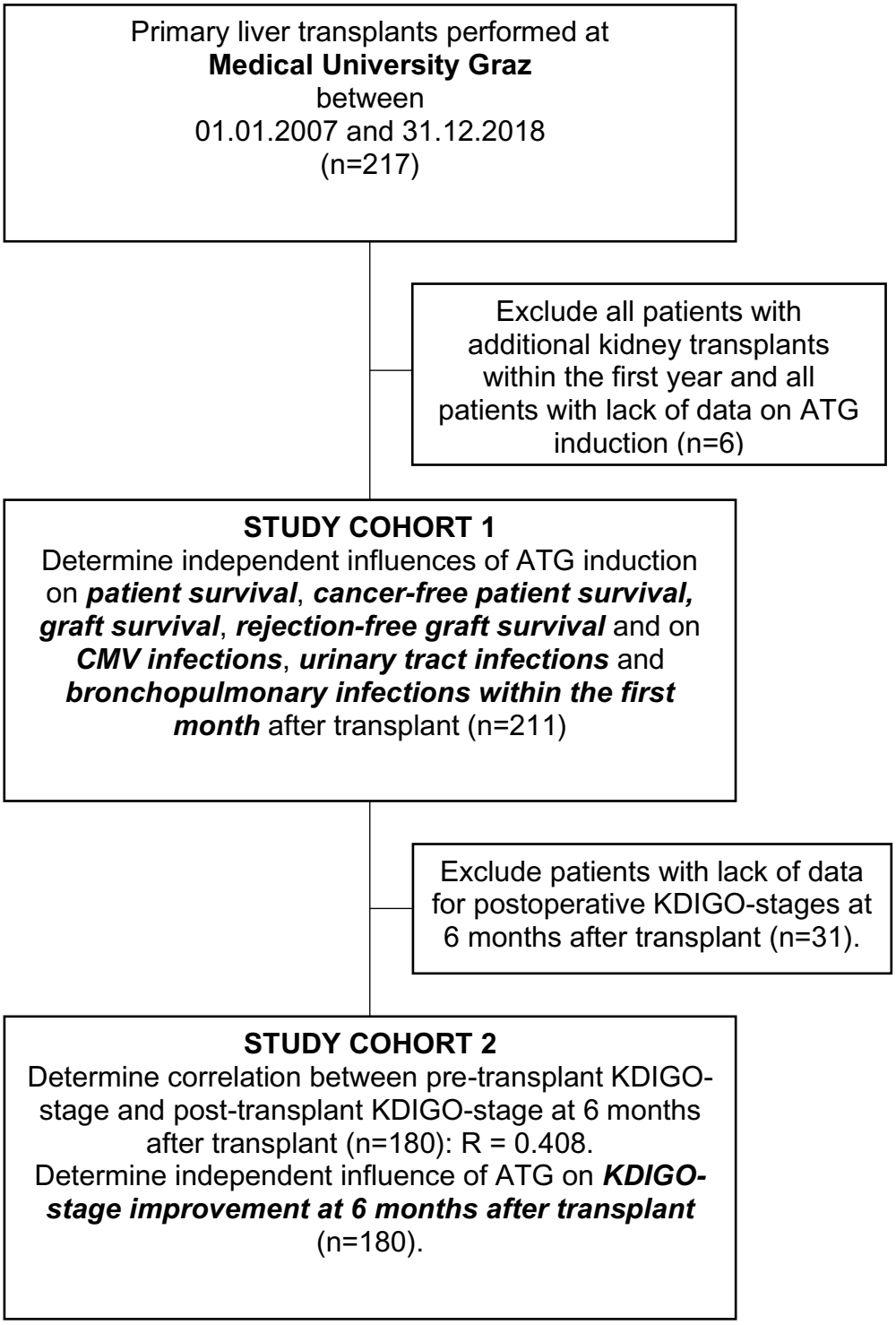

Figure 1 Shown is the flow of patients through the study

2.3 Immunosuppression

During the first week after liver transplantation, all patients received Thymoglobulin© 0.5 to 1.0 mg/kg/d for 3-4 days and a Methylprednisolone taper starting with 70 mg every 8 hours. Calcineurin Inhibitors (CNIs) were initiated according to kidney function on days 1 to 3, and standard trough levels were intended to be reached on day 4. Tacrolimus trough levels were between 5 and 8 ng/ml and Cyclosporine levels between 100 and 150 ng/ml. From day 7, Prednisolone was given at 15 mg/d and was tapered after the first month. Steroids were stopped not later than six months after liver transplantation. MMF was started orally between days 2 and 3 with target through levels about 1 µg/mL.

Maintenance therapy consisted of CNIs (preferably with Tacrolimus) and MMF. Tacrolimus trough levels were during this period around 5 ng/ml and Cyclosporine levels around 100 ng/ml, and MMF target through levels were about 1 µg/mL. Reasons to add mTor inhibitors (Everolimus or Sirolimus) to the immunosuppressive regimen were renal or neurological impairment, hepatocellular carcinoma as indication for liver transplantation, de-novo cancer of any origin or dignity and viral hepatitis. Trough levels of mTor inhibitors ranged from 3-5 ng/ml.

2.4 Definitions of variables

Bronchopulmonary infections were defined for the purpose of this study as bronchitis, pneumonia, ARDS, pneumonitis and events with acute respiratory insufficiency treated with antibiotics.

Urinary tract infections were defined for the purpose of this study by all episodes with formal diagnosis of urinary tract infection, positive urine test stripes including nitrogen and leukocyte positivity in the urine, as well as positive “Uricult tests”, and reported positive responses after antibiotic treatment.

Kidney graft function was staged into five ordinal Kidney Disease Improving Global Outcomes-categories (KDIGO): Stage 1 equals an estimated glomerular filtration rate (eGFR) with ≥90 ml/min (normal kidney function); stage 2 with an eGFR 60-89 ml/min (mildly decreased renal function); stage 3a resembles an eGFR 45-60 ml/min and stage 3b an eGFR 30-44 ml/min (moderately decreased kidney function); stage 4 equals an eGFR 15-29 ml/min (severely decreased renal function); stage 5 equals

an eGFR <15 ml/min or dialysis (graft failure) (Stevens, et al., 2013).eGFR values were calculated for this staging using the CKD Epi formula as published before (48).

2.5 Study endpoints

Primary study endpoints were patient survival, graft survival, cancer-free survival and rejection-free graft survival.

Patient survival was defined as the time in years between transplantation and death or date last seen alive. The chosen unit for all survival time analyses in this study was years with one decimal.

In this study graft survival was defined as the time between primary liver transplantation and either the first retransplant or patient death in those patients who did not receive a retransplant.

Cancer-free survival was defined as the time between primary liver transplantation and either the date of the first diagnosis of malignancy or patient death in those patients who were not diagnosed with a malignancy during follow-up.

Rejection-free graft survival was defined as the time between primary liver transplantation and the occurrence of the first rejection episode or graft loss defined by either retransplantation or patient death.

All survivors during follow-up of this study were censored in Kaplan-Meier analyses and Cox regression analyses.

Secondary study end-points were CMV PCR positive episodes within the first month (yes/no) and within the first six months after liver transplantation (yes/no), bronchopulmonary and/or urinary tract infections within the first month after liver transplantation as a combined study-endpoint (yes/no) as well as bronchopulmonary infections within the first month after liver transplantation (yes/no) and urinary tract infections within the first month after liver transplantation (yes/no) after liver transplantation as separate study endpoints.

Primary and secondary study endpoints were investigated using Study Cohort 1.

The tertiary study-endpoint was defined as KDIGO stage improvement six months after liver transplantation (yes/no) in comparison to individual pre-transplant KDIGO stages.

2.6 Statistical methods

The significance of differences in the distribution of categorical variables and continuous variables between groups was tested with the Pearson's Chi^2 test and the Wilcoxon test, respectively.

Risk factors for primary survival study endpoints were evaluated in univariable Cox regression analysis and in Kaplan-Meier analyses with log rank tests for categorical variables.

Multivariable Cox regression analysis was performed to determine the independent influence of ATG induction therapy on primary survival study-endpoints adjusted for risk factors with an influence on the respective primary study endpoint in univariable Cox regression analysis with p-values <0.200. The results of univariable Cox regression analyses are summarized in the Supplementary Tables in the Appendix.

Univariable binary logistic regression analysis was used to determine the influence of variables on all binary secondary and tertiary study-endpoints. Multivariable binary logistic regression analysis was used to determine the independent influence of ATG induction adjusted for risk factors with an influence on these binary study endpoints in the respective univariable logistic regression analyses with p-values <0.200. The results of univariable binary logistic regression analyses are summarized in the Supplementary Tables in the Appendix.

ROC-curve analysis was performed to determine the area under the receiver operating curve (AUROC) to assess the sensitivity and specificity of the models' predictions of secondary and tertiary binary study-endpoints. AUROCs >0.700 are widely regarded as a prerequisite for clinically useful prognostic models (49,50)

P-values < 0.05 were defined as significant. JMP Pro 13.0 statistics software was used to conduct statistical analyses (SAS Institute, Cary, NC, USA).

3 Results

3.1 Patients

Figure 1 shows the flow of patients through the study. A total of 211 patients fulfilled the inclusion and exclusion criteria of this study and could be evaluated. 31 patients with lack of data for postoperative KDIGO-stages at 6 months after transplant were excluded from these 211 patients for the determination of the independent influence of ATG on KDIGO-stage improvement at 6 months after transplant patients (see Figure 1). The following Table 1 summarizes the distribution of all relevant pre- and post-transplant variables in the investigated cohort of 211 patients.

Variables			Distribution	Missing values (n; %)
Pre-transplant variables		Gender male (n; %)	170 ; 80.6%	0 ; 0%
		Gender female (n; %)	41 ; 19.4%	0 ; 0%
		Age at transplant (years) (median; range)	58.7 ; 15.2 – 74.8	0 ; 0%
	Indications	Autoimmune hepatitis (n; %)	6 ; 2.8%	0 ; 0%
		Alcoholic liver disease (n; %)	87 ; 41.2%	0 ; 0%
		Acute liver failure (n; %)	3 ; 1.4%	0 ; 0%
		Hepatitis B virus cirrhosis (n; %)	4 ; 1.9%	0 ; 0%
		Hepatocellular carcinoma (HCC) (n; %)	44 ; 20.9%	0 ; 0%
		Hepatitis C virus cirrhosis (n; %)	10 ; 4.7%	0 ; 0%
		Hepatitis C virus cirrhosis + HCC (n; %)	28 ; 13.3%	0 ; 0%
		Primary sclerosing cholangitis (n; %)	8 ; 3.8%	0 ; 0%
		Secondary sclerosing cholangitis (n; %)	6 ; 2.8%	0 ; 0%
		Other (n; %)	15 ; 7.1%	0 ; 0%
		Height (m) (median; range)	1.76 ; 1.33 – 1.97	0 ; 0%
		Weight (kg) (median; range)	80 ; 40 – 122	1 ; 0.47%
		BMI (kg/m²) (median; range)	25.4 ; 16.4 – 43.7	1 ; 0.47%
	Kidney function	KDIGO stage I (n; %)	86 ; 41.0%	1 ; 0.47%
		KDIGO stage II (n; %)	86 ; 41.0%	
		KDIGO stage IIIa (n; %)	22 ; 10.5%	
		KDIGO stage IIIb (n; %)	11 ; 5.2%	
		KDIGO stage IV (n; %)	3 ; 1.4%	
		KDIGO stage V (n; %)	2 ; 1.0%	
		Donor CMV IgG pos (n; %)	99 ; 47.6%	3 ; 1.4%
		Recipient CMV IgG pos (n; %)	136 ; 65.1%	2 ; 0.95%
Post-transplant variables	Immunosuppression	Induction with Basiliximab (n; %)	6 ; 2.9%	1 ; 0.47%
		Induction with ATG (n; %)	131 ; 62.1%	0 ; 0%
		Induction with ALG (n; %)	12 ; 5.7%	0 ; 0%
		Cyclosporine A (n; %)	41 ; 19.5%	1 ; 0.47%
		Tacrolimus (n; %)	197 ; 93.8%	1 ; 0.47%
		Mycophenolate Mofetil (n; %)	203 ; 96.7%	1 ; 0.47%
		Azathioprin (n; %)	4 ; 1.9%	1 ; 0.47%
		Everolimus (n; %)	76 ; 36.2%	1 ; 0.47%
		Sirolimus (n; %)	44 ; 21.0%	1 ; 0.47%
		De novo cancer (n; %)	26 ; 12.4%	1 ; 0.47%
	Infections	Urinary tract infection within 1st month (n; %)	18 ; 8.5%	0 ; 0%
		Bronchopulmonary infection within 1st month (n; %)	34 ; 16.1%	0 ; 0%
		Post-Tx CMV PCR + within 1st month (n; %)	48 ; 22.7%	0 ; 0%
		CMV positive episodes (median, range)	0 ; 0 – 6	0 ; 0%
		Subsequent Re-Tx (n; %)	7 ; 3.3%	0 ; 0%
		Number of deaths (n; %)	52 ; 24.6%	0 ; 0%
		Kaplan-Meier: Estimated 1-year survival (%)	84.2%	0%
		Kaplan-Meier: Estimated 1-year graft survival (%)	82.7%	0%

Table 1 Shown is the distribution of variables in the investigated cohort (n = 211) and the frequencies and percentages of missing values. Displayed data on immunosuppression indicates types of drugs administered during anytime follow-up after transplantation which may be temporary.

Figure 2 shows the distribution of the number of patients with ATG induction therapy versus without ATG induction therapy separated per year from 2007 to 2018.

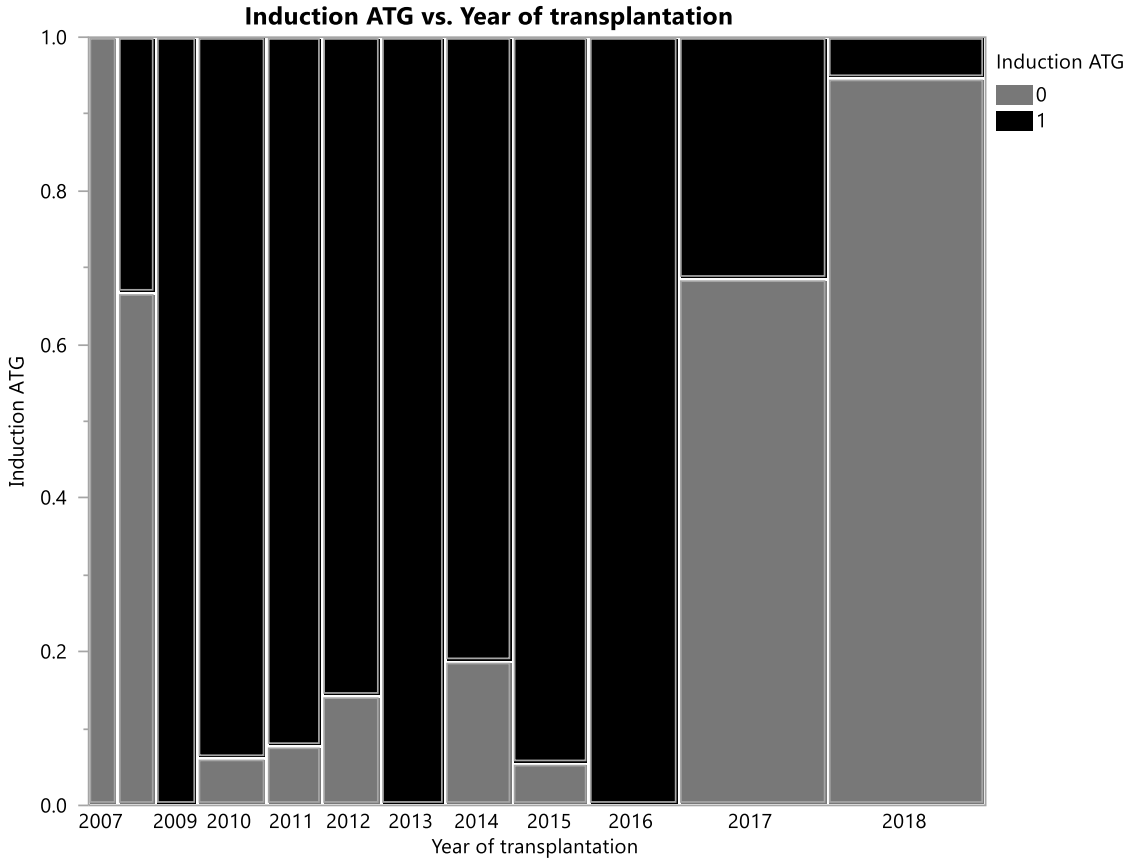

Figure 2 Shown is the distribution between **ATG induction versus No-ATG induction** therapy separated per year. Case numbers per year are reflected by the width of the bars for each year in which the investigated primary transplants were performed.

3.2 Comparison of ATG induction versus no ATG induction

Table 2 summarizes the statistical significance of differences in the distribution of pre- and post-transplant variables between those patients who were treated with ATG induction therapy (n = 131) versus those who did not receive any ATG induction (n = 88). Significant differences could only be detected for the distribution of the indication hepatocellular carcinoma (16.0 % with ATG versus 28.8 % without ATG, p = 0.027), induction with ALG (0.0% with ATG versus 15.0% without ATG, p = <0.001), immunosuppression with Everolimus (42.7% with ATG versus 25.3% without ATG , p = 0.011), immunosuppression with Sirolimus (26.7 with ATG induction versus 11.4% without ATG, p = 0.008), post-transplant de novo cancer (16.0% with ATG versus 6.3% without ATG, p = 0.039), and post-transplant CMV PCR positive episodes within the first month (31.3% with ATG versus 8.8% without ATG, p < 0.001) (Table 2).

Variables			ATG Induction (n=131)	No ATG Induction (n=80)	p-value*
Pre-transplant variables		Gender male (n; %)	104 ; 79.4%	66 ; 82.5%	0.580
		Gender female (n; %)	27 ; 20.6%	14 ; 17.5%	0.580
		Age at transplant (years) (median; range)	58.0 ; 18.1 – 74.7	59.7 ; 15.2 – 74.8	0.212
	Indications	Alcoholic liver disease (n; %)	55 ; 42.0%	32 ; 40.0%	0.776
		Hepatitis C virus cirrhosis + HCC (n; %)	20 ; 15.3%	8 ; 10.0%	0.274
		Viral liver cirrhosis (n; %)	9 ; 6.9%	5 ; 6.3%	0.861
		Hepatocellular carcinoma (HCC) (n; %)	**21 ; 16.0%**	**23 ; 28.8%**	**0.027**
		Other (n; %)	26 ; 19.8%	12 ; 15.0%	0.374
		Height (m) (median; range)	1.76 ; 1.52 – 1.97	1.75 ; 1.33 – 1.94	0.923
		Weight (kg) (median; range)	77.5 ; 40 – 122	80 ; 48 – 108	0.484
		BMI (kg/m²) (median; range)	25.4 ; 16.4 – 43.7	25.4 ; 17.6 – 33.7	0.536
	Kidney function	KDIGO stage I (n; %)	46 ; 35.1%	40 ; 50.6%	0.228
		KDIGO stage II (n; %)	57 ; 43.5%	29 ; 36.7%	
		KDIGO stage IIIa (n; %)	16 ; 12.2%	6 ; 7.6%	
		KDIGO stage IIIb (n; %)	8 ; 6.1%	3 ; 3.8%	
		KDIGO stage IV (n; %)	3 ; 2.3%	0 ; 0%	
		KDIGO stage V (n; %)	1 ; 0.8%	1 ; 1.3%	
		Donor CMV IgG pos (n; %)	57 ; 44.2%	42 ; 53.2%	0.208
		Recipient CMV IgG pos (n; %)	87 ; 66.4%	49 ; 62.8%	0.598
Post-transplant variables	Immunosuppression	Induction with Basiliximab (n; %)	5 ; 3.8%	1 ; 1.3%	0.282
		Induction with ALG (n; %)	**0 ; 0%**	**12 ; 15.0%**	**<0.001**
		Cyclosporine A (n; %)	28 ; 21.4%	13 ; 16.5%	0.384
		Tacrolimus (n; %)	125 ; 95.4%	72 ; 91.1%	0.212
		Mycophenolate Mofetil (n; %)	129 ; 98.5%	74 ; 93.7%	0.060
		Azathioprin (n; %)	4 ; 3.1%	0 ; 0%	0.117
		Everolimus (n; %)	**56 ; 42.7%**	**20 ; 25.3%**	**0.011**
		Sirolimus (n; %)	**35 ; 26.7%**	**9 ; 11.4%**	**0.008**
		De novo cancer (n; %)	**21 ; 16.0%**	**5 ; 6.3%**	**0.039**
		Urinary tract infection within first month (n; %)	10 ; 7.6%	10 ; 12.5%	0.551
		Bronchopulmonary infection within first month (n; %)	24 ; 18.3%	8 ; 10.0%	0.264
		Post-Tx CMV PCR + within first month (n; %)	**41 ; 31.3%**	**7 ; 8.8%**	**<0.001**
		Subsequent Re-Tx (n; %)	3 ; 2.3%	4 ; 5.0%	0.286

Table 2 Shown is the distribution of variables between those patients who were treated with ATG versus those who did not receive ATG (* Chi² test for nominal variables, Wilcoxon test for continuous variables).

3.3 Influence of ATG on primary study endpoints

3.3.1 Influence of ATG on patient survival

Kaplan-Meier analysis revealed that ATG had no statistically significant influence on patient survival (log rank test: p=0.737) (Figure 3a).

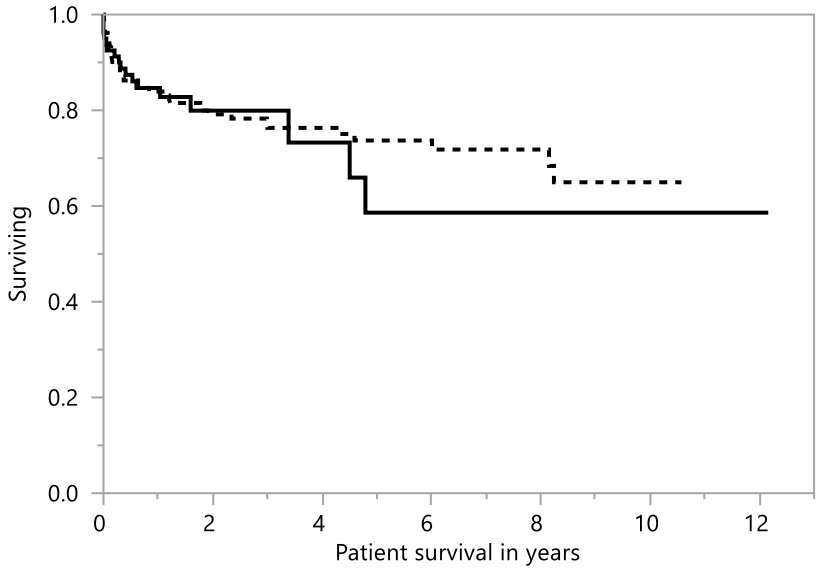

Figure 3a Shown is the influence of ATG induction (dotted line, n=131) versus no ATG induction (straight line, n=80) on patient survival (Kaplan-Meier analysis, log rank test: p=0.737).

Supplementary Table 1 in the Appendix summarizes the results of univariable Cox regression analysis to determine the impact of investigated variables on patient survival. Multivariable Cox regression analysis adjusted for ATG induction therapy demonstrated that higher pre-transplant KDIGO stages (I-V) are an independent risk factor for early death after transplantation (hazard ratio = 1.369, 95%-CI = 1.077-1.700, p = 0.012) while a donor CMV IgG positive status was identified as an independent protective factor for early death after transplantation (hazard ratio = 0.464, 95%-CI = 0.245-0.841, p = 0.011). This multivariable analysis further revealed that ATG induction treatment has no independent significant influence on early death after transplantation (hazard ratio = 0.917, 95%-CI = 0.493-1.791, p = 0.791) (see Table 3).

Variables	Hazard ratio	95% - CI	p-value
KDIGO stage (I-V)	**1.369**	**1.077-1.700**	**0.012**
Donor CMV IgG pos (yes/no)	**0.464**	**0.245-0.841**	**0.011**
Induction with ATG (yes/no)	0.917	0.493-1.791	0.791

Table 3 Shown are the results of multivariable Cox regression analysis to determine the independent influence of ATG induction on **early death after transplantation** adjusted for risk factors with an influence on patient survival in univariable Cox regression analysis with p-values <0.200 (see Supplementary Table 1 in the Appendix). Variables with independent significant influences are marked in bold letters (p<0.050).

3.3.2 Influence of ATG on graft survival

Kaplan-Meier analysis revealed that ATG had no statistically significant influence on graft survival (log rank test: p=0.539) (Figure 3b).

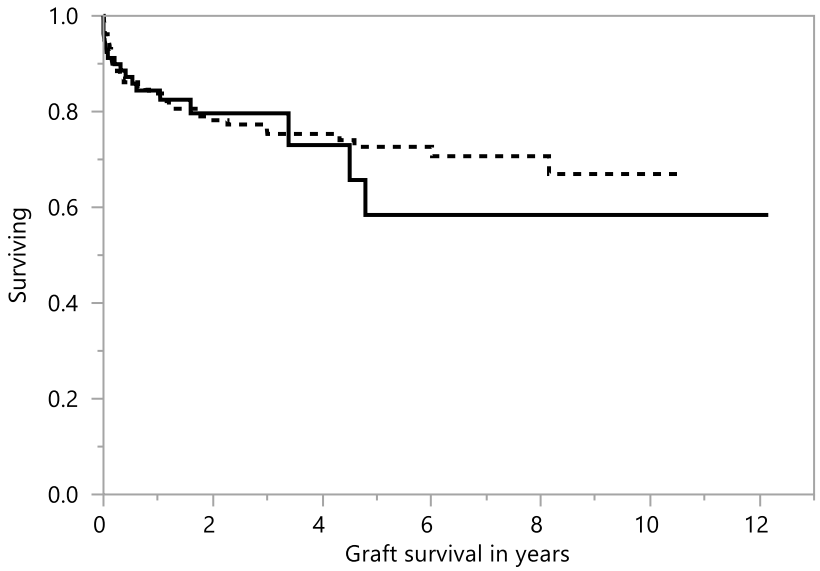

Figure 3b Shown is the influence of ATG induction (dotted line, n=131) versus no ATG induction (straight line, n=80) on graft survival (Kaplan-Meier analysis, log rank test: p=0.539).

Supplementary Table 2 in the Appendix summarizes the results of univariable Cox regression analysis to determine the impact of investigated variables on graft survival. Adjusted multivariable Cox regression analysis revealed that the combined indication of Hepatitis C virus cirrhosis with Hepatocellular Carcinoma (hazard ratio

= 2.033, 95-%CI = 0.981-3.890, p = 0.041) and higher pre-transplant KDIGO stages (hazard ratio = 1.329, 95-%CI = 1.039-1.660, p = 0.017) were independently significant risk factors for early graft loss after transplantation. In this analysis a donor CMV IgG positive status (hazard ratio = 0.557, 95-%CI = 0.306-0.982, p = 0.048) was an independently significant protective factor for early graft loss after transplantation adjusted for ATG induction therapy (see Table 4).

Variables	Hazard ratio	95% - CI	p-value
Hepatitis C virus cirrhosis + HCC (yes/no)	**2.033**	**0.981-3.890**	**0.041**
KDIGO stage (I-V)	**1.329**	**1.039-1.660**	**0.017**
Donor CMV IgG pos (yes/no)	**0.557**	**0.306-0.982**	**0.048**
Induction with ATG (yes/no)	0.808	0.445-1.527	0.495

Table 4 Shown are the results of multivariable Cox regression analysis to determine the independent influence of ATG induction on **early graft loss** after transplantation adjusted for risk factors with an influence on early graft loss in univariable Cox regression analysis with p-values <0.200 (see Supplementary Table 2 in the Appendix). Variables with independent significant influences are marked in bold letters (p<0.050).

3.3.3 Influence of ATG on cancer-free survival

Kaplan-Meier analysis revealed that ATG had no statistically significant influence on patient survival (log rank test: p=0.737), graft survival (log rank test: p=0.539) and cancer-free survival (log rank test: p=0.946) (Figure 3c).

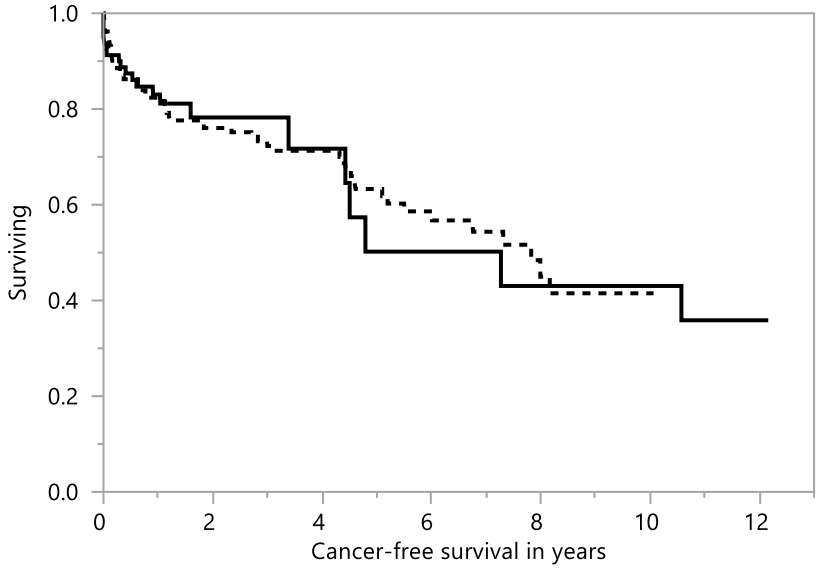

Figure 3c Shown is the influence of ATG induction (dotted line, n=131) versus no ATG induction (straight line, n=80) on cancer-free survival (Kaplan-Meier analysis, log rank test: p=0.946).

Supplementary Table 3 in the Appendix summarizes the results of univariable Cox regression analysis to determine the impact of investigated variables on cancer-free survival. Multivariable Cox regression analysis adjusted for ATG induction therapy detected higher KDIGO stages (hazard ratio = 1.332, 95-%CI = 1.069-1.630, p = 0.012) as an independently significant risk factor for cancer-free survival after transplantation while a donor CMV IgG positive status (hazard ratio = 0.714, 95%-CI = 0.433-1.160, p = 0.175) was identified as an independent protective factor after adjustment for ATG induction (Table 5).

Variables	Hazard ratio	95% - CI	p-value
Pseudometric KDIGO stage (1-6)	**1.332**	**1.069-1.630**	**0.012**
Donor CMV IgG pos (yes/no)	0.714	0.433-1.160	0.175
Induction with ATG (yes/no)	1.006	0.580-1.826	0.985

Table 5 Shown are the results of multivariable Cox regression analysis to determine the independent influence of ATG induction on **cancer-free survival** after transplantation adjusted for risk factors with an influence on cancer-free survival in univariable logistic regression analysis with p-values <0.200 (see Supplementary Table 3 in the appendix). Variables with independent significant influences are marked in bold letters (p<0.050).

3.3.4 Influence of ATG on rejection-free graft survival

Kaplan-Meier analysis showed that higher numbers of rejection episodes during follow-up decreased patient survival significantly (log rank test: p < 0.001) (Figure 3d). As soon as there was one rejection episode apparent survival decreased significantly.

Figure 3d Shown is the significant influence of the number of rejection episodes during follow-up on **rejection free graft survival** [straight black line = 0 episodes (n=171, 81.0%), dotted black line = 1 episode (n=30, 14.2%), straight grey line = 2 episodes (n=8, 3.8%), dotted grey line ≥ 3 episodes (n=2, 0.1%)] after liver transplantation (log rank test: $p < 0.001$).

Kaplan-Meier analysis revealed no significant influence of ATG induction therapy on rejection-free graft survival (log rank test: $p = 0.920$) (Figure 3e).

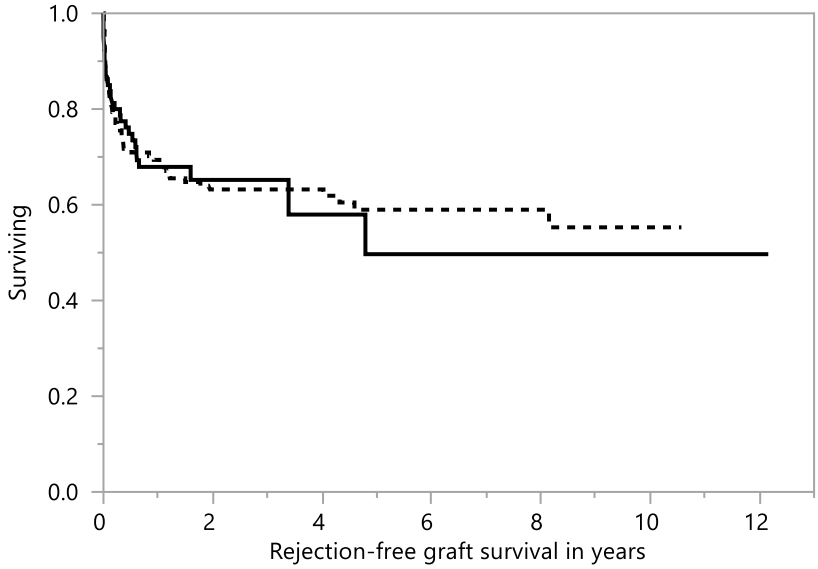

Figure 3e Shown is the influence of ATG on **rejection-free graft survival** (dotted line = ATG; straight line = no ATG induction therapy) (log rank test: $p = 0.920$).

Supplementary Table 4 in the Appendix summarizes the results of univariable Cox regression analysis to determine the impact of investigated variables on rejection-free graft survival. Multivariable Cox regression analysis adjusted for ATG induction therapy identified higher pre-transplant KDIGO stages (hazard ratio = 1.268, 95%-CI = 1.023-1.548, p = 0.031) as an independently significant risk factor for rejection-free graft survival after liver transplantation. In contrast, a donor positive CMV status combined with a recipient negative CMV status (hazard ratio = 0.427, 95%-CI = 0.173-0.914, p = 0.027) was determined as a protective factor for rejection-free graft survival.

Variables	Hazard ratio	95% - CI	p-value
Age at transplant (years)	0.981	0.958-1.005	0.111
Alcoholic liver disease (yes/no)	0.739	0.416-1.314	0.301
Viral liver cirrhosis (yes/no)	1.958	0.801-4.318	0.133
Other (yes/no)	1.140	0.526-2.391	0.734
KDIGO stage (I-V)	**1.268**	**1.023-1.548**	**0.031**
D- / R+	1.368	0.833-2.244	0.214
D+ / R-	**0.427**	**0.173-0.914**	**0.027**
Induction with ATG (yes/no)	0.795	0.482-1.338	0.381

Table 6 Shown are the results of multivariable Cox regression analysis to determine the **independent influence of ATG induction on rejection-free graft survival** after transplantation adjusted for risk factors with an influence on early graft loss in univariable Cox regression analysis with p-values <0.200 (see Supplementary Table 4 in the Appendix). Variables with independent significant influences are marked in bold letters (p<0.050).

3.4 Influence of ATG on secondary study endpoints

3.4.1 Influence of ATG on post-transplant infections

The only identified independently significant risk factor for urinary tract and/or bronchopulmonary infections within the first month after transplantation adjusted for ATG induction were high BMI values > 25 kg/m² (odds ratio = 0.418, 95%-CI = 0.212-0.824, p = 0.010) (see Table 7).

Variables	Odds ratio	95% - CI	p-value
Hepatocellular carcinoma (HCC) (yes/no)	0.512	0.195-1.343	0.155
BMI > 25 kg/m² (yes/no)	**0.418**	**0.212-0.824**	**0.010**
Induction with ALG (yes/no)	0.248	0.029-2.144	0.143
Induction with ATG (yes/no)	1.052	0.510-2.172	0.890

Table 7 Shown are the results of a multivariable binary logistic regression analysis to determine the independent influence of ATG induction on **urinary tract and/or bronchopulmonary infections within the first month after transplant** adjusted for risk factors with an influence on this endpoint in univariable logistic regression analysis with p-values < 0.200 (Supplementary Table 5 in the Appendix). Variables with independent significant influences are marked in bold letters (p<0.050).

ROC-curve analysis of the final adjusted multivariable logistic regression model demonstrated an AUROC = 0.653 for the prediction of urinary tract and/ or bronchopulmonary infections within the first month after transplantation in a cohort of 211 patients (Figure 4).

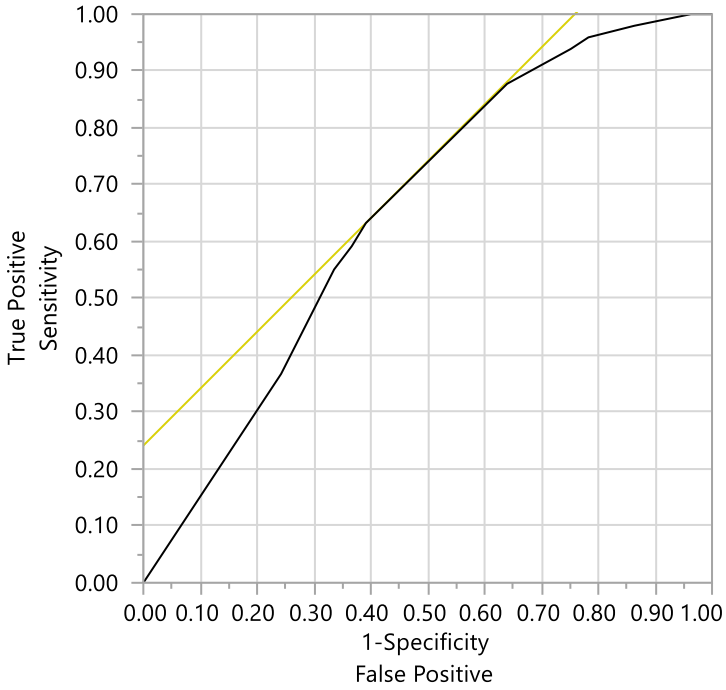

Figure 4 Shown is the result of ROC-curve analysis of the final multivariable logistic regression model for the prediction of **urinary tract and/or bronchopulmonary infections within the first month** in 211 patients after liver transplantation (AUROC=0.653).

Kaplan-Meier analysis revealed that a urinary tract and/ or bronchopulmonary infection within the first month after transplantation reduces long term patient survival significantly (log rank test: p = 0.029) (Figure 5a).

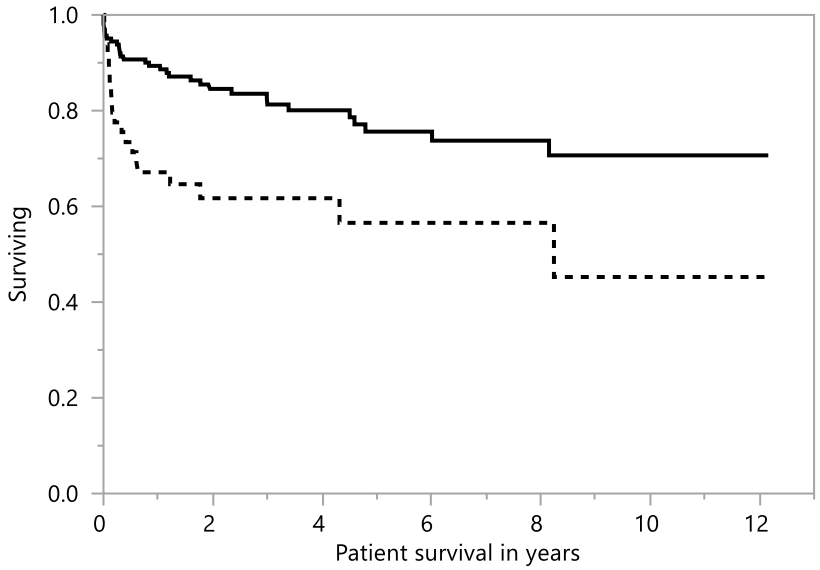

Figure 5a Shown is the significant influence of ***urinary tract and/or bronchopulmonary infections within the first month*** (dotted line, n = 49) ***versus no urinary tract and/or bronchopulmonary infections within the first month*** (straight line, n = 162) on patient survival after liver transplantation (log rank test: p = 0.029).

Kaplan-Meier analysis showed that patient survival is not significantly influenced by urinary tract infections within the first month after transplantation (log rank test: p = 0.877) (Figure 5b).

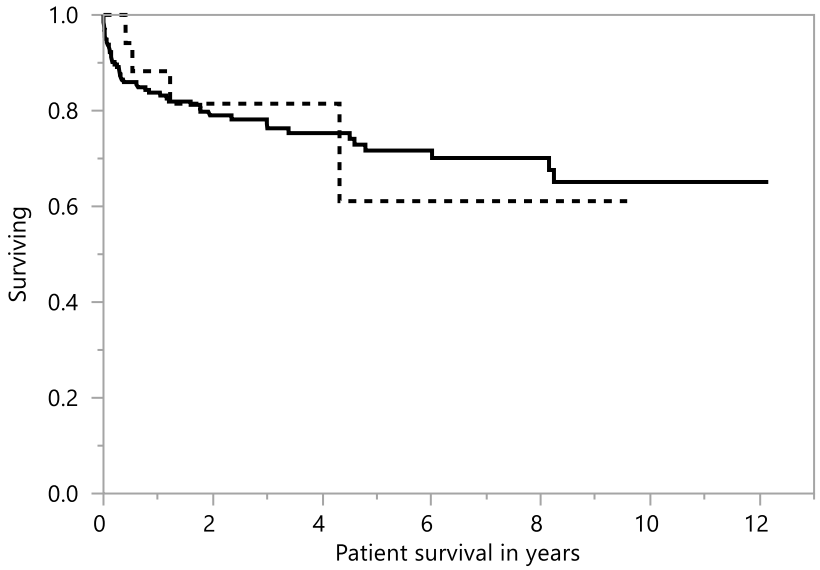

Figure 5b Shown is the significant influence of **urinary tract infections within the first month** (dotted line, n = 18) **versus no urinary tract infections within the first month** (straight line, n = 193) on patient survival after liver transplantation (log rank test: p = 0.877).

Bronchopulmonary infections within the first month after transplantation reduced patient survival significantly as identified in Kaplan-Meier analysis (log rank test: $p < 0.001$) (Figure 5c).

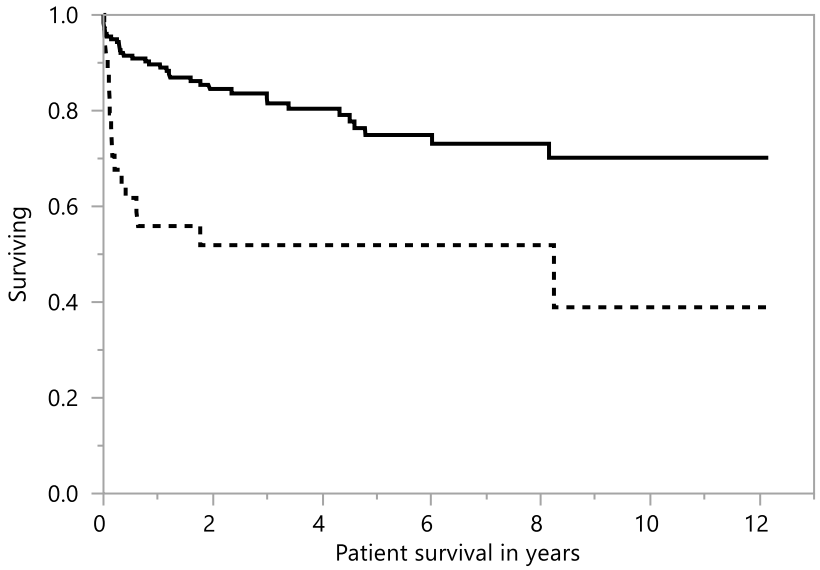

Figure 5c Shown is the significant influence of **bronchopulmonary infections within the first month** (dotted line, n = 34) **versus no bronchopulmonary infections within the first month** (straight line, n = 177) on patient survival after liver transplantation (log rank test: $p < 0.001$).

3.4.2 Influence of ATG on post-transplant CMV PCR positive episodes

Multivariable binary logistic regression analysis adjusted for ATG induction therapy showed that a CMV negative donor combined with a CMV negative recipient status (odds ratio = 0.070, 95-%CI = 0.009-0.548, p <0.001) is an independently significant protective factor against CMV PCR positive episodes within the first month after transplantation. In contrast, ATG induction therapy was an independently significant risk factor for CMV PCR positive episodes within the first month after transplantation (odds ratio = 6.160, 95%-CI = 2.410-15.741, p < 0.001) (Table 8).

Variables		Odds ratio	95% - CI	p-value
Pre-transplant variables	Hepatocellular carcinoma (HCC) (yes/no)	0.940	0.369-2.400	0.897
	BMI < 19 kg/m² (yes/no)	3.452	0.687-17.331	0.132
	D- / R-	**0.070**	**0.009-0.548**	**<0.001**
	D+ / R+	1.187	0.561-2.512	0.655
	Induction with ATG (yes/no)	**6.160**	**2.410-15.741**	**<0.001**

Table 8 Shown are the results of a multivariable binary logistic regression analysis to determine the independent influence of ATG induction and of all possible donor CMV IgG and recipient CMV IgG combinations on **CMV PCR positive episodes within the first month after transplant** adjusted for risk factors with an influence on this endpoint in univariable logistic regression analysis with p-values <0.200 (see Supplementary Table 6 in the Appendix). Variables with independent significant influences are marked in bold letters (<0.050).

ROC-curve analysis of the final adjusted multivariable logistic regression model demonstrated an AUROC = 0.756 for the prediction of CMV PCR positive episodes within the first month after transplantation with high sensitivity and specificity (Figure 6).

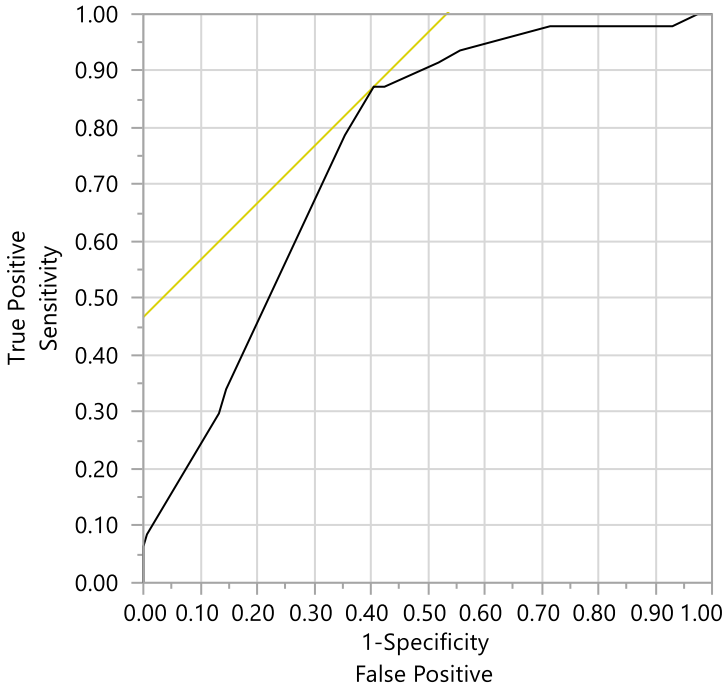

Figure 6 Shown is the result of ROC-curve analysis of the final multivariable logistic regression model for the prediction of **CMV PCR positive episodes within the first month** in 211 patients after liver transplantation (AUROC=0.756).

Kaplan-Meier analysis demonstrated that CMV PCR positive episodes within the first month after transplantation had a significant influence on patient survival (log rank test: p = 0.029) (see Figure 7).

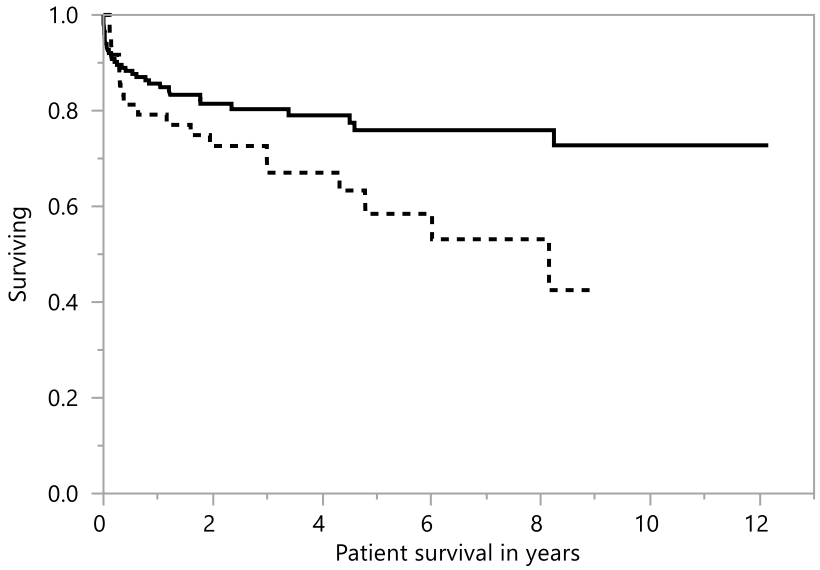

Figure 7 Shown is the significant influence of **CMV PCR positive episodes within the first month** (dotted line, n = 48) **versus no CMV PCR positive episodes within the first month** (straight line, n = 163) on patient survival after liver transplantation (log rank test: p = 0.029).

Multivariable binary logistic regression analysis adjusted for ATG induction determined the combination of donor negative CMV status with recipient negative CMV status (odds ratio = 0.145, 95%-CI = 0.040-0.521, p < 0.001), as an independently significant protective factor against CMV PCR positive episodes within the first 6 months after transplantation. A donor positive CMV status combined with a recipient positive CMV status (odds ratio = 2.269, 95%-CI = 1.138-4.524, p = 0.019) was revealed being an independently significant risk factor for CMV PCR positive episodes within the first 6 month after transplantation as well as the induction therapy with ATG (odds ratio = 3.607, 95%-CI = 1.779-7.314, p < 0.001) had an independently significant negative influence on the emergence of CMV positive episodes within the first 6 months after transplant (Table 9).

Variables		Odds ratio	95% - CI	p-value
Pre-transplant variables	Gender female (yes/no)	1.494	0.669-3.338	0.327
	Hepatocellular carcinoma (HCC) (yes/no)	0.725	0.313-1.677	0.448
	KDIGO stage (I-V)	1.209	0.875-1.671	0.250
	D- / R-	**0.145**	**0.040-0.521**	**<0.001**
	D+ / R+	**2.269**	**1.138-4.524**	**0.019**
	Induction with ATG (yes/no)	**3.607**	**1.779-7.314**	**<0.001**

Table 9 Shown are the results of multivariable binary logistic regression analysis to determine the independent influence of ATG induction on **CMV PCR positive episodes within the first 6 months after transplant** adjusted for risk factors with an influence on this endpoint in univariable logistic regression analysis with p-values <0.200 (see Supplementary Table 7 in the Appendix) Variables with independent significant influences are marked in bold letters (<0.050).

ROC-curve analysis of the final adjusted multivariable logistic regression model demonstrated an AUROC = 0.764 for the prediction of CMV PCR positive episodes within the first 6 months after transplantation (Figure 8).

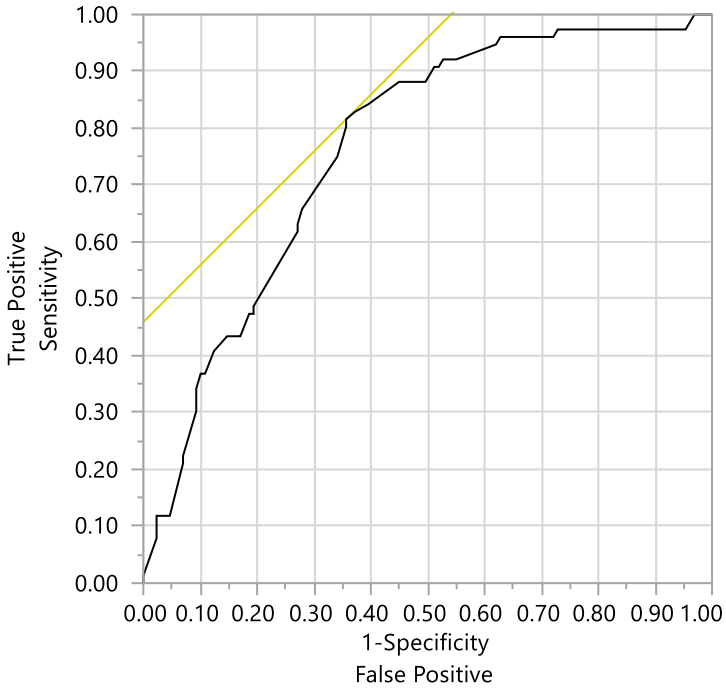

Figure 8 Shown is the result of ROC-curve analysis of the final multivariable logistic regression model for the prediction of **CMV PCR positive episodes within the first 6 months** after transplant in 211 patients after liver transplantation (AUROC=0.764).

3.4.3 Influence of ATG on KDIGO-stage improvement

Multivariable binary logistic regression analysis adjusted for ATG induction therapy identified a BMI > 25 kg/m² (odds ratio = 0.297, 95%-CI = 0.092-0.966, p = 0.036) as an independently significant risk factor for lower or no KDIGO stage improvement within the first 6 months after transplantation. In this analysis patients with higher pre-transplant KDIGO stages (odds ratio = 3.746, 95%-CI = 2.146-6.537, p = <0.001) were at an independently and significantly increased odds for larger KDIGO stage improvement within the first 6 months after transplantation. Furthermore, ATG induction therapy (odds ratio = 3.636, 95%-CI = 0.934-14.161, p = 0.043) showed in this analysis an independently significant influence on larger KDIGO stage improvement 6 months after transplantation (Table 10).

Variables		Odds ratio	95% - CI	p-value
Pre-transplant variables	Gender female (yes/no)	1.306	0.407-4.190	0.656
	BMI > 25 kg/m² (yes/no)	**0.297**	**0.092-0.966**	**0.036**
	BMI < 19 kg/m² (yes/no)	2.378	0.344-16.471	0.395
	KDIGO stage (I-V)	**3.746**	**2.146-6.537**	**<0.001**
	Donor CMV IgG pos (yes/no)	0.709	**0.241-2.087**	0.532
	Recipient CMV IgG pos (yes/no)	1.494	0.436-5.122	0.516
	Induction with ATG (yes/no)	**3.636**	**0.934-14.161**	**0.043**

Table 10 Shown are the results of a multivariable binary logistic regression analysis to determine the independent influence of ATG induction on **KDIGO-stage improvement 6 month after transplantation** adjusted for risk factors with an influence on this endpoint in univariable logistic regression analysis with p-values <0.200 (see Supplementary Table 8 in the Appendix). Variables with independent significant influences are marked in bold letters (p<0.050).

ROC-curve analysis of the final multivariable logistic regression model shows high sensitivity and specificity for the prediction of KDIGO stage improvement 6 months after liver transplantation (AUROC=0.916) (Figure 9).

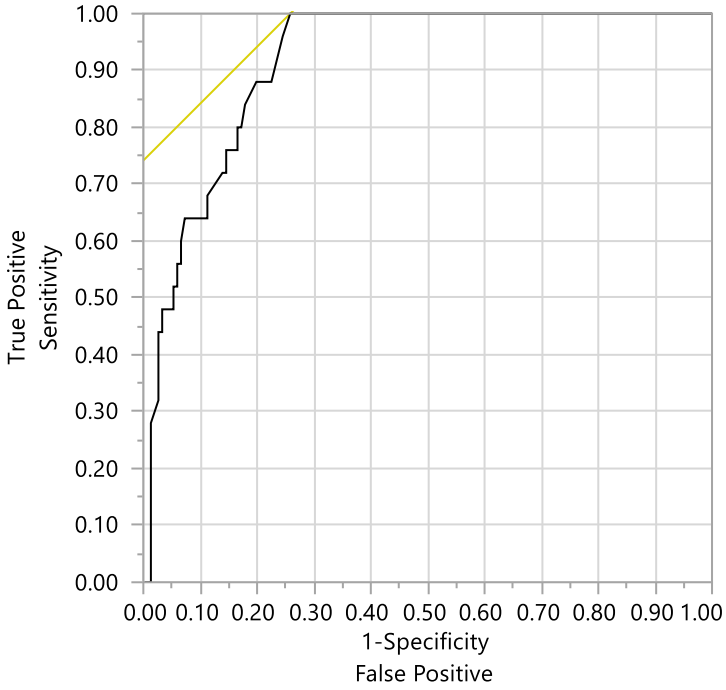

Figure 9 Shown is the result of ROC-curve analysis of the final multivariable logistic regression model for the prediction of **KDIGO-stage improvement** in 180 patients 6 months after liver transplantation (AUROC=0.916).

3.4.4 Influence of ATG on Tacrolimus levels 3 months after transplantation

The blood level of Tacrolimus 3 months after transplantation was determined to be significantly lower in patients with low-dose ATG-induction therapy. The two-sided Wilcoxon test showed a high significance ($p<0.001$).

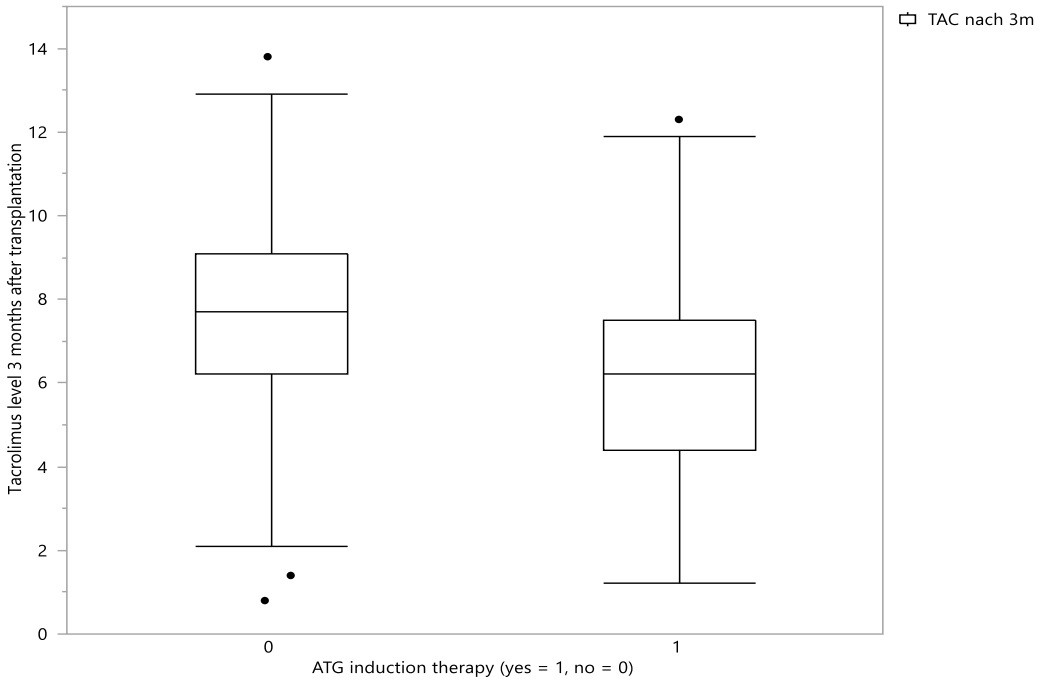

Figure 10 Shown is box plot of Tacrolimus levels 3 months after transplantation with low-dose ATG induction therapy versus without ATG induction therapy. 170 patients (80.6%) from Study Cohort 1 (n=211) were investigated due to known Tacrolimus levels.

4 Discussion

4.1 Principle findings

4.1.1 Effects of ATG induction therapy

This study shows that low-dose ATG induction therapy is safe in regard to patient survival, graft survival and cancer-free survival. It is noteworthy that ATG had no significant influence on rejection-free graft survival in this study.

ATG increased the risk for CMV-PCR positive episodes within the first month and within the first six months after transplantation independently and significantly, whereas a Donor and Recipient CMV IgG negative status (D-/R-) was shown to be an independently significant protective factor. As shown in this study, a CMV-PCR positive episode within the first month after transplantation decreases patient survival significantly during follow up. These findings strongly suggest the requirement of CMV prophylaxis in patients treated with ATG induction therapy.

Low-dose ATG induction therapy showed an independently significant influence on KDIGO stage improvement within the first 6 months after transplantation compared to pre-transplant KDIGO stages. This effect is significantly greater in those patients with higher pre-transplant KDIGO stages and in patients with BMI values ≤ 25 kg/m² as shown in multivariable regression analysis. This result implies that patients with higher pre-transplant KDIGO stages and BMI values ≤ 25 kg/m² profit most from ATG induction therapy.

Moreover, the logit of this multivariable logistic regression model was able in this study to predict the improvement of renal function within the first 6 months with high sensitivity and specificity (AUROC = 0,916) implicating a potentially useful clinical prognostic model which requires external validation before it is introduced into clinical practice.

The most likely causal explanation for these observations on improved kidney function in those patients with ATG induction lies in the observed fact that they had

significantly lower Tacrolimus blood levels 3 months after transplantation which is known to be associated with lower nephrotoxicity (51–53).

4.1.2 Other relevant factors for patient's prognosis

Higher pre-transplant KDIGO-stages were identified as an independently significant risk factor for rejection-free graft survival, patient survival, early graft loss and cancer-free survival.

Donor CMV IgG positivity and recipient CMV IgG negativity were revealed as independently significant protective factors for rejection free graft survival. A donor CMV IgG positive status was identified as an independently significant protective factor for patient survival and graft survival. It is interesting to note, that a similar influence of the donor CMV IgG-Status on graft survival was published recently. (54).

This study demonstrates unsurprisingly that a donor and recipient CMV IgG negative status are independent significant protective factors against CMV-positive episodes during the first month and the first 6 months after transplantation. In contrast a donor and recipient CMV-IgG positive status were identified as independent significant risk factors for CMV-PCR positive episodes during the first 6 months after transplantation.

The combined indication of Hepatitis C Virus induced cirrhosis with hepatocellular carcinoma was identified as an independently significant risk factor for early graft loss after transplantation.

As expected, higher numbers of rejection episodes during follow-up are shown here to decrease patient survival significantly.

BMI values > 25kg/m² independently increased the risk for post-transplant bronchopulmonary and/ or urinary tract infections. Patients who had a bronchopulmonary infection within the first month after transplantation showed a significantly reduced patient survival during follow up.

4.2 Methodological Limitations

The most relevant limitations of this study lie in the retrospective and unicenter nature of the investigation and the comparatively low number of investigated cases. Therefore, a major center bias cannot be ruled out in this study. A further relevant limitation is, that relevant factors for prognosis may either not yet be known and/or were not available for retrospective analysis.

Unfortunately, the follow-up time is unequally split between the ATG and no-ATG induction groups. ATG was mainly used before 2017 and so the follow-up time of these patients is longer than in those patients, who did not receive ATG induction therapy. For this reason, we decided to investigate for most end-points follow-up periods of 1 month and 6 months after transplantation, which allowed the analysis of nearly all included patients. The validity for patient survival, graft survival, cancer-free survival and rejection free survival is therefore restricted due to the dissimilar follow up times of the induction group versus the non-induction group.

Further the number of transplantations per year increased since 2017 significantly (Figure 2), and so most transplantations were performed during the last 2 years of this study.

In order to limit the effects of overfitting in multivariable regression we decided to include those variables into multivariable regression modelling which demonstrated p-values <0.200 in univariable regression analysis plus ATG induction (yes/no), because this variable is the focus of this study. This approach is established and labeled as purposeful selection of covariates in multiple regression analysis as described before. (55)

Some variables were only positive for a small percentage of analyzed patients as shown in Table 2. Therefore, the validity of those analyses that are based or rely on these variables are likely limited in their validity. Therefore, such variables were excluded from analysis which may have had an influence on the results of multivariable regression analysis.

Due to small case numbers of some liver transplant indications we grouped all indications with case numbers <4 in the group labeled as “Others”. This subgroup included Alpha-1 Antitrypsin Deficiency (AAT-deficiency), liver-trauma, non-alcoholic steatohepatitis, primary biliary cirrhosis, Budd-Chiary syndrome, cystic liver diseases, primary sclerosing cholangitis, secondary sclerosing cholangitis,

autoimmune hepatitis and acute liver failure. This grouping decision may be controversial.

ALG-induction therapy was used in the cohort analyzed in this study before the introduction of ATG-induction therapy. In this study we investigated 12 patients with ALG induction therapy and grouped them into the group without ATG-induction therapy. Due to the fact that ATG and ALG are similar medications with similar effects this decision may introduce certain bias, which is in our view likely negligible due to small case numbers.

All patients received Prograf©, which contains the active component Tacrolimus in combination with Cellcept©, which contains the active component MMF and Corticosteroids as first line therapy maintenance immunosuppression after transplantation. As an exceptional treatment, HCV positive patients received Cyclosporine A, Cellcept© (MMF) and Corticosteroids as standard treatment. These differences in maintenance immunosuppression may have introduced a further bias.

4.3 Comparison with previous work

As reported before the delayed onset of immunosuppression can be considered as safe and effective while patients are receiving ATG induction therapy (16,56–59). The results of our study confirm the value of this previously published approach.

4.3.1 Survival rates

There are only few studies on patient and graft survival in relation to induction therapy. In an analysis with 60.000 patients a significantly higher patient and graft survival rate was detected 5 years after transplantation in patients who received an induction therapy of any type. This ameliorated survival rate was significant within the first year after transplantation and remained almost the same percentage during follow up. This finding indicated an early benefit of induction therapy after transplantation (60). In another analysis with fewer case numbers there was no significant difference shown in patient and graft survival after induction therapy of any type versus no induction therapy. Only a trend towards improved graft survival rates could be shown (61). Due to high risk of bias, limited case numbers and lack of follow-up time in this previously published study and the mix of different induction therapies in the aforementioned previous studies no finally conclusive evidence on the value of ATG induction therapy after liver transplantation could be derived .This was the reason why we conducted the present study. Patient survival or graft

survival rates might differ in future investigations because of longer follow up time. Nevertheless, it is possible to say that ATG induction therapy does not harm patients or graft survival during the first months and years after transplantation. Late effects might emerge in the future but cannot be predicted at that time.

4.3.2 Cancer free survival

Because of its effects in depleting immunoregulatory cells many immunosuppressive agents are suspected to increase the rate of post-transplant lympho-proliferative diseases (PTLD). Immunosuppression with CNI showed an increased risk for developing PTLD and especially Non-Hodgkin lymphomas (NHL), so the initiation of immunosuppression with induction therapy and delayed onset and lower dosage of CNI's after transplantation needs to be evaluated if it decreases the risk for PTLD (62–64). Tacrolimus increases the risk for PTLD significantly after heart transplantation in pediatric patients compared with Cyclosporine administration alone (65). MMF does not seem to increase the risk for PTLD after kidney transplantation (66). Especially during the 1980s and 1990s when OKT3 was routinely used as induction therapy the rate of PTLDs was elevated. Since the late 1990s thymocyte depleting medication like ATG was used as a standard regimen in induction therapy after solid organ transplantations (67–69). The influence of ATG induction therapy on lymphoproliferative diseases is described as controversial.

The investigation in our study derives limited results for cancer-free survival or the evolution of PTLD due to different follow-up times of the two cohorts with and without ATG induction. The genesis of cancer or PTLD usually takes time and does not emerge within one year so this data needs to be seen critically and further investigation after longer follow-up needs to be undertaken. According to a more recent study the new regimen and lower dosages of immunosuppression does not markedly increase the risk for PTLD (29).

During the period of 2004-2013 a study of induction therapy after kidney transplantation evaluated the incidence of Non-Hodgkin Lymphoma or PTLD genesis. There was no increase in NHL with exclusion of older lymphocyte depleting agents and lower doses of ATG found (70). Generally, studies on de novo carcinoma risk associated with ATG did not find an increased risk for malignancies, but due to rarity of PTLD and NHL clinical trials are still too rare for a definitive conclusions (61).

4.3.3 Kidney function

Compromised kidney function before liver transplantation is a well-known complication of end-stage liver diseases. Pre-transplant kidney function is one of the most important factors in post-transplant renal outcome and improvement of renal function (71,72).

Studies try to figure out a therapy plan to protect or even improve renal function at the time of liver transplantation. Induction therapies try to bridge the vulnerable phase during the first days after transplantation. In a study of Dopazo et al. 2018 the difference between ATG and Basiliximab as induction therapy was investigated. In a prospective single-center study it was shown that a low-dose ATG administration as well as a Basiliximab induction therapy preserves renal function in patients with end-stage liver disease. There were no significant differences found in acute rejection episodes, renal improvement or infection rates. In this previously published study, the costs of immunosuppressive agents have been investigated too, and Basiliximab was shown to be approximately 6-fold more expensive than an ATG regimen (73). The theory behind an improved renal function is a well-functioning liver graft and the delayed initiation of CNI's. In comparison to the findings of this previously published study we show an independently significantly improvement of renal function with ATG induction therapy. Patients with compromised kidney function prior to transplantation take a demonstrable benefit out of receiving a low-dose ATG induction regimen administered. Maybe because of small case numbers (n=20) the significance of renal improvement could not be demonstrated. This significance might be revealed in greater cohort studies (73).

The different usage of immunosuppressive agents like Sirolimus or IL-2R antagonists is reported controversially regarding benefit to renal function. The only known significant fact is that the delayed initiation of the nephrotoxic CNIs including Tacrolimus reduces the risk for renal failure and improves renal outcome after transplantation (74–76). The evaluation of IL-2R antibodies in a CNI-sparing model was compared in three multicenter randomized controlled trials (RCTs). In those published studies IL-2R antibodies were administered with MMF, Corticosteroids and delayed and/or reduced Tacrolimus dosage. In one study patients had significant GFR preservation (74), in the second one there was also an improvement of renal function within one month and 6 months after transplantation when compared to a control group (77). The third study did not find any statistically

significant differences in renal function based on Daclizumab with delayed Tacrolimus initiation in patients with good renal function pretransplant (78).
The evaluation of Sirolimus in a renal sparing strategy showed in a meta-analysis based on 11 studies no significant improvement of renal function in patients with compromised kidney function within the first year post-transplant (42).
An induction therapy with ATG for 3 days, and delayed initiation of CNI's showed to have a beneficial effect on renal function immediately as well as later on during follow up after liver transplantation. The rejection rates were lower in patients receiving ATG induction therapy but patient and graft survival did not differ significantly (79).

4.3.4 Infective complications

This current study defines as infective episodes bronchopulmonary and/ or urinary tract infections. It is hard to compare infective events with other studies due to different definitions. Bronchopulmonary and urinary tract infections are very common complications during immunosuppression. Especially bronchopulmonary infections influence patient survival independently and significantly. In a previously published study there was no difference in infectious complications detectable after ATG induction therapy (73). Other studies demonstrated no significant difference in infection rates postoperatively neither by induction with rATG (79,80) nor with IL2-RA (74,77,78). According to these studies an induction therapy with delayed CNI initiation does not elevate the risk for infectious complications.

4.3.5 CMV infections

Studies investigating CMV-infections after transplantation are rare. CMV risk constellation and suggested CMV prophylaxis regimens are regarded controversially. In a study from Low et al. 2017 an induction therapy with >3.0mg/kg rATG is considered to increase the risk for CMV infections in CMV R+ liver transplant recipients during the first year with a maximum risk at 50 days post transplantation. In this current study as well as in other publications a D-/R- status is associated with the lowest risk for an CMV positive infection after transplantation. It was shown that CMV prophylaxis given for more than 6 weeks can significantly reduce CMV infections in transplant recipients who received rATG as an induction therapy (81). Compared to our study we also found a D-/R- CMV status to be an

independently significant protective factor against CMV positive infections after transplantation within the first 6 months after transplantation. A recipient positive status showed to be an independently significant risk factor for CMV-PCR positive episodes equally shown in our study. ATG induction therapy was in this study an independently significant risk factor for the emergence of CMV-PCR positivity within the first month after transplantation (OR=6.160, 95%-CI=2.410-15.741). The relation between CMV PCR positive episodes after transplantation and reduced patient survival was significant in this study. Patients who had an CMV infection during the first month after transplantation showed a significantly decreased survival during follow-up. Hence, we believe that administering a CMV prophylaxis in patients who receive ATG induction therapy is required which is even more relevant for recipient CMV positive patients. According to the study by Low et al, 2017 the administration of prophylaxis should be given for at least 6 weeks after transplantation (81).

4.4 Clinical Implications of this Study

Patients with higher pre-transplant KDIGO stages and a BMI < 25 kg/m² profit most from ATG-induction therapy leading to a higher likelihood of renal function improvement after transplantation and consecutive better patient survival.

Furthermore, all patients with ATG-induction therapy require CMV prophylaxis because these patients are at increased risk of experiencing CMV-PCR positive episodes post transplantation which diminishes patient and graft survival.

The above mentioned implications of this study are of very high clinical relevance because they have significant potential to improve patient and graft survival as well as the quality of life of our patients.

4.5 Further Studies

Studies with higher case numbers and external center participation are required to confirm or dismiss the findings of this study which may be influenced by a high center bias. Because this investigation was a retrospective analysis there are non-changeable factors which must be accepted and may relativize some results. Therefore, a prospective multicenter study would be ideal to verify or deny the findings of this study.

4.6 Conclusions

In conclusion a delayed and/or reduced dosage of Calcineurin Inhibitor administration can be safely enabled by ATG induction therapy when combined with CMV prophylaxis. Patients with compromised pre-transplant renal function with higher KDIGO stages, a BMI ≤ 25 kg/m2 and at higher risk for renal insufficiency after transplantation profit most from Thymoglobulin© induction therapy resulting in the improvement of kidney function 6 months after transplantation. Patients with Thymoglobulin© induction therapy require CMV-prophylaxis due to the increased associated risk for early CMV-PCR positive events. The optimal dosage and administration strategy of ATG induction therapy need to be evaluated in further multicenter studies.

Low-dose ATG induction therapy is a good alternative without setting patients at risk for early graft loss or reduced patient survival.

5 References

1. Mazzaferro V, Regalia E, Doci R, Andreola S, Pulvirenti A, Bozzetti F, et al. Liver transplantation for the treatment of small hepatocellular carcinomas in patients with cirrhosis. N Engl J Med. 1996;11(693–699).

2. Burra P, Burroughs A, Graziadei I, Pirenne J, Valdecasas JC, Muiesan P, et al. EASL Clinical Practice Guidelines: Liver transplantation. J Hepatol. 2016;64(2):433–85.

3. Furukawa H, Todo S. Evolution of immunosuppression in liver transplantation: Contribution of cyclosporine. Transplant Proc. 2004;36(2 SUPPL.):274–84.

4. Boillot O, Seket B, Dumortier J, Pittau G, Boucaud C, Bouffard Y, et al. Thymoglobulin induction in liver transplant recipients with a tacrolimus, mycophenolate mofetil, and steroid immunosuppressive regimen: A five-year randomized prospective study. Liver Transplant. 2009;15(11):1426–34.

5. Lancet. European Mycophenolate Mofetil Cooperative Study Group. Placebo-controlled study of mycophenolate mofetil combined with

cyclosporine and corticosteroids for prevention of acute rejection. 1995;345:1321–5.

6. Wiesner R, Edwards E, Freeman R, Harper A, Kim R, Kamath P, et al. Model for end-stage liver disease (MELD) and allocation of donor livers. Gastroenterology [Internet]. 2003 Jan;124(1):91–6. Available from: http://www.ncbi.nlm.nih.gov/pubmed/12512033

7. Habib S, Berk B, Chang C-CH, Demetris AJ, Fontes P, Dvorchik I, et al. MELD and prediction of post-liver transplantation survival. Liver Transpl [Internet]. 2006 Mar;12(3):440–7. Available from: http://www.ncbi.nlm.nih.gov/pubmed/16498643

8. Freeman RB, Gish RG, Harper A, Davis GL, Vierling J, Lieblein L, et al. Model for end-stage liver disease (MELD) exception guidelines: results and recommendations from the MELD Exception Study Group and Conference (MESSAGE) for the approval of patients who need liver transplantation with diseases not considered by the standar. Liver Transpl [Internet]. 2006 Dec;12(12 Suppl 3):S128-36. Available from: http://www.ncbi.nlm.nih.gov/pubmed/17123284

9. Préville X, Flacher M, LeMauff B, Beauchard S, Davelu P, Tiollier J, et al. Mechanisms involved in antithymocyte globulin immunosuppressive activity in a nonhuman primate model. Transplantation. 2001;71:460–8.

10. Tchervenkov J, Flemming C, Guttmann R, des Gachons G. Use of thymoglobulin induction therapy in the prevention of acute graft rejection episodes following liver transplantation. Transpl Proc. 1997;29:13S-15S.

11. Montenovo MI, Jalikis FG, Li M, Yeh M, Dick A, Hansen R, et al. Superior patient and graft survival in adult liver transplant with rabbit antithymocyte globulin induction: Experience with 595 patients. Exp Clin Transplant. 2017;15(4):425–31.

12. Storb R, Gluckman E, Thomas E, Buckner C, Clift R, Fefer, A. et al. Treatment of established human graft-versus-host disease by antithymocyte globulin. Blood. 1974;44:56–75.

13. Beiras-Fernandes A, Thein E, Chappel D, Gallego R, Fernandez-Roel D, Kemming G, et al. Polyclonal anti-thymocyte globulins influence apoptosis in reperfused tissues after ischaemia in a non-human primate model. Transpl Int. 2004;17:453–7.

14. Filo R, Smith E, Leapman S. Reversal of acute allograft rejection with adjunctive ATG therapy. Transpl Proc. 1981;13:482–90.

15. Zietse R, Van Steenberge E, Hesse C, Vaessen L, Ijzermans J, Weimar W. Single-shot, high-dose rabbit ATG for rejection prophylaxis after kidney transplanation. Transpl Int. 1993;6:337–40.

16. Tector AJ, Fridell JA, Mangus RS, Shah A, Milgrom M, Kwo P, et al. Promising early results with immunosuppression using rabbit anti-thymocyte globulin and steroids with delayed introduction of tacrolimus in adult liver transplant recipients. Liver Transplant. 2004;10(3):404–7.

17. Starzl TE, Murase N, Abu-Elmagd K, Gray EA, Shapiro R, Eghtesad B, et al. Tolerogenic immunosuppression for organ transplantation. Lancet (London, England) [Internet]. 2003 May 3;361(9368):1502–10. Available from: http://www.ncbi.nlm.nih.gov/pubmed/12737859

18. Platz KP, Mueller AR, Blumhardt G, Bachmann S, Bechstein WO, Kahl A, et al. Nephrotoxicity following orthotopic liver transplantation. A comparison between cyclosporine and FK506. Transplantation [Internet]. 1994 Jul 27;58(2):170–8. Available from: http://www.ncbi.nlm.nih.gov/pubmed/7518975

19. Pawarode A, Fine DM, Thuluvath PJ. Independent risk factors and natural history of renal dysfunction in liver transplant recipients. Liver Transpl [Internet]. 2003 Jul;9(7):741–7. Available from: http://www.ncbi.nlm.nih.gov/pubmed/12827563

20. Paramesh AS, Roayaie S, Doan Y, Schwartz ME, Emre S, Fishbein T, et al. Post-liver transplant acute renal failure: factors predicting development of end-stage renal disease. Clin Transplant [Internet]. 2004 Feb;18(1):94–9. Available from: http://www.ncbi.nlm.nih.gov/pubmed/15108777

21. Campbell MS, Kotlyar DS, Brensinger CM, Lewis JD, Shetty K, Bloom RD, et al. Renal function after orthotopic liver transplantation is predicted by duration of pretransplantation creatinine elevation. Liver Transpl [Internet]. 2005 Sep;11(9):1048–55. Available from: http://www.ncbi.nlm.nih.gov/pubmed/16123966

22. Bilbao I, Charco R, Balsells J, Lazaro JL, Hidalgo E, Llopart L, et al. Risk factors for acute renal failure requiring dialysis after liver transplantation. Clin Transplant [Internet]. 1998 Apr;12(2):123–9. Available from: http://www.ncbi.nlm.nih.gov/pubmed/9575400

23. Lebrón Gallardo M, Herrera Gutierrez ME, Seller Pérez G, Curiel Balsera E, Fernández Ortega JF, Quesada García G. Risk factors for renal dysfunction in the postoperative course of liver transplant. Liver Transpl [Internet]. 2004 Nov;10(11):1379–85. Available from: http://www.ncbi.nlm.nih.gov/pubmed/15497160

24. Popow I, Leitner J, Grabmeier-Pfistershammer K, Majdic O, Zlabinger GJ, Kundi M, et al. A comprehensive and quantitative analysis of the major specificities in rabbit antithymocyte globulin preparations. Am J Transplant. 2013;13(12):3103–13.

25. Hirose R, Roberts J, Quan D, Osorio R, Freise C, Ascher N et al. Experience with daclizumab inliver transplantation: renal transplant dosing without calcineurin inhibitors in insufficient to prevent acute rejection in liver transplantation. Transplantation. 2000;69:307–11.

26. De Pietri L, Serra V, Preziosi G, Rompianesi G, Begliomini B. Perioperative effects of high doses of intraoperative thymoglobulin induction in liver transplantation. World J Transplant. 2015;5(4):320–8.

27. Tchervenkov J, Flemming C, Guttman R, des Gachons G. Use of thymoglobulin induction therapy in the prevention of acute graft rejection episodes following liver transplantation. Transpl Proc. 1997;29:13–5.

28. Bogetti D, Sankary HN, Jarzembowski TM, Manzelli A, Knight PS, Thielke J, et al. Thymoglobulin induction protects liver allografts from

ischemia/reperfusion injury. Clin Transplant. 2005;19:507–11.

29. Hertig A, Zuckermann A. Rabbit antithymocyte globulin induction and risk of post-transplant lymphoproliferative disease in adult and pediatric solid organ transplantation: An update. Vol. 32, Transplant Immunology. 2015. p. 179–87.

30. Eason JD. The role of antibody induction in liver transplantation. Curr Opin Organ Transplant [Internet]. 2007;12(3):242–4. Available from: http://www.ncbi.nlm.nih.gov/pubmed/27711011

31. Eason JD, Nair S, Cohen AJ, Blazek JL, Loss GE. Steroid-free liver transplantation using rabbit antithymocyte globulin and early tacrolimus monotherapy. Transplantation. 2003;75(8):1396–9.

32. De Ruvo N, Cucchetti A, Lauro A, Masetti M, Cautero N, Di Benedetto F, et al. Preliminary results of a “prope” tolerogenic regimen with thymoglobulin pretreatment and hepatitis C virus recurrence in liver transplantation. Transplantation [Internet]. 2005 Jul 15;80(1):8–12. Available from: http://www.ncbi.nlm.nih.gov/pubmed/16003226

33. Starzl T, Klintmalm G, Porter K et al. Liver transplantation with the use of cyclosporin A and prednisone. N Engl J Med. 1981;305:266.

34. Iwatsuki S, Starzl TE, Todo S, Gordon RD, Esquivel CO, Tzakis AG, et al. Experience in 1,000 liver transplants under cyclosporine-steroid therapy: a survival report. Transplant Proc [Internet]. 1988 Feb;20(1 Suppl 1):498–504. Available from: http://www.ncbi.nlm.nih.gov/pubmed/3279643

35. European FK506 Multicentre Liver Study Group. Randomised trial comparing tacrolimus (FK506) and cyclosporin in prevention of liver allograft rejection. European FK506 Multicentre Liver Study Group. Lancet (London, England) [Internet]. 1994 Aug 13;344(8920):423–8. Available from: http://www.ncbi.nlm.nih.gov/pubmed/7520105

36. U.S. Multicenter FK506 Liver Study Group. A comparison of tacrolimus (FK 506) and cyclosporine for immunosuppression in liver transplantation. N

Engl J Med [Internet]. 1994;331(17):1110–5. Available from: http://www.ncbi.nlm.nih.gov/pubmed/7523946

37. de Mattos AM, Olyaei AJ, Bennett WM. Nephrotoxicity of immunosuppressive drugs: long-term consequences and challenges for the future. Am J Kidney Dis [Internet]. 2000 Feb;35(2):333–46. Available from: http://www.ncbi.nlm.nih.gov/pubmed/10676738

38. McCauley J, Van Thiel DH, Starzl TE, Puschett JB. Acute and chronic renal failure in liver transplantation. Nephron [Internet]. 1990;55(2):121–8. Available from: http://www.ncbi.nlm.nih.gov/pubmed/2362625

39. Lynn M, Abreo K, Zibari G, McDonald J. End-stage renal disease in liver transplants. Clin Transplant [Internet]. 2001;15 Suppl 6:66–9. Available from: http://www.ncbi.nlm.nih.gov/pubmed/11903391

40. Cantarovich M, Tzimas GN, Barkun J, Deschênes M, Alpert E, Tchervenkov J. Efficacy of mycophenolate mofetil combined with very low-dose cyclosporine microemulsion in long-term liver-transplant patients with renal dysfunction. Transplantation [Internet]. 2003 Jul 15;76(1):98–102. Available from: http://www.ncbi.nlm.nih.gov/pubmed/12865793

41. Kaltenborn A, Schrem H. Mycophenolate mofetil in liver transplantation: a review. Ann Transplant [Internet]. 2013 Dec 18;18:685–96. Available from: http://www.ncbi.nlm.nih.gov/pubmed/24346057

42. Asrani SK, Leise MD, West CP, Murad MH, Pedersen RA, Erwin PJ, et al. Use of sirolimus in liver transplant recipients with renal insufficiency: a systematic review and meta-analysis. Hepatology [Internet]. 2010 Oct;52(4):1360–70. Available from: http://www.ncbi.nlm.nih.gov/pubmed/20815021

43. Murgia M, Jordan S, Kahan B. The side effect profile of sirolimus: a phase I study in quiescent cyclosporine-prednisone-treated renal transplant patients. Kidney Int. 1996;49:209–16.

44. McAlister V, Peltekian K, Malatjalian D, Colohan S, MacDonald S, Bitter-

Suermann H et al. Orthotopic liver transplantation using low-dose tacrolimus and sirolimus. Liver Transpl. 2001;7:701–8.

45. Dunkelberg J, Trotter J, Wachs M, Bak T, Kugelmas M, Steinberg T et al. Sirolimus as primary immunosuppression in liver transplantation is not associated with hepatic artery or wound complications. Liver Transl. 2003;9:463–8.

46. McKenna GJ, Trotter JF. Sirolimus – It doesn't deserve its bad Rap(a). J Hepatol [Internet]. 2012 Jan;56(1):285–7. Available from: https://linkinghub.elsevier.com/retrieve/pii/S0168827811005150

47. Stevens PE, Levin A, Kidney Disease: Improving Global Outcomes Chronic Kidney Disease Guideline Development Work Group Members. Evaluation and management of chronic kidney disease: synopsis of the kidney disease: improving global outcomes 2012 clinical practice guideline. Ann Intern Med [Internet]. 2013 Jun 4;158(11):825–30. Available from: http://www.ncbi.nlm.nih.gov/pubmed/23732715

48. Levey AS, Stevens LA, Schmid CH, Zhang YL, Castro AF, Feldman HI, et al. A new equation to estimate glomerular filtration rate. Ann Intern Med [Internet]. 2009 May 5;150(9):604–12. Available from: http://www.ncbi.nlm.nih.gov/pubmed/19414839

49. Hanley JA, McNeil BJ. The meaning and use of the area under a receiver operating characteristic (ROC) curve. Radiology [Internet]. 1982 Apr;143(1):29–36. Available from: http://www.ncbi.nlm.nih.gov/pubmed/7063747

50. Reichert B, Kaltenborn A, Goldis A, Schrem H. Prognostic limitations of the Eurotransplant-Donor Risk Index in liver transplantation. J Negat Results Biomed [Internet]. 2013 Dec 24;12:18. Available from: http://www.ncbi.nlm.nih.gov/pubmed/24365258

51. Nankivell BJ, P'Ng CH, O'Connell PJ, Chapman JR. Calcineurin Inhibitor Nephrotoxicity Through the Lens of Longitudinal Histology: Comparison of Cyclosporine and Tacrolimus Eras. Transplantation [Internet]. 2016

Aug;100(8):1723–31. Available from: http://www.ncbi.nlm.nih.gov/pubmed/27306529

52. Naesens M, Kuypers DRJ, Sarwal M. Calcineurin inhibitor nephrotoxicity. Clin J Am Soc Nephrol [Internet]. 2009 Feb;4(2):481–508. Available from: http://www.ncbi.nlm.nih.gov/pubmed/19218475

53. Myers BD, Ross J, Newton L, Luetscher J, Perlroth M. Cyclosporine-associated chronic nephropathy. N Engl J Med [Internet]. 1984 Sep 13;311(11):699–705. Available from: http://www.ncbi.nlm.nih.gov/pubmed/6382005

54. Dogan N, Hüsing-Kabar A, Schmidt HH, Cicinnati VR, Beckebaum S, Kabar I. Acute allograft rejection in liver transplant recipients: Incidence, risk factors, treatment success, and impact on graft failure. J Int Med Res [Internet]. 2018 Sep;46(9):3979–90. Available from: http://www.ncbi.nlm.nih.gov/pubmed/29996675

55. Hosmer DW, Lemeshow S, Sturdivant RX. Applied Logistic Regression (Wiley Series in Probability and Statistics). 3rd ed. Wiley; 2013. 528 p.

56. Tchervenkov JI, Tzimas GN, Cantarovich M, Barkun JS, Metrakos P. The impact of thymoglobulin on renal function and calcineurin inhibitor initiation in recipients of orthotopic liver transplant: A retrospective analysis of 298 consecutive patients. In: Transplantation Proceedings. 2004. p. 1747–52.

57. Knight RJ, Kerman RH, Schoenberg L, Podder H, Van Buren CT, Katz S, et al. The selective use of basiliximab versus thymoglobulin in combination with sirolimus for cadaveric renal transplant recipients at low risk versus high risk for delayed graft function. Transplantation [Internet]. 2004 Sep 27;78(6):904–10. Available from: http://www.ncbi.nlm.nih.gov/pubmed/15385812

58. Shaffer D, Langone A, Nylander WA, Goral S, Kizilisik AT, Helderman JH. A pilot protocol of a calcineurin-inhibitor free regimen for kidney transplant recipients of marginal donor kidneys or with delayed graft function. Clin Transplant [Internet]. 2003;17 Suppl 9:31–4. Available from:

http://www.ncbi.nlm.nih.gov/pubmed/12795665

59. Eason JD, Loss GE, Blazek J, Nair S, Mason AL. Steroid-free liver transplantation using rabbit antithymocyte globulin induction: Results of a prospective randomized trial. Liver Transplant. 2001;7(8):693–7.

60. Moonka DK, Kim D, Kapke A, Brown KA, Yoshida A. The influence of induction therapy on graft and patient survival in patients with and without hepatitis C after liver transplantation. Am J Transplant [Internet]. 2010 Mar;10(3):590–601. Available from: http://www.ncbi.nlm.nih.gov/pubmed/19958339

61. Penninga L, Wettergren A, Wilson CH, Chan A-W, Steinbrüchel DA, Gluud C. Antibody induction versus placebo, no induction, or another type of antibody induction for liver transplant recipients. Cochrane database Syst Rev [Internet]. 2014 Jun 5;(6):CD010253. Available from: http://www.ncbi.nlm.nih.gov/pubmed/24901467

62. Opelz G, Henderson R. Incidence of non-Hodgkin lymphoma in kidney and heart transplant recipients. Lancet (London, England) [Internet]. 1993;342(8886–8887):1514–6. Available from: http://www.ncbi.nlm.nih.gov/pubmed/7902900

63. van Leeuwen MT, Grulich AE, Webster AC, McCredie MRE, Stewart JH, McDonald SP, et al. Immunosuppression and other risk factors for early and late non-Hodgkin lymphoma after kidney transplantation. Blood [Internet]. 2009 Jul 16;114(3):630–7. Available from: http://www.ncbi.nlm.nih.gov/pubmed/19443660

64. Sampaio MS, Cho YW, Shah T, Bunnapradist S, Hutchinson I V. Association of immunosuppressive maintenance regimens with posttransplant lymphoproliferative disorder in kidney transplant recipients. Transplantation [Internet]. 2012 Jan 15;93(1):73–81. Available from: http://www.ncbi.nlm.nih.gov/pubmed/22129761

65. Dayton JD, Richmond ME, Weintraub RG, Shipp AT, Orjuela M, Addonizio LJ. Role of immunosuppression regimen in post-transplant

lymphoproliferative disorder in pediatric heart transplant patients. J Heart Lung Transplant [Internet]. 2011 Apr;30(4):420–5. Available from: http://www.ncbi.nlm.nih.gov/pubmed/21147001

66. Robson R, Cecka JM, Opelz G, Budde M, Sacks S. Prospective registry-based observational cohort study of the long-term risk of malignancies in renal transplant patients treated with mycophenolate mofetil. Am J Transplant [Internet]. 2005 Dec;5(12):2954–60. Available from: http://www.ncbi.nlm.nih.gov/pubmed/16303010

67. Dharnidharka VR, Sullivan EK, Stablein DM, Tejani AH, Harmon WE, North American Pediatric Renal Transplant Cooperative Study (NAPRTCS). Risk factors for posttransplant lymphoproliferative disorder (PTLD) in pediatric kidney transplantation: a report of the North American Pediatric Renal Transplant Cooperative Study (NAPRTCS). Transplantation [Internet]. 2001 Apr 27;71(8):1065–8. Available from: http://www.ncbi.nlm.nih.gov/pubmed/11374404

68. Dharnidharka VR, Tejani AH, Ho P-L, Harmon WE. Post-transplant lymphoproliferative disorder in the United States: young Caucasian males are at highest risk. Am J Transplant [Internet]. 2002 Nov;2(10):993–8. Available from: http://www.ncbi.nlm.nih.gov/pubmed/12482154

69. Meier-Kriesche H-U, Li S, Gruessner RWG, Fung JJ, Bustami RT, Barr ML, et al. Immunosuppression: evolution in practice and trends, 1994-2004. Am J Transplant [Internet]. 2006;6(5 Pt 2):1111–31. Available from: http://www.ncbi.nlm.nih.gov/pubmed/16613591

70. Opelz G, Unterrainer C, Süsal C, Döhler B. Efficacy and safety of antibody induction therapy in the current era of kidney transplantation. Nephrol Dial Transplant. 2016;31(10):1730–8.

71. Ruebner RL, Reese PP, Abt PL. Donation after cardiac death liver transplantation is associated with increased risk of end-stage renal disease. Transpl Int [Internet]. 2014 Dec;27(12):1263–71. Available from: http://www.ncbi.nlm.nih.gov/pubmed/25070497

72. Fabrizi F, Dixit V, Martin P, Messa P. Pre-transplant kidney function predicts chronic kidney disease after liver transplant: meta-analysis of observational studies. Dig Dis Sci [Internet]. 2011 May;56(5):1282–9. Available from: http://www.ncbi.nlm.nih.gov/pubmed/21221799

73. Dopazo C, Charco R, Caralt M, Pando E, Lázaro JL, Gómez-Gavara C, et al. Low Total Dose of Anti-Human T-Lymphocyte Globulin (ATG) Guarantees a Good Glomerular Filtration Rate after Liver Transplant in Recipients with Pretransplant Renal Dysfunction. Can J Gastroenterol Hepatol. 2018;2018.

74. Neuberger JM, Mamelok RD, Neuhaus P, Pirenne J, Samuel D, Isoniemi H, et al. Delayed introduction of reduced-dose tacrolimus, and renal function in liver transplantation: the “ReSpECT” study. Am J Transplant [Internet]. 2009 Feb;9(2):327–36. Available from: http://www.ncbi.nlm.nih.gov/pubmed/19120077

75. Gerhardt T, Terjung B, Knipper P, Palmedo H, Woitas RP, Kalff J, et al. Renal impairment after liver transplantation - a pilot trial of calcineurin inhibitor-free vs. calcineurin inhibitor sparing immunosuppression in patients with mildly impaired renal function after liver transplantation. Eur J Med Res [Internet]. 2009 May 14;14(5):210–5. Available from: http://www.ncbi.nlm.nih.gov/pubmed/19541578

76. Saner FH, Cicinnati VR, Sotiropoulos G, Beckebaum S. Strategies to prevent or reduce acute and chronic kidney injury in liver transplantation. Liver Int [Internet]. 2012 Feb;32(2):179–88. Available from: http://doi.wiley.com/10.1111/j.1478-3231.2011.02563.x

77. Yoshida EM, Marotta PJ, Greig PD, Kneteman NM, Marleau D, Cantarovich M, et al. Evaluation of renal function in liver transplant recipients receiving daclizumab (Zenapax), mycophenolate mofetil, and a delayed, low-dose tacrolimus regimen vs. a standard-dose tacrolimus and mycophenolate mofetil regimen: a multicenter randomized clinic. Liver Transpl [Internet]. 2005 Sep;11(9):1064–72. Available from: http://www.ncbi.nlm.nih.gov/pubmed/16123958

78. Calmus Y, Kamar N, Gugenheim J, Duvoux C, Ducerf C, Wolf P, et al. Assessing renal function with daclizumab induction and delayed tacrolimus introduction in liver transplant recipients. Transplantation [Internet]. 2010 Jun 27;89(12):1504–10. Available from: http://www.ncbi.nlm.nih.gov/pubmed/20495510

79. Soliman T, Hetz H, Burghuber C, Györi G, Silberhumer G, Steininger R, et al. Short-term induction therapy with anti-thymocyte globulin and delayed use of calcineurin inhibitors in orthotopic liver transplantation. Liver Transplant. 2007;13(7):1039–44.

80. Bajjoka I, Hsaiky L, Brown K, Abouljoud M. Preserving renal function in liver transplant recipients with rabbit anti-thymocyte globulin and delayed initiation of calcineurin inhibitors. Liver Transpl. 2008;14:66–72.

81. Low CY, Hosseini-Moghaddam SM, Rotstein C, Renner EL, Husain S. The effect of different immunoprophylaxis regimens on post-transplant cytomegalovirus (CMV) infection in CMV-seropositive liver transplant recipients. Transpl Infect Dis. 2017;19(5):1–10.

6 Appendix

Variables			Hazard ratio	95% - CI	p-value
Pre-transplant variables	Gender female (yes/no)		0.991	0.469-1.895	0.979
	Age at transplant (years)		1.005	0.980-1.034	0.690
	Indications	Alcoholic liver disease (yes/no)	1.021	0.582-1.762	0.941
		Hepatitis C virus cirrhosis + HCC (yes/no)	1.294	0.589-2.539	0.496
		Viral liver cirrhosis (yes/no)	0.965	0.291-2.377	0.945
		Hepatocellular carcinoma (HCC) (yes/no)	1.084	0.511-2.088	0.822
		Other (yes/no)	0.695	0.286-1.445	0.351
	BMI > 25 kg/m² (yes/no)		1.028	0.595-1.781	0.922
	BMI < 19 kg/m² (yes/no)		1.295	0.313-3.560	0.677
	KDIGO stage (I-V)		**1.381**	**1.088-1.712**	**0.009**
	Donor CMV IgG pos (yes/no)		**0.428**	**0.227-0.769**	**0.004**
	Recipient CMV IgG pos (yes/no)		1.144	0.644-2.124	0.652
	Induction with ALG (yes/no)		0.928	0.277-2.318	0.887
	Induction with ATG (yes/no)		0.903	0.506-1.672	0.738

Supplementary Table 1: Shown are the results of univariable Cox regression analysis to determine significant ***pre-transplant risk factors for early death*** after transplantation. Induction with Basiliximab was not investigated due to small case numbers (n=6). Variables with p-values <0.200 are marked in bold letters.

Variables			Hazard ratio	95% - CI	p-value
Pre-transplant variables	Gender female (yes/no)		1.019	0.500-1.901	0.956
	Age at transplant (years)		1.002	0.978-1.029	0.889
	Indications	Alcoholic liver disease (yes/no)	0.922	0.530-1.571	0.678
		Hepatitis C virus cirrhosis + HCC (yes/no)	**1.677**	**0.822-3.135**	**0.147**
		Viral liver cirrhosis (yes/no)	0.919	0.277-2.258	0.870
		Hepatocellular carcinoma (HCC) (yes/no)	0.960	0.455-1.835	0.907
		Other (yes/no)	0.754	0.330-1.510	0.447
	BMI > 25 kg/m² (yes/no)		1.074	0.632-1.835	0.792
	BMI < 19 kg/m² (yes/no)		1.254	0.304-3.426	0.713
	KDIGO stage (I-V)		**3.622**	**1.092-10.628**	**0.036**
	Donor CMV IgG pos (yes/no)		**0.531**	**0.294-0.925**	**0.025**
	Recipient CMV IgG pos (yes/no)		1.278	0.726-2.357	0.404
	Induction with ALG (yes/no)		0.929	0.278-2.303	0.887
	Induction with ATG (yes/no)		0.837	0.479-1.505	0.543

Supplementary Table 2: Shown are the results of univariable Cox regression analysis to determine significant ***pre-transplant risk factors for early graft loss*** after transplantation. Induction with Basiliximab was not investigated due to small case numbers (n=6). Variables with p-values <0.200 are marked in bold letters.

Variables			Hazard ratio	95% - CI	p-value
Pre-transplant variables		Gender female (yes/no)	0.956	0.501-1.688	0.884
		Age at transplant (years)	1.010	0.988-1.035	0.404
	Indications	Alcoholic liver disease (yes/no)	1.248	0.777-1.987	0.356
		Hepatitis C virus cirrhosis + HCC (yes/no)	1.119	0.581-1.995	0.723
		Viral liver cirrhosis (yes/no)	0.556	0.169-1.350	0.215
		Hepatocellular carcinoma (HCC) (yes/no)	1.029	0.524-1.862	0.929
		Other (yes/no)	0.788	0.392-1.442	0.458
		BMI > 25 kg/m² (yes/no)	1.017	0.637-1.627	0.945
		BMI < 19 kg/m² (yes/no)	0.755	0.183-2.067	0.625
		KDIGO stage (I-V)	**1.349**	**1.084-1.646**	**0.008**
		Donor CMV IgG pos (yes/no)	**0.660**	**0.403-1.064**	**0.088**
		Recipient CMV IgG pos (yes/no)	1.132	0.693-1.909	0.628
		Induction with ALG (yes/no)	0.827	0.316-1.791	0.655
		Induction with ATG (yes/no)	1.015	0.607-1.760	0.956

Supplementary Table 3: Shown are the results of univariable Cox regression analysis to determine significant ***risk factors for cancer-free survival*** after transplantation. Induction with Basiliximab was not investigated due to small case numbers (n=6). Variables with p-values <0.200 are marked in bold letters.

Variables			Hazard ratio	95% - CI	p-value
Pre-transplant variables		Gender female (yes/no)	1.148	0.651-1.916	0.618
		Age at transplant (years)	**0.984**	**0.967-1.016**	**0.107**
	Indications	**Alcoholic liver disease (yes/no)**	**0.630**	**0.391-0.992**	**0.046**
		Hepatitis C virus cirrhosis + HCC (yes/no)	1.391	0.733-2.437	0.295
		Viral liver cirrhosis (yes/no)	**1.945**	**0.905-3.695**	**0.084**
		Hepatocellular carcinoma (HCC) (yes/no)	0.727	0.383-1.274	0.278
		Other (yes/no)	**1.538**	**0.884-2.545**	**0.123**
		BMI > 25 kg/m² (yes/no)	0.958	0.617-1.489	0.847
		BMI < 19 kg/m² (yes/no)	1.377	0.419-3.326	0.553
		KDIGO stage (I-V)	**1.249**	**1.019-1.505**	**0.033**
		D- / R-	1.253	0.687-2.140	0.444
		D- / R+	**1.511**	**0.956-2.362**	**0.072**
		D+ / R-	**0.410**	**0.171-0.829**	**0.011**
		D+ / R+	0.850	0.509-1.370	0.514
		Induction with ALG (yes/no)	1.101	0.425-2.342	0.824
		Induction with ATG (yes/no)	0.976	0.618-1.573	0.919

Supplementary Table 4: Shown are the results of univariable Cox regression analysis to determine significant ***pre-transplant risk factors for rejection-free graft survival*** after liver transplantation. Variables with p-values <0.200 are marked in bold letters.

Variables			Odds ratio	95% - CI	p-value
Pre-transplant variables		Gender female (yes/no)	1.487	0.692-3.197	0.317
		Age at transplant (years)	0.993	0.965-1.022	0.643
	Indications	Alcoholic liver disease (yes/no)	1.509	0.793-2.870	0.211
		Hepatitis C virus cirrhosis + HCC (yes/no)	1.385	0.569-3.375	0.481
		Viral liver cirrhosis (yes/no)	0.532	0.115-2.462	0.419
		Hepatocellular carcinoma (HCC) (yes/no)	**0.455**	**0.180-1.152**	**0.097**
		BMI > 25 kg/m² (yes/no)	**0.388**	**0.200-0.756**	**0.005**
		BMI < 19 kg/m² (yes/no)	2.035	0.468-8.838	0.343
		KDIGO stage (I-V)	1.099	0.805-1.500	0.557
		Donor CMV IgG pos (yes/no)	0.966	0.509-1.835	0.916
		Recipient CMV IgG pos (yes/no)	0.902	0.463-1.756	0.762
		Induction with ALG (yes/no)	**0.286**	**0.036-2.273**	**0.164**
		Induction with ATG (yes/no)	1.347	0.686-2.646	0.383

Supplementary Table 5: Shown are the results of univariable binary logistic regression analysis to determine significant risk factors for ***urinary tract and/or bronchopulmonary infections within the first month after transplant***. Induction with Basiliximab was not investigated due to small case numbers (n=6). Variables with p-values <0.200 are marked in bold letters. Variables with independent significant influences are marked in bold letters.

Variables			Odds ratio	95% - CI	p-value
Pre-transplant variables		Gender female (yes/no)	1.540	0.715-3.316	0.278
		Age at transplant (years)	1.014	0.982-1.046	0.389
	Indications	Alcoholic liver disease (yes/no)	1.023	0.533-1.967	0.945
		Hepatitis C virus cirrhosis + HCC (yes/no)	1.430	0.586-3.488	0.440
		Viral liver cirrhosis (yes/no)	1.990	0.634-6.248	0.254
		Hepatocellular carcinoma (HCC) (yes/no)	**0.706**	**0.303-1.642**	**0.048**
		BMI > 25 kg/m² (yes/no)	0.793	0.416-1.512	0.481
		BMI < 19 kg/m² (yes/no)	**3.591**	**0.863-14.940**	**0.087**
		KDIGO stage (I-V)	**1.434**	**1.058-1.942**	**0.021**
		Recipient CMV IgG pos (yes/no)	**2.860**	**1.297-6.303**	**0.005**
		Induction with ALG (yes/no)	1.141	0.296-4.392	0.850
		Induction with ATG (yes/no)	**4.751**	**2.012-11.215**	**<0.001**
		D- / R-	**0.079**	**0.010-0.592**	**0.0003**
		D- / R+	1.538	0.790-2.995	0.208
		D+ / R-	0.917	0.389-2.165	0.843
		D+ / R+	**1.665**	**0.841-3.294**	**0.148**

Supplementary Table 6: Shown are the results of univariable binary logistic regression analysis to determine significant risk factors for ***CMV PCR positive episodes within the first month after transplant***. Induction with Basiliximab was not investigated due to small case numbers (n=6). Variables with p-values <0.200 are marked in bold letters.

Variables			Odds ratio	95% - CI	p-value
Pre-transplant variables		**Gender female (yes/no)**	**1.668**	**0.836-3.328**	**0.149**
		Age at transplant (years)	1.006	0.980-1.033	0.648
	Indications	Alcoholic liver disease (yes/no)	1.314	0.745-2.318	0.346
		Hepatitis C virus cirrhosis + HCC (yes/no)	1.147	0.507-2.595	0.743
		Viral liver cirrhosis (yes/no)	1.814	0.612-5.383	0.286
		Hepatocellular carcinoma (HCC) (yes/no)	**0.588**	**0.583-1.224**	**0.147**
		BMI > 25 kg/m² (yes/no)	0.825	0.471-1.448	0.503
		BMI < 19 kg/m² (yes/no)	1.767	0.429-7.277	0.433
		KDIGO stage (I-V)	**1.272**	**0.961-1.683**	**0.091**
		Induction with ALG (yes/no)	0.563	0.148-2.146	0.382
		D- / R-	**0.125**	**0.037-0.425**	**<0.001**
		D- / R+	9.61	0.530-1.741	0.895
		D+ / R-	1.063	0.511-2.212	0.871
		D+ / R+	**2.752**	**1.492-5.078**	**0.001**
		Induction with ATG (yes/no)	**3.132**	**1.657-5.918**	**<0.001**

Supplementary Table 7: Shown are the results of univariable binary logistic regression analysis to determine significant risk factors for ***CMV PCR positive episodes within the first 6 months after transplant***. Variables with p-values <0.200 are marked in bold letters.

Variables			Odds ratio	95% - CI	p-value
Pre-transplant variables		**Gender female (yes/no)**	**2.000**	**0.791-5.059**	**0.156**
		Age at transplant (years)	0.994	0.958-1.031	0.754
	Indications	Alcoholic liver disease (yes/no)	0.706	0.296-1.683	0.426
		Hepatitis C virus cirrhosis + HCC (yes/no)	0.927	0.254-3.385	0.908
		Viral liver cirrhosis (yes/no)	0.520	0.064-4.207	0.506
		Hepatocellular carcinoma (HCC) (yes/no)	0.840	0.295-2.394	0.745
		BMI > 25 kg/m² (yes/no)	**0.362**	**0.148-0.890**	**0.022**
		BMI < 19 kg/m² (yes/no)	**3.261**	**0.565-18.825**	**0.186**
		KDIGO stage (I-V)	**3.559**	**2.218-5.713**	**<0.001**
		Donor CMV IgG pos (yes/no)	**0.563**	**0.240-1.318**	**0.186**
		Recipient CMV IgG pos (yes/no)	**1.871**	**0.709-4.936**	**0.188**
		Induction with ALG (yes/no)	Instable regression due to low case number (n=11) and no patients with ALG treatment who improved their KDIGO stage		
		Induction with ATG (yes/no)	**3.911**	**1.286-11.898**	**0.007**
		Infections within the first 6 months after transplant	0.916	0.343-2.448	0.860

Supplementary Table 8: Shown are the results of univariable binary logistic regression analysis to determine significant prognostic factors for an ***improvement of the KDIGO-stage of kidney function 6 months after transplantation in comparison to the pre-transplant KDIGO-stage***. Induction with Basiliximab was not investigated due to small case numbers (n=6). Variables with p-values <0.200 are marked in bold letters.

6.1 Further Publication

The content of this diploma thesis will we published in an appropriate journal.