Oslo, January 2019

# LIVER TRANSPLANTATION IN ADULTS BEYOND ESTABLISHED DONOR- AND RECIPIENT CRITERIA

Trygve Thorsen, MD

Section for Transplantation Surgery

Department of Transplantation Medicine

Oslo University Hospital, Rikshospitalet

Oslo, Norway

Oslo University Hospital

UiO : Institute of Clinical Medicine
University of Oslo

*Series of dissertations submitted to the*
*Faculty of Medicine, University of Oslo*

ISBN 978-82-8377-420-7



Cover: Hanne Baadsgaard Utigard.
Print production: Reprosentralen, University of Oslo.

## TABLE OF CONTENTS

## AKNOWLEDGEMENTS

The present work has been performed at the Section for Transplantation Surgery, Department of Transplantation Medicine at Oslo University Hospital, Rikshospitalet.

Firstly, I would like to thank my former chief and main supervisor Dr Aksel Foss for strongly encouraging me to engage in a PhD program. This project would certainly not have been started if it wasn't for his initiative and hard-working abilities. However, my most important co-worker and contributor during this work has without doubt been my current main supervisor and colleague Dr Einar Martin Aandahl. His broad knowledge, excellent writing-capabilities and enthusiasm are truly impressive. At times when encountering scientific challenges and seemingly insurmountable feedback from reviewers have made life miserable, his comforting abilities and research skills have been invaluable. I could not ask for a better supervisor. Although probably not aware of the arrangement while taking place, I would also like to thank his wife Cecilie for letting him spend most of his last paternity leave finalizing the two first papers in this thesis. Also, I wish to express my gratitude to my co-supervisor Prof Tom Hemming Karlsen. His high-quality feedback and comments on this project have always been very helpful. Further, I would like to thank the head of our department, Prof Pål-Dag Line, for hiring me in the first place and then facilitating my working-conditions in a way that have made it possible to combine research and clinical work. Also thanks to my former leader Dr Bjarte Fosby for providing me with sufficient leave for finalizing my thesis during the autumn of 2018, and thanks to my good colleagues Rune Horneland, Bjørn Lien, Morten Hagness, Morten Skauby, Harald Hugenschmidt, Jon Solheim, Kristine Fasting, Ammar Khan, William Indrevåg, Casper Beiske and Olav Liavåg for support and taking care of my duties in the last few months when my focus was on finish the thesis rather than contributing to the daily clinical work. I also wish to thank Dr Kirsten Muri Boberg and her colleagues at the Section of

Gastroenterology for their help and cooperation in the handling and follow-up of our common liver transplant patients. Thanks to Dr Ulrika Dahlgren and Dr William Bennet in Gothenburg and all other collaborators at the Scandinavian transplant centers that have participated in the project. Thanks to Dr Knut Jørgen Labori, Department of Hepato-Pancreato-Biliary Surgery, for his important contribution to paper 3. Thanks to Dr Christian Naper at Institute of Immunology and Dr Krzysztof Grzyb at Department of Pathology for their contributions to paper 2.

Last but not least, I wish to thank my family. To my parents Oddrun and Svein for continuous support in every possible way and for taking care of the kids and dog at busy times. To my wife Linda, although perhaps not expressing enthusiasm towards my PhD project on a daily basis, this work would definitely been impossible to complete without her backing and support. I am fully aware that taking care of our great kids Vilma and Johannes at times can be a lot more challenging than the writing of any doctoral thesis.

Oslo, March 2019, Trygve Thorsen

## ABBREVIATIONS

ABCc, ABO compatible

ABOi, ABO incompatible

ACR, acute cellular rejection

AFP, alfa-fetoprotein

AIH, autoimmune hepatitis

ALD, alcoholic liver disease

ALF, acute liver failure

AMR, antibody-mediated rejection

APC, antigen presenting cell

ATG, anti-thymocyte globulin

BAR, balance of risk

BC, biliary complications

BMI, body mass index

CBD, common biliary duct

CCA, cholangiocarcinoma

CIT, cold ischemia time

CMV, cytomegalovirus

CNI, calcineurin inhibitor

CRLM, colorectal liver metastasis

CYA, cyclosporine A

DAA, direct acting antivirals

DBB, brain dead donor

DCD, donation after cardiac death

DDLT, deceased donor liver transplantation

DGF, delayed graft function

DM, diabetes mellitus

D-MELD, donor age and recipient model for end-stage liver disease

DRI, donor risk index

DSA, donor specific antibodies

ECD, extended criteria donors

ET-DRI, Eurotransplant donor risk index

GS, graft survival

GSC, glycosorb® selective columns

HAT, hepatic artery stenosis

HBV, hepatitis B virus

HCC, hepatocellular carcinoma

HCV, hepatitis C virus

HLA, human leucocyte antigen

IL-2, interleukin 2

IRI, ischemia-reperfusion injury

IS, immunosuppresives

IVC, inferior vena cava

IVIG, intravenous immunoglobulins

LDLT, living donor liver transplantation

LT, liver transplantation

MC, Milan criteria

MELD, model for end-stage liver disease

MFI, mean fluorescence intensity

MPA, mycophenolic acid

mTor, mammalian target of rapamycin

NAFLD, non-alcoholic fatty liver disease

NASH, non-alcoholic steatohepatitis

PBC, primary biliary cirrhosis

PNF, Primary nonfunction

PP, plasmapheresis

PS, patient survival

PSC, primary sclerosing cholangitis

PVT, portal vein thrombosis

RBC, red blood cells

Re-tx, retransplantation

SIRS, systemic inflammatory response syndrome

SOFT, survival outcomes following liver transplantation

SRTR, Scientific Registry of Transplant recipients

TAC, tacrolimus

TCMR, T cell-mediated rejection

UNOS, United Network for Organ Sharing

VVB, veno-venous bypass

## LIST OF PUBLICATIONS

### Paper I

**Transplantation With Livers From Deceased Donors Older Than 75 Years**

Thorsen T, Aandahl EM, Bennet W, Olausson M, Ericzon BG, Nowak G, Duraj F, Isoniemi H, Rasmussen A, Karlsen TH, Foss A.

Transplantation. 2015 Dec;99(12):2534-42. doi: 10.1097/TP.0000000000000728.

### Paper II

**Liver transplantation with deceased ABO-incompatible donors is life-saving but associated with increased risk of rejection and post-transplant complications**

Thorsen T, Dahlgren US, Aandahl EM, Grzyb K, Karlsen TH, Boberg KM, Rydberg L, Naper, Foss A1, Bennet W.

Transpl International. 2015 Jul;28(7):800-12. doi: 10.1111/tri.12552.

### Paper III

**Liver transplantation as a lifesaving procedure for posthepatectomy liver failure and iatrogenic liver injuries**

Trygve Thorsen, Jon Magnus Solheim, Knut Jørgen Labori, Pål-Dag Line, Einar Martin Aandahl.

Submitted for publication.

# 1 INTRODUCTION

From being an experimental procedure in the 1960s and 1970s, liver transplantation (LT) is now the established and often only curative treatment for a variety of diseases resulting in end-stage liver disease. Both benign liver diseases and some malignancies are established indications, and the definitive success of LT has become its greatest dilemma; in most parts of the world, there is a huge discrepancy between the rising number of patients on the waiting lists and the availability of donor organs. At the end of December 2016 approximately 5600 patients were listed for LT in Europe, while almost 20% of the patients died while waiting the same year [1]. As of September 2018, nearly 14000 patients are listed for LT in USA, and the waitlist-mortality for 2016 was 16% [1,2]. Several strategies have been explored to improve the organ availability such as using living donors and split livers. Severe complications and even fatal incidents with donor deaths have prohibited expansive use of living donors in most western countries [3-6] and in 2016, the percentage of living donors in LT was only 4.4% in USA and 2.9% in Europe[1]. Dividing a liver graft into two viable split-livers is another way of achieving more organs for LT. Unfortunately, utilization of this method is also declining in the western world, probably due to higher frequencies of complications compared to whole organ LT[7-9]. To expand the donor pool, the transplant community has been forced to explore the use of marginal donors, so called extended criteria donors (ECD). As opposed to renal transplant donors, there is no precise definition of the ECD-criteria in LT. In general, ECD-grafts are believed to be of lower than average quality and associated with worse patient outcomes or an increase in disease transmission[10]. High donor age the is single most important ECD-criteria resulting in inferior outcomes[8] and increased use of ECD-donors has significantly lowered wait-list mortality and reduced the gap in organ shortage[11]. In paper 1 of this thesis, we have explored the Scandinavian

experience using liver donors above 75 years of age and compared the results with a control group utilizing donors aged 20-49 years.

Blood group incompability (ABOi) between the donor and the recipient is regarded as a major risk factor for acute rejection, poor graft function, early graft loss and increased risk for complications after LT. In the Western world ABOi LT has mainly been used in urgent cases when no ABO compatible donors were available[12-18]. In paper 2 we have analyzed the common experience in Gothenburg and Oslo with ABOi LT.

As results and patient survival after LT constantly improved, the spectrum of LT-indications has been expanding, and limits for operability and acceptable risk have been pushed. Conditions that previously were regarded as contraindications in some parts of the world, have been regarded as acceptable at centers with better availability of donor organs. In Norway, the situation has been fortunate for a long period, with good access to donors combined with at times a very short waiting list. This has allowed exploration of new indications for LT such as colorectal liver metastasis (CRLM)[19-22], or expansion of criteria beyond what is internationally accepted for patients with hepatocellular carcinoma (HCC) [23]. In paper 3, we have investigated LT as a lifesaving procedure for patients with acute liver failure (ALF) after liver resections or after iatrogenic liver injuries. This patient group is heterogeneous, and the various indications have been little described in current medical literature.

# 2 TRANSPLANT IMMUNOLOGY

## 2.1 The immune system

The human immune system consists of the *innate immune system* and *the adaptive immune system.* The innate immune system includes both immune cells and mechanisms that involve the mucosal barriers. The innate immune cells execute an immediate, but non-specific immune response towards intruding agents, acting directly or by inducing an inflammatory response leading to recruitment of other immune cells[24]. It is a rapid immune response, initiated within minutes or hours after an encounter with a pathogen, but it does no generate immunologic memory. If the intruding agent evades the innate response, vertebrates have a second layer of protection called *the adaptive immune system,* which is activated by the innate immune response and inflammation. This system adapts its response during an infection to improve its recognition of the intruding pathogen, and targets highly specific peptide antigens presented by human leukocyte antigen (HLA) class 1 (endogenous/intracellular peptides) or HLA class 2 (exogenous/extracellular peptides) molecules[25]. The adaptive immune system has the capacity to generate immunologic memory, which enables the host to set mount a more rapid and efficient immune response upon later exposure to the same antigen. The main cell categories in the adaptive immune system are T and B cells, of which multiple subgroups exist.

All components of the human immune system are involved in the immune response against an organ transplant, but T cell dependent mechanisms are crucial in the initiation of alloreactivity towards the transplant. Most of the immunosuppressive drugs used in organ transplantation are directed towards T cells. HLA molecule variants together with the ABO blood group system represent the most important alloantigens.

There are two distinct pathways for recognition of an alloantigen (Figure 1)[26]. Through the *direct pathway* T cells are able to directly recognize intact non-self HLA molecules on the surface of donor cells. The *indirect pathway* characterizes the T cells capability to identify donor HLA molecules that have been processed and presented as peptides by self-HLA class II molecules on the surface of antigen presenting cells (APC). T cells require at least two signals to be activated and acquire effector functions. Signal 1 is generated by the interaction of the T cell receptor (TCR) with its ligand, while signal 2 is generated via an interaction between costimulatory molecules on the antigen-presenting cell and the ligands on the T cells[27]. The costimulatory signals are crucial to evoke a potent response against the allograft, and therapeutic blocking of these signals has been an area of research in the development of new immunosuppressive drugs[28].

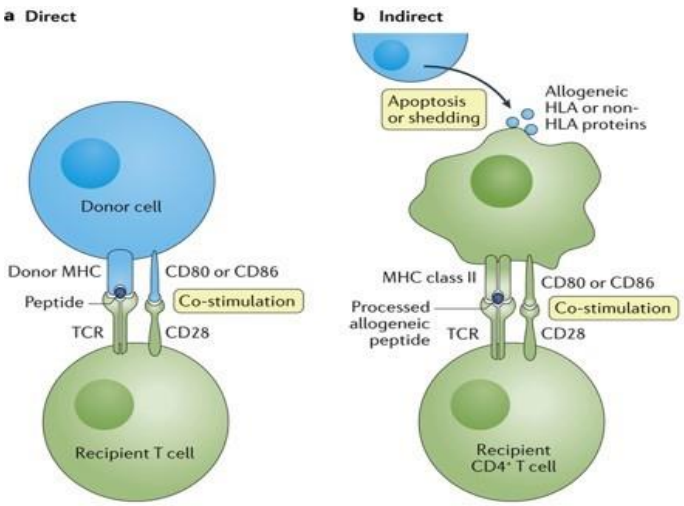



**Figure 1. a**) In the direct pathway, which is important in the early phase of allorecognition of host antigens and graft rejection, polyclonal recipient T cells recognize intact donor major histocompatibility complex (MHC) molecules directly via their T cell receptors (TCRs). **b**) By contrast, the indirect pathway is oligoclonal and dependent on a restricted set of T cells that display a specific repertoire of TCRs. These T cells recognize only a limited number of dominant peptides that are displayed on the MHC of recipient antigen-presenting cells (APCs), and they play an important part in late and chronic rejection. The indirect pathway is also responsible for the alloantibody responses seen in patients who have received organ transplants. Adapted with permission from Yang, Transplant genetics and genomics, Nature Reviews Genetics volume 18, (2017). |

## 2.2 Rejection in organ transplantation

Allograft rejection is one of leading causes of inferior graft function and graft survival (GS) in LT recipients. Improved immunosuppressive (IS) drugs and combination regimens have significantly reduced the incidence of rejection over time[29]. Liver-biopsy is required in the diagnostics of all types of rejection described below

### 2.2.1 T cell-mediated rejection (TCMR)

Most rejection-episodes after LT is considered to be T cell-mediated (TCMR), formerly known as acute cellular rejection (ACR)[30,31]. From the early days of the organ transplantation era, it has been clear that the liver has an immunological advantage compared to other organs by being more robust against rejection[31]. Tolerance after LT in humans is not that unusual, and it has been shown that 5-15% of patients can be taken of IS with no obvious damage to the liver graft[32]. Early TCMR is typically within 90 days of transplant and is characterized by inflammatory bile duct damage and portal inflammation. The incidence is between 10 to 30%, and most studies show little impact on graft and patient survival[29]. Late TCMR occurring more than 90 days post LT is observed in 7-23% of the patients, and various studies have documented association with reduced graft survival[33-36]. Risk factors include younger age, autoimmune etiology, prior episode of early TCMR, female gender and non-compliance[29]. A small percentage of the patients suffer from steroid-resistant late TCMR, of who up to 25% progress to a state of chronic rejection, resulting in potentially irreversible bile duct and/or vascular injury to the graft[37]. The incidence of chronic rejection in adults is around 1-5%, and risk factors include autoimmune etiology, cyclosporine-based IS, number and severity of TCMR episodes, retransplantation for rejection and donor/recipient sex mismatch[38,39]. Diagnostic histological

criteria include bile duct loss affecting more than 50% of the portal tracts, atrophy of the majority of bile ducts or foam cell obliterative arteriopathy[37].

### 2.2.2 Antibody-mediated rejection and donor specific antibodies

Although most rejections episodes after LT are T cell-mediated, there has been an increasing focus on antibody-mediated rejection and the impact of donor specific antibodies (DSA) in the recent years. Unlike the situation with other solid organ transplants, liver grafts seem to be less prone to be affected by antibody-mediated rejection (AMR)[40,41]. Multiple mechanisms of resistance to AMR have been proposed, including unique sinusoidal microvasculature of the liver, the ability to secrete soluble HLA, complement phagocytosis by Kupffer cells, the dual arterial and portal circulation and the ability to regenerate[42]. However, presence of persistent high level of DSA or development of de-novo HLA class 2 DSA has been associated with rejection[42] and decreased patient and graft survival[43], and evidence indicating that DSA is linked to adverse outcomes in LT continues to emerge[42].

#### 2.2.2.1 Acute AMR

Even though preformed DSA are present in nearly 20% of all liver transplant recipients[44], biopsy proven AMR is rare (<1% of all and <5% of sensitized candidates)[45-49]. AMR should be considered in cases with TCMR not responding to standard treatment. Typically, acute AMR presents with delayed peak in liver transaminases, refractory thrombocytopenia and resistance to steroid treatment. Combined AMR and TCMR is not uncommon[43]. According to the latest guidelines from the Banff Working Group on Liver Allograft Pathology, all the following criteria for a definitive diagnosis of AMR should be present: presence of DSA, diffuse C4d-positive staining, AMR-pattern of injury on biopsy and exclusion of other causes of liver

injury[44]. Initial treatment of mild AMR should be done with steroid-boluses, while treatment of moderate and severe acute AMR may include plasmapheresis (PP) and intravenous immunoglobulins (IVIG) with or without B-cell depleting agents like rituximab. Recent research in the field have focused on differentiating pathogenic from nonpathogenic DSA, as well as trying to define the threshold-values for DSA resulting in actual rejection[50]. Moderate to low DSA with Mean Fluorescence Intensity (MFI) levels <5000, in particular of HLA class 1, appear to not have any clinical significance. However, preformed HLA class 2 DSA with MFI >5000 are associated with increased risk of early TCMR, combined TCMR/AMR and potentially also AMR alone[43]. The picture is complex and several studies have shown that 95% of class 1 with MFI >5000 and 67% of class 2 with MFI>10000 pre-transplant DSA are spontaneously cleared after transplantation, and probably have no clinical significance[51-53]. C1q-fixing class 2 DSA have been showed to represent the greatest risk of initiating early rejection[54]. The clinical status of the patient and the quality of the allograft might be factors that affect the patients risk for AMR. It has been proposed that presence of DSA may play a more significant role in deceased donor LT than in the setting of living donor LT, due to more severe ischemia-reperfusion injury (IRI) and thereby increased exposure of endothelial and biliary HLA to circulating DSA[46,47].

#### 2.2.2.2 Chronic AMR

The hallmarks of chronic AMR are mild to moderate inflammation with low-grade interface activity and fibrosis seen on biopsy, positive/negative C4d-staining together with circulating DSA present at least 3 months[44]. However, the same histologic picture can also be seen in biopsies from patients with normal liver tests, making the diagnosis of chronic AMR difficult[29]. The progressive fibrosis is the most characteristic feature of chronic AMR and occurs in 8-15%

of patients who develop de-novo or persistent DSA after transplantation[49]. The majority of the cases are thought to be caused by HLA class 2 DSA against the DQ locus[55,56]. Liver tests can be normal, but slow progression with loss of bile ducts due to destruction of the supplying capillary vessels can ultimately lead to graft failure[46,56]. As for acute AMR, chronic AMR can co-exist with TCMR[44]. The threshold-level of MFI in the posttransplant setting associated with development of chronic AMR is not known but has been estimated to lie around 10 000[54]. A positive C4d-staining is firmly associated with presence of DSA, and a combination of C4d positivity on liver biopsies together with detection of class 2 DSA has been found to be the strongest predictor for inferior 5-year survival[57]. It has been suggested that there is a variable expression of HLA class 2 in the liver, and this may be the reason why some patients for periods do not develop AMR despite having circulating DSA in serum. An inflammatory insult to the liver may cause upregulation of HLA class 2 receptors, which then is targeted by DSA[43]. This phenomenon has been seen in HVC-patients with disease recurrence accompanied with accelerated fibrosis in presence of HLA class 2 DSA[58]. Currently, there is no defined treatment strategy in the setting of established chronic AMR[29], and future studies are needed both for exploring the magnitude of the problem as well as pointing out possible therapeutic interventions.

#### 2.2.2.3 AMR and ABO-incompatible LT

Antibody mediated rejection is a well-known complication associated with ABO-incompability between donor and recipient. Here, the rejection occurs secondary to preformed ABO-antibodies rather than the donor specific antibodies discussed in the sections above. Both the clinical picture, the histological features and the extent of immunological reactions diverge compared to what can be observed in “regular” DSA-induced AMR. AMR in ABOi LT usually appears as a hyper-acute rejection with a dramatic picture dominated by hepatic necrosis 1-2

weeks after transplantation, or as delayed diffuse intrahepatic biliary strictures[59], the latter usually as a more subtle and less dramatic situation although often resulting in need for re-transplantation in the long run. In contrast to regular donor specific antibodies, the ABO antibodies are naturally occurring and found universally, and they are highly reactive[60]. In addition to being expressed in variable levels on vascular endothelium, ABO antigens are also present on the surface of red blood cells, sinusoidal endothelium of the liver as well as on biliary epithelium and on a wide variety of other tissues[44,61]. Normally, adaption of the graft to the recipients ABO type occurs within 2-3 weeks after transplant[62], a process called accommodation. However, vascular and biliary epithelium of hepatic allografts may continue to express donor blood group antigens up to 150 days after transplant[63].

Criteria for diagnosis of acute AMR in ABOi LT are included in the latest guidelines from The Banff Working Group on Liver Allograft Pathology where the histologic picture differs somewhat from regular AMR with edema and periportal hepatocyte necrosis being more prominent in ABOi AMR[44]. Further aspects regarding ABO incompatible liver transplantation are discussed under section **3.6.2.**

## 2.3 Immunosuppression in current use

The continuous improvement in graft and patient survival after LT is associated with many factors including the efficacy of immunosuppression. Effective IS management is a key factor in achieving optimal results after LT. Even though there is evidence that some liver recipients that have stopped taking IS-medication still maintaining allograft function and seemingly have developed immunological tolerance[30], the vast majority of patients will need life-long treatment with IS to avoid allograft rejection and the associated complications[64]. IS in LT can broadly be divided into the induction phase and the maintenance phase, as well as eventual resumption of

these phases when managing episodes of rejection[29]. Induction therapy is usually accomplished by a single high dose of intravenous corticosteroids given at time of LT. The use of other induction medication, such as interleukin 2 (IL-2) receptor antibodies or lymphocyte-depleting therapy with anti-thymocyte globulin (ATG), is increasing due to need for delayed introduction of calcineurin inhibitors (CNI) in patients suffering from kidney failure at the time of LT. Induction therapy is also indicated in patients with increased immunological risk (re-transplantation, immune-mediated liver disease)[29].

The IS currently in use are targeted at either depleting, diverting or blocking of T-cells. Most LT patients are treated with a combination of 3 different drugs at time of discharge from the hospital; a CNI in combination with an antiproliferative agent and cortocosteroids[30,64]. However, the overall approach to IS varies widely between different transplant centers around the world. Figure 2 illustrates actions of the various immunosuppressive drugs in use.

CNIs, which includes cyclosporine A (CyA) and tacrolimus (TAC), have immunosuppressive effect mainly by preventing activation of T lymphocytes[65]. These drugs inhibit intracellular signal pathways in T-cells by blocking the function of calcineurin and thereby preventing production of IL-2, which is necessary for activation of T-cells[66]. Both drugs are associated with similar toxicities, although tacrolimus is regarded as more diabetogenic and have higher neurotoxicity while CyA is hampered with more renal toxicity[66]. Second to renal toxicity, the most important commons side effects of CNIs are hyperlipidemia (CyA), diabetes and hypertension. Multiple studies have proved better patient- and graft survival in patients receiving tacrolimus compared to CyA with less risk for acute rejection, thus in most centers tacrolimus is the preferred CNI[29,67].

The major antiproliferative drug in current use is the mycophenolic acid derivates (MPA). T and B lymphocytes are dependent of de-novo synthesis of purines for proliferation, and MPA exerts in immunosuppressive effect by blocking the purine-synthesis, resulting in a potent

cytostatic effect on both cell types[68]. Contrary to CNIs, MPA does not cause diabetes, hypertension of renal toxicity. It is however associated with cytopenia and gastrointestinal side effects like nausea and diarrhea, and many patients discontinue MPA due to these adverse effects[66].

Corticosteroids have been one of the major components of IS since the beginning of liver transplantation. Although the mechanisms by how the drug exerts the effects are not very well elucidated, steroids have multiple effects on the immune system. Corticosteroids causes a reduction of circulating T cells by inhibition of IL-2, impaired release from lymphoid tissue and induction of apoptosis, as well as an inhibitory effect on leukocyte adhesion and inflammatory mediators[69]. Unfortunately, steroids cause a wide range of side effects like hypertension, diabetes, osteoporosis, hypercholesterolemia, delayed wound healing and increased risk for infection[30,69]. For this reason, there has been an increasing interest in steroid reduction regimens after LT in the recent years, which has resulted in a trend towards early withdrawal and in some centers even steroid-free protocols[70]. However, patients transplanted due to autoimmune liver disease and patients with recurrent rejection episodes are less likely to be successfully withdrawn from steroids and should be kept on a small dose life-long[71].

Another class of immunosuppressive drugs is comprised of the biologic agents, which include anti-thymocyte globulin (ATG), basiliximab, daclizumab, alemtuzumab and muromomab-CD3 (OKT3). These drugs are antibodies to molecules on the cell surface. Basiliximab, an IL-2 receptor antibody, is widely used as induction therapy at time of LT. ATG causes complement-mediated lysis and depletion on circulating T cells, and is used both for induction as well as treatment for steroid-resistant rejection[66]. Cytokine release syndrome caused by ATG may lead to hypotension, fever, pulmonary edema and SIRS[72].

The last class of IS used in LT are the inhibitors of the mammalian target of rapamycin, the mTor-inhibitors. Sirolimus and everolimus are the two agents available for clinical use. mTor-

inhibitors works by blocking signals from multiple T cell surface receptors (including IL-2) resulting in suppression of cytokine-driven proliferation[73]. These agents are not linked to nephrotoxicity, diabetes or hypertension, but are associated with other serious adverse effects like thrombosis, impaired wound healing, leukopenia, anemia and mouth ulcers[74]. At the same time mTor-inhibitors have demonstrated potentially important positive effects on prevention of neoplasia, and is currently used as standard immunosuppression in patients undergoing LT for colorectal metastatic disease according to the SECA-protocol[75,76]

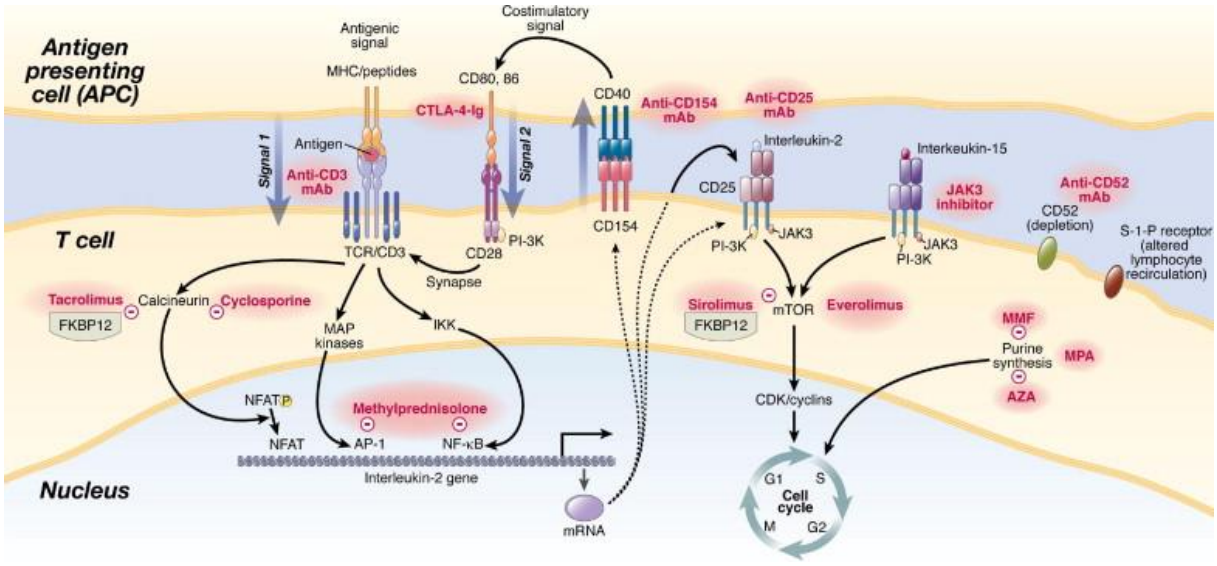



**Figure 2.** Illustrating actions of various immunosuppressive drugs in use and sites of action. Adapted with permission from Halloran, Immunosuppressive drugs for kidney transplantation, N Engl J Med. 2004 Dec 23;351(26)

# 3 LIVER TRANSPLANTATION

## 3.1 Historical background of liver transplantation

The first attempts of orthotopic liver transplantation were performed in dogs as early as 1952 by V. Staudacher at the University of Milan[77,78]. In 1955, C Stuart Welch of Albany, New York, described an auxiliary placement of a liver graft into the right paravertebral gutter of non-immunosuppressed dogs. However, the technical aspects of the operation, including the crucial need for portal blood flow, were first enlightened by Thomas Starzl in 1960[79]. In 1963, Starzl published the first report on clinical LT in humans[80]. The patient was a 3-year old boy with liver failure due to biliary atresia. Unfortunately, he died on-table of hemorrhage and coagulopathy. This first procedure was followed by six unsuccessful attempts of LT in Denver, Boston and Paris[80 81,82]. These disastrous outcomes of the first series in human LT led to a worldwide pessimism. The LT-procedure seemed too difficult and hazardous to be allowed in a clinical setting. In addition, methods of preservation were assumed inadequate for avoiding ischemic damage, and researches began to wonder if the available immunosuppressive medications were too primitive. These considerations were augmented by the fact that long-term survival had not yet been achieved in experimental animal models[83]. A moratorium in further operations lasted more than 3 years into the summer of 1967, when a 19-months old girl with hepatoma was successfully transplanted by Starzl in Denver[84]. By then, many long-term canine survivors had been achieved, some surviving more than 3 years after LT. Improvements in surgical techniques and preservation, along with introduction of immunosuppressive medications as antilymphocyte globulin used together with azathioprine and prednisone facilitated further development and successful implementation of clinical LT with long-time patient survival. In 1968, a LT unit was opened in Cambridge, UK, by Roy Calne and coworkers[85]. The book entitled “Experience in Hepatic Transplantation” from 1969 describes

the 33 first human LT, of which 25 were performed in Denver and four in Cambrigde[86]. The improved survival of patients in both USA and UK was a true proof-of-concept for liver transplantation in humans. However, the procedure was still hampered with significant mortality rates, which led to questions whether the risk was unacceptable. In 1979, the immunosuppressive medication cyclosporine became available for clinical use, which turned out to be a crucial turning point in the field of human organ transplantation. Cyclosporine in combination with prednisone or lymphocyte depleting agents had a remarkable positive effect on long-term patient and graft survival after LT[87]. With this new drug-regimen, it was now possible to achieve a 1-year survival of at least 70%, and new liver transplant programs were started at multiple hospitals worldwide in the years to come. In 1989 the new drug FK506, later know as tacrolimus, was released for clinical use. When cyclosporine was substituted with tacrolimus in LT patients, 1-year patient and graft survival was further improved as first documented from Pittsburgh[88], and later confirmed in multicenter studies from the US[89] and Europe[90]. In relative short time LT became the treatment of choice for a many different diseases causing end-stage liver failure, and even some malignant conditions.

It soon became evident that the future challenge would be the small supply of livers compared to an ever-increasing need. Figure 2 summarizes some of the most central milestones in the development of liver transplantation since the beginning in the early 50ies.

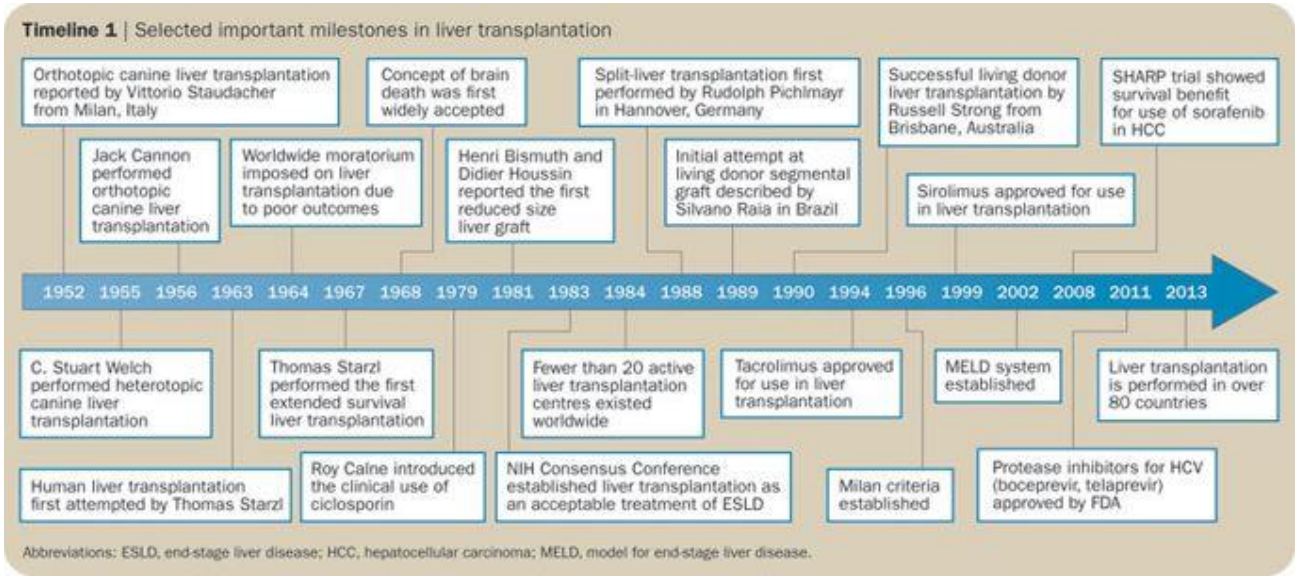



**Figure 3.** Timeline illustrating important achievements in the field of liver transplantation. Adapted with permission from Zarrinpar, Nat. Rev Gastroenterol. Hepatol. 10, (2013).

## 3.2 Surgical procedure in whole liver LT

### 3.2.1 Surgical technique

The technique has been progressively developed and refined since the start of LT in humans in 1963. In most cases, the hepatectomy is the most challenging part of the procedure, especially in patients with advanced cirrhosis or in patients that previously have undergone upper abdominal surgery, with massive bleeding being the most common complication. In the first described classic technique, the hepatectomy was performed with transverse clamping and division of the vena cava below and above the liver (IVC) with resection of the retrohepatic portion of cava. With this method, involving clamping of the portal vein and IVC, venous return to the right atrium was greatly diminished in the anhepatic phase, which typically reduced

the cardiac output by up to 50%[91]. This can lead to hemodynamic instability. As a consequence, the concept of veno-venous bypass (VVB) was developed in the late 1980s, where blood from the portal vein and IVC is routed directly to the right atrium in the anhepatic phase using a motor-driven pump and heparin-coated cannulas (Figure 4)[92].

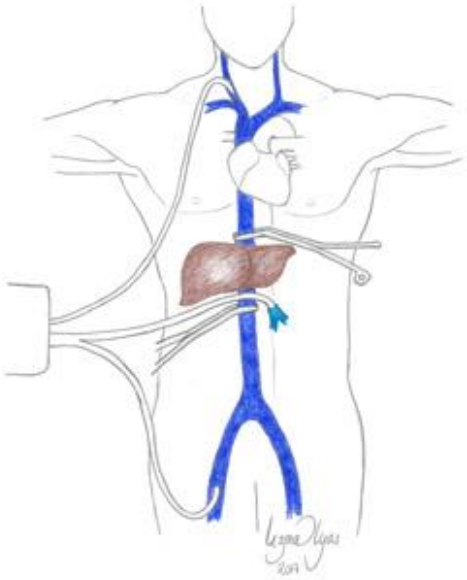


**Figure 4.** Illustration of the veno-venous circuit. Venous blood is collected from the portal vein and the inferior caval vein into the pump and re-enters the body through a cannula in the deep jugular vein, thereby by-passing the liver. Illustration by Uzma Ilyas.

VVB can be used routinely, or selectively in patients with signs of hemodynamic instability after clamping the portal vein and IVC before removing the recipients native liver[93]. However, several complications and disadvantages are related to the use of VVB, for instance longer operating time, cannula- and incision-related complications, hypothermia and hemodilution[93]. During the latest two decades, there has been a trend towards avoiding the use of VVB entirely [94,95]. The technique with preservation of the retrohepatic vena cava during hepatectomy was described as far back as 1968 by Roy Calne[85], and later revived in 1989 by Tzaki as the "piggyback" procedure[96,97]. This technique preserves caval flow during the whole procedure and therefore reduces hemodynamic instability and the corresponding negative effect on renal function. In cases where closure of the portal vein is poorly tolerated, especially in patients with metabolic disease or acute liver failure who lack portosystemic collaterals, a temporary

portocaval shunt can be constructed[98,99], and many centers use this as standard technique in LT. The evidence for renal protection using piggyback is however not strong and is still unclear if the protective effect on renal function is due to the reduction in blood loss or due to preservation of the caval flow during the procedure.

Implantation of the liver graft consists of several vascular anstomoses. With the classic technique, the cava on the graft are anastomosed end-to-end to the corresponding upper and lower caval cuffs in the recipient. If the piggy-back technique with full preservation of the recipient's cava during hepatectomy is used, the hepatic veins on the graft are anastomosed to the recipient's cava in an end-to-side manner. The two different techniques are illustrated in Figure 5.

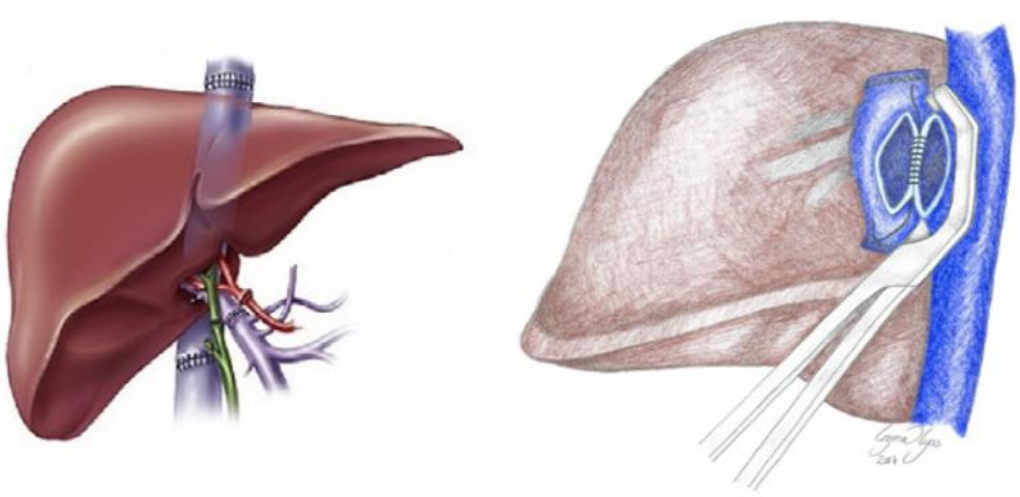


**Figure 5.** The left illustration shows the classic technique with the superior and inferior caval anastomoses. The figure on the right shows the piggy-back technique where the recipients cava is preserved, and the venous anastomoses are constructed using a part of the donor-cava as a conduit fashioning a side-to-side anastomosis. Right illustration by Uzma Ilyas.

When present, the portocaval shunt is taken down, and the portal vein on the graft is anastomosed to the patient's portal vein, and reperfusion of the graft is started. After construction of the arterial anastomosis, the biliary reconstruction is performed. This can be achieved either by a biliodigestive anastomoses or by a direct choledocho-choledocho anastomosis. Figure 6 illustrates the situation when all anastomoses are completed.

a) b)

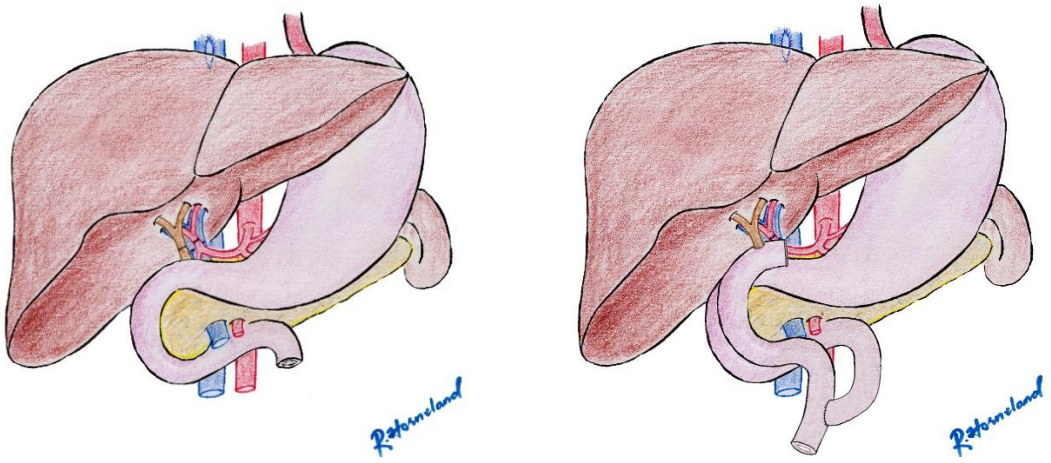


**Figure 6.** a) Illustrating the situation where the biliary tract is reconstructed a choledocho-choledochostomy or "duct-to-duct" anastomosis. b) Showing biliary reconstruction with a biliodigestive or "Roux-en-y" anastomosis where the choledochus of the donorliver is anastomosed to a jejunal loop. Illustrations by Rune Horneland.

### 3.2.2 Complications after LT

The most important surgical complications after LT includes primary nonfunction (PNF), portal vein thrombosis (PVT), hepatic artery thrombosis (HAT), hepatic outflow obstruction and biliary complications. PNF is characterized by coagulopathy, little or no biliary output, encephalopathy together with renal and multiorgan failure with rising liver enzymes and serum

lactate. The reported incidence of PNF varies from 1 to 8% and is associated with poor prognosis[100]. Hepatic artery thrombosis is the most common vascular complication after LT, where the reported incidence in large patient cohorts varies between 1.6 and 4.4 % in adults[101]. Complete and symptomatic HAT is dramatic and often requires early re-transplantation. Portal vein complications (stenosis and thrombosis) are less common and are reported to occur in 1-2% of the patients after LT[102]. Hepatic venous outflow obstruction can be caused by either an anastomotic stricture or by kinking/rotation of the graft with a reported incidence of 2,5 to 6%[103]. This problem is associated with a high risk for morbidity and mortality, and re-transplantation may be the only solution in cases where endovascular treatment with stent or surgical correction is not an option. Biliary complications continue to be a major problem in LT, with an overall incidence of 10% to 30% in current literature, most of which are stenosis at the site of or slightly proximal to the biliary anastomosis[104-106]. Although most biliary problems can be managed by early re-operation or endoscopic intervention, these complications remain a major challenge after LT causing morbidity and reduced quality of life for many patients.

Around 20% to 65% of LT patients develop acute T cell-mediated rejection (TCMR) (formerly known as acute cellular rejection (ACR))[107-109], of which 20% to 40% of the recipients develops at least one episode of TCMR that requires additional immunosuppressive treatment. However, contrary to renal transplantations where rejection is associated with long-term loss of graft function, occurrence of treated TCMR during the first 6 weeks after LT may in fact improve outcome in non-HCV patients[110,111].

## 3.3 Epidemiology

The spectrum of diseases and major indications for LT varies among different parts of the world. In the United States, chronic hepatitis C infection (HCV) remained for a long time the

major indication for LT. However, after introduction of the highly effective direct-acting antiviral agents (DAA) in late 2013 and its impact on successful HCV-treatment, alcoholic liver disease (ALD) is now the major indication for LT comprising approximately 24% of the performed liver transplant procedures in 2016. Non-alcoholic steatohepatitis (NASH), a disease closely related to over-weight and metabolic syndrome[112] is the second leading cause (19%) and HCV the third most common cause (18%)[113].

In Europe, ALD (20,6%) and HCV-related disease (decompensated cirrhosis and hepatocellular carcinoma) (20,6%) have been the two major indications during the last 10 years, followed by hepatitis B (HBV) (9,8%) and cholestatic disease (9,5%) [114,115]. However, after the advent of DAAs, there has been a dramatic decline in HCV as indication for LT in Europe, both with a reduction in HCV-cirrhosis of almost 60% and HCV-related HCC of 40%. DAA-treatment has also led to a major improve in HCV-recipient survival after LT. In the DDA-era from 2014 to 2017, the main indications in Europe were ALD (27,6 %), HCV (17,4 %) and HBV (8,4%)[115]. It is expected that HCV-related disease will continue to decline, and in the first semester of 2017 it went down to 10,6%. NASH as an indication for LT has progressively increased to 6% of all performed LTs in Europe during the same period and is expected to rise further in the years to come, although not to the same degree as in USA[116].

In Asian countries the prevalence of viral hepatitis is much higher than in the Western world, and the most common indications for LT in adults are HBV-related HCC and HBV cirrhosis, followed by HVC-related cirrhosis. Certain regions of Asia have the highest rates of HBV in the world, comprising more than 75% of the world's HBV carriers[117]. Asian regions also have the highest prevalence of HCV , and it has been estimated that >60% of the worlds HCC-cases arise in these regions[118].

In contrast, the situation and epidemiology in the Nordic countries is quite different. In the period from 2013 to 2017, HCC and primary sclerosing cholangitis (PSC) were the two leading

causes for LT accounting for 17,5% and 17,2% of the cases, respectively. The third most common indication was ALD (12.2%) followed by metabolic disease (8,7%)[119]. In 2017, 40.3% of the patients listed for LT with a primary diagnosis of HCC were also HCV-positive. After the introduction of DAA in 2013/2014, the percentage of patients listed for transplantation with HVC-cirrhosis has declined markedly.

## 3.4 Indications for LT

In general, LT should be considered as treatment for any patient suffering from liver disease where the operation can be expected to increase life expectancy beyond what the natural history of the liver disease would predict, or where the patient's quality of life is expected to be significantly improved by the procedure [120].

This includes acute liver failure, chronic liver failure, cirrhosis and some metabolic disorders which can be cured by LT. LT is also indicated for certain hepatobiliary malignancies, with HCC being the most common indication. Some of the major indications for LT are briefly summarized in the following chapter.

### 3.4.1 Acute liver failure

Acute liver failure accounts for approximately 10 % of the liver transplantations performed in both Europe and United States[66]. The most common cause of ALF in the western world is paracetamol toxicity, followed by viral hepatitis[121]. Before the advent of LT, the death rate for ALF was more than 80%. The prognosis for patients who undergo liver transplantation is very good, with 2-years survival rates of more than 92%[122]. Around 15-20% of ALF patients have no identifiable cause for their liver disease[123].

### 3.4.2 Chronic liver disease

The term “Chronic liver disease” is defined as a disease of the liver which lasts over a period of more than six months. It is comprised of a wide range of liver pathologies but can in most cases be classified according to viral, autoimmune, alcoholic or metabolic etiologies.

Worldwide, infection with HVC and HBV are among the most common causes of end-stage chronic liver disease with hepatic cirrhosis as the end result. LT has been the only option for curable treatment for these patients. However, the prognosis after LT used to be disappointing due to recurrence in the new liver. After the introduction of antiviral medication combined with prophylactic hepatitis B immunoglobulins, the outcome after LT has improved significantly[124]. The same development has occurred for HCV-patients after the advent of second generation DAA-drugs in 2013-14[115].

Auto-immune liver disease in adults is mainly comprised of patients with primary biliary cirrhosis (PBC), PSC and autoimmune hepatitis (AIH). PBC is an immune-mediated inflammatory disorder affecting small intrahepatic bile ducts that at a late stage progresses to biliary cirrhosis. One third of the patients develops to a state of decompensated cirrhosis and need for LT, after which the prognosis is excellent[125]. PSC is characterized by chronic inflammation and fibrosis of both intra- and extrahepatic bile ducts and is closely related to inflammatory bowel disease. It is a rare condition worldwide, however, individuals from Northern Europe are affected much more frequently[126]. LT is recommended in patients with late stage PSC and may also be considered with signs of dysplasia due to risk of cancer development. Outcome after LT is very good but carries a risk of more than 10% risk for recurrence after long term follow-up[125]. AIH is an autoimmune inflammatory disease of the liver, the etiology is so far unknown. Most patients respond to standard treatment with steroids or other immunosuppressive drugs, but around 10 % progress to end-stage liver cirrhosis where

LT is the only curative option[66]. Outcome after LT is good, but with a long-term risk for recurrence of about 20%[125].

Alcoholic liver disease is common, and worldwide a significant number of patients receive LT due to ALD-cirrhosis. Most centers demand at least 6-months complete abstinence from alcohol before the patients are accepted for the waiting list. However, this requirement remains controversial. In different series, the risk of relapse of drinking is estimated to be between 15-40% depending on the duration of follow-up after LT[127]. Patients with acute alcoholic hepatitis in a rapid deteriorating condition represent a dilemma as these patients very often will be unlikely to survive without LT[128,129]. However, the overall survival of patients transplanted for ALD is comparable or higher than patients transplanted for other etiologies of liver disease[130]. A recent a study of patients who underwent early LT (before 6 months of abstinence) for severe alcoholic hepatitis found a 1-year survival of 94% and 3-year survival of 84%, which is similar to patients receiving liver transplants for other indications[131].

Non-alcoholic fatty liver disease (NAFLD) and non-alcoholic steatohepatitis are becoming increasingly more common medical problems and represent the fastest growing indication for LT in the developed world. It is closely related to obesity, insulin resistance and metabolic syndrome[112]. NAFLD is a spectrum of hepatic manifestations ranging from simple steatosis to NASH with severe cirrhosis. Individuals with end-stage NASH are often morbidly obese, which has led to a significant debate regarding concomitant risk at LT in these patients[132]. Perhaps surprisingly, patient survival after LT seems to be comparable to other indications when concomitant cardiovascular disease has been excluded before transplantation[133,134]. However, there is a true risk for further weight-gain, hypertension and dyslipidemia after LT related to use of steroids and immunosuppressive drugs. Approximately 5-10% of patients that are transplanted for NASH will experience NASH-related cirrhosis of the liver graft, resulting in graft-loss in 50% of these patients[66]. Although survival after LT is good, patients with high

BMI often require longer hospital stay and have more postoperative complications such as prolonged stay in the intensive care unit and reduced wound healing[135].

Hereditary hemochromatosis, Wilson's disease and alfa1-antitrypsin deficiency are less common metabolic liver diseases that in some patients ultimately require LT.

### 3.4.3 Hepatobiliary malignancy

LT is internationally accepted as treatment of choice for certain malignant conditions. HCC is the most common hepatic malignancy. It usually develops in the setting of liver fibrosis/ cirrhosis and is therefore closely related to viral hepatitis and alcoholic liver disease. The 5-year incidence of HCC-development in a cirrhotic liver is somewhere between 10% and 20%[136]. The diagnosis of HCC is usually established by radiological imaging and tumor markers have limited clinical value. In some cases, a biopsy is needed. Small tumors can be treated by percutaneous ablation or by liver resection, which is the first option for curative treatment in a non-cirrhotic patient[137,138]. However, even in selected patients, the rate of recurrence is over 50 % at 3 years after resection. LT is indicated therapy for patients when diagnosed with HCC at an early stage, and particularly in the setting of chronic liver disease with cirrhosis. The seminal study by Mazzaferro et al from 1996 established LT as a viable treatment under certain strict circumstances where long term survival similar to benign conditions was achieved[139]. Selection of patients accepted for L was solely based on tumor size and number of nodules. Since then, many groups around the world have tried to expand these criteria, mostly at the expense of reduced long-term survival compared to the original Milan-criteria (MC)[140]. However, two recent models combining the level of alpha-fetoprotein (AFP), number of nodules and size of the largest nodule, have been shown to be better than MC in identifying patients with low risk of HCC recurrence or those who will survive for 5 years after liver transplantation[141]. LT for

HCC is undoubtedly the optimal treatment for many patients, unfortunately its widespread use is limited due to global organ shortage.

Cholangiocarcinoma (CCA) is the second most common primary hepatobiliary cancer and is associated with poor prognosis[142]. Liver transplantation in patients with unresectable hilar CCA remains a controversial subject. 5-year survival was only 5-15% in early series[143,144]. A protocol from the Mayo clinic combining strict selection with neoadjuvant chemoradiation showed promising results with over 80 % 5-year survival and low recurrence rates[145]. In the latest study published from the Mayo group, 5-year patient survival is estimated to be 65-70%[146]. Despite this, protocols for LT in CCA is still not in widespread use.

LT can be indicated for certain patients with unresectable liver metastases from neuroendocrine tumors (NET). The liver is the most common site of NET metastases, occurring in up to 85% of the cases[147]. Carcinoid syndrome most frequently occur in the presence of liver metastases and LT is a treatment option for NET-patients with unresectable liver metastases or uncontrolled symptoms. A meta-analysis from 2015 reported an overall 5-year post-transplant patient survival of only 50% in 706 patients treated with LT for metastatic NET[148]. However, smaller studies have reported much more favorable outcomes utilizing stricter selection criteria, with 5 years survival of 80-97%[149,150].

### 3.4.4 LT beyond established indications

Colorectal liver metastases have classically been considered a contraindication against LT. Colorectal cancer (CRC) is the third most common cancer in men and the second in women[151]. CRLM develop in more than 50% of these patients[152] , but only about 20% of the patients are resectable[153].

Prior to 1995 several LT for CRLMs were performed, but this was abandoned due to dismal results with a 5-year survival rates under 20%[154,155]. In the following two decades, CRLM was considered a contraindication for LT. Since then, the survival rate after liver transplantation in general has improved by almost 30%[20]. In a prospective pilot study from Oslo (The SECA study) with LT for nonresectable CRLM, a 5-year overall survival rate of 60% was demonstrated[20]. However, 19 of 21 patients experienced recurrence of disease, mostly in the form of resectable liver metastases. In later publications, the same group demonstrated that LT in nonresectable CRLM patients with extensive tumor load and progression on the last line of chemotherapy had increased survival compared with any other treatment option reported in the literature[156], and that a low-risk group of patients with unresectable CRLM had a 5-year survival after LT similar to that of patients with HCC with lesions within the Milan criteria[23]. This has led to renewed interest of the topic, and a French group has recently published their experience with similar results to what was achieved in the SECA1-study[157]. Currently, four clinical trials on liver transplantation for unresectable colorectal liver metastases are registered at https://clinicaltrials.gov[154].

Considering our institutional experience with LT for colorectal liver metastasis combined with a fortunate donor situation and short waiting list, it has been possible to explore new indications for liver transplantations beyond those established internationally. Among those are patients that have developed acute liver failure due of iatrogenic injuries during surgery, or due to remnant liver failure after previous resection surgery. Transplanting these patients may be practiced at different transplant centers around the world, but the literature describing this patient-group is scarce and, in most cases, confined to small patient-series or even single case reports. In our own series of a total of 13 rescue-patients, 7 were diagnosed with some form of cancer prior to LT of which 4 had CRLM as indication for original surgery. In paper 3 of this

thesis we have investigated the indications and outcome for this group of patients, where LT has been performed outside traditional settled criteria.

Table 1 summarizes internationally accepted indications for LT, while table 2 shows generally acknowledged contraindications to performing the procedure.

| Indications for Liver Transplantation | |
|---|---|
| **Acute Liver Failure** | **Metabolic Disorders Originating from the Liver** |
| Acute viral hepatitis | Hyperoxaluria |
| Drug or toxin induced hepatotoxicity | Familial Amyloidosis |
| Acetaminophen overdose | Urea cycle defects |
| Autoimmune hepatitis | Branched-chain amino acid disorders |
| Wilson's disease | Familial homozygous hypercholesterolemia |
| **Cirrhosis from Chronic Liver Disease** | **Malignancies** |
| Chronic viral hepatitis | Hepatocellular carcinoma |
| Alcoholic liver disease | Cholangiocarcinoma (limited) |
| Autoimmune hepatitis | Hepatoblastoma |
| Cholestatic liver disease | Fibrolamellar hepatocellular carcinoma |
| Wilson's disease | Metastatic neuroendocrine tumors |
| Hereditary and neonatal hemochromatosis | Hemangioendothelioma |
| Alpha-1-antitrypsin deficiency | **Miscellaneous** |
| Non-alcoholic steatohepatitis | Polycystic liver disease |
| Cryptogenic liver disease | Hereditary hemorrhagic telangiectasia |
| Budd-Chiari syndrome | Erythropoietic protoporphyria |
| Tyrosinemia | |
| Glycogen storage diseases | |

**Table 1.** Internationally established indications for LT**.** Adapted from www.hepatitisc.uw.edu/go/management-cirrhosis-related-complications/liver-transplantation-referral/core-concept/all

| | |
|---|---|
| *Absolute* | Severe cardiac and/or pulmonary diseases and severe pulmonary hypertension (mPAP >45 mm Hg) |
| | Alcohol addiction without motivation for alcohol abstinence and untreated/ongoing substance abuse |
| | Hepatocellular carcinoma with extrahepatic metastases |
| | Current extrahepatic malignancies (eventually reevaluation after successful therapy) |
| | Sepsis |
| *Relative* | Untreated alcohol abuse and other drug-related addiction |
| | Cholangiocellular carcinoma |
| | Hepatic metastatic neuroendocrine tumors (NET), metastatic hemangioendothelioma |
| | Morbid obesity |
| | Persistent non-adherence |

**Table 2.** Commonly accepted contraindications to performing LT. Adapted from Graziadei, Indications for liver transplantation in adults : Recommendations of the Austrian Society for Gastroenterology and Hepatology (ÖGGH) in cooperation with the Austrian Society for Transplantation, Transfusion and Genetics (ATX), Wiener klinische Wochenschrift 128(19), 2016.

## 3.5 Outcome after LT

Over the years there has been a steady improvement in the results after liver transplantation, as documented in reports from the European, American as well as Scandinavian registries[119,158,159]. According to the latest European Liver Transplant Registry (ELTR) report from 2018, the overall 1- and 5-year survival rates for patients transplanted in the period 2010-2014 are now estimated to be 86% and 74%, respectively. For comparison, the corresponding values for patients receiving LT in Europe during 1990-1994 were 75% and 64%[159]. Even though the 5-year patient survival (PS) has improved in the recent years for all indications, the most important gain in 5-year PS for LT in Europe has been seen for patients with primary liver tumors (67%), liver metastases (61%) and acute liver failure (69%)[159].

The corresponding survival rates observed in the Scandinavian Nordic Liver Transplant Registry (NLTR) shows even better results than in ELTR, with overall 5-year survival of 82%.

There are however, clearly differences in the long-term patient and graft survival for different indications. For instance, patients transplanted due to PSC and metabolic disease have 5-year survival rate of 89%, while patients with HCC and cirrhosis have a corresponding rate of 72%[119].

In general, the most critical period for the outcome after LT is the first year; according to the ELTR-report, 46% of the deaths and 67% of the retransplantations (Re-tx) occur during this period of time. In 44% of patients requiring re-tx, it is performed in the first postoperative month after the primary LT. More than half of the patients who die, do so within 6 months after LT[159]. Typically, graft dysfunction and technical complications dominate during the first postoperative months, infections in the next intermediate period (6-12 months) while malignancies represent the main cause of death in the later postoperative course. Re-tx is necessary in approximately 5-10% of the patients and is associated with significantly lower survival rates compared to primary LT[108,114,159].

## 3.6 Donor considerations

The key to successful liver transplantation starts with selection and procurement of an acceptable liver graft. The source of the graft can be both living and deceased donors. In this thesis we will focus mainly on the latter as the vast majority of LT-procedures in the Western world is performed utilizing grafts from deceased donors. Herein, most of the liver grafts are procured from brain-dead donors (DBD), although the use of nonheart-beating donors (donation referred to as donation after cardiac arrest or DCD) clearly is increasing in both Europe and in the United States[160].

As mortality on the waiting lists are increasing[161] and the number of patients on the waiting lists clearly outnumbers the number of livers available for LT, the use of donors that in the past were

not considered suitable has been increasing. In the past, an ideal liver donor was defined according the following criteria: age < 40 year, trauma as the cause of death, DBD-donor, hemodynamic stability at time of procurement and no steatosis or underlying chronic liver diease[66]. A clear and uniform definition of the term Extended Criteria Donors in liver transplantation has not yet been defined in the transplant community. Broadly speaking, ECD-grafts are thought to be of lower than average quality and associated with an increase in risk for disease transmission and/or linked to poor transplant outcome[10]. Table 3 shows often cited characteristics of an ECD-donor.

**Box 1**
**Definition of extended criteria donors**

Advanced age

Macrovesicular steatosis

DCD

Organ dysfunction at procurement
- ICU stay greater than 7 days
- Hypernatremia greater than 165
- Bilirubin greater than 3
- Elevated aspartate aminotransferase/alanine aminotransferase
- Vasopressor use

Cause of death: anoxia, cerebrovascular accident

Disease transmission
- HBcAb+
- HBsAg+
- Hepatitis C virus
- CDC high-risk donors
- HIV positive
- Extrahepatic malignancy

CIT greater than 12 hours

**Table 3.** Frequently cited characteristics of an ECD donors in LT. Adapted with permission from Vodkin, Extended Criteria Donors in Liver Transplantation, Clin Liver Dis 21 (2017)

### 3.6.1 Donor age

A complete review of the field of ECD-donors in Lt is beyond the scope of this thesis. However, the donor age is perhaps the single most important donor related factor associated with increased risk for inferior post-transplant outcome[162,163], a perceived fact that was further

strengthened by the seminal report by Feng et al in 2006 that introduced the Donor Risk Index (DRI)[8]. In this study, which included more than 20 000 LT performed in the US between 1998 and 2002, the authors identified 7 donor characteristics that were associated with graft failure, amongst them donor age as the most important factor with an isolated relative risk of 1.65 for graft failure compared to an ideal donor. In a concurrent study from the ELTR, it was shown that donor age over 60 years significantly increased 3-month patient mortality[164].

#### 3.6.1.1 Structural and functional changes in the aging liver

From the era before the new HCV-antiviral drugs became available for clinical use, it was known that the risk for HCV-relapse was markedly increased in older livers[165-167]. The reason for this is not clear, but it has been suggested that the speedier progression of fibrosis and development of cirrhosis could be related to hepatocyte telomere shortening correlated to old age[168,169]. However, after the introduction of the highly effective and well-tolerated DAAs to eradicate HCV either before or after transplantation, the established opinion regarding the use of older livers to this patient group is likely to be changed.

The liver seems to be aging slowly, and age-related changes are not reflected in standard laboratory tests[10]. There is, however, a decline in hepatobiliary function, with reduction in bile flow and lower content of cytochrome 450[170]. The rate of DNA repair is decreased together with a reduction in the ability to respond to oxidative stress[171]. With older age, the hepatocyte volume decreases with fewer and larger hepatocytes, which means that the mass of functional hepatocytes can be reduced even though the total organ mass is unchanged[172]. The hepatic blood flow is also reduced[173]. Changes in the aging liver may lower the regenerative abilities of the transplanted liver and make it more vulnerable to ischemia- and reperfusion-injury, especially in the setting of increasing cold ischemia time (CIT)[174]. Some reports have demonstrated a

higher rate of HAT and biliary complications[175-179]. Furthermore, the increased prevalence of steatosis in older livers may result in delayed graft function (DGF) [180,181]. On the histological level there is pseudo-capillarization in the endothelium of the sinusoids with thickening and defenestration, resulting in restricted availability of oxygen and other substances to the liver parenchyma[180]. Atherosclerotic age-related changes are predominantly observed in aorta and in the proximal portions of the its branches, but can in some cases also impact more distal sites and cause occlusive pathology involving the hepatic artery, which theoretically can predispose to vascular complications[182,183]. Summarized, aged livers likely adapt less well to stress compared to livers from younger donors, but the overall hepatic function is well preserved in healthy old livers. Figure 7 illustrates different age-related alterations in old livers.

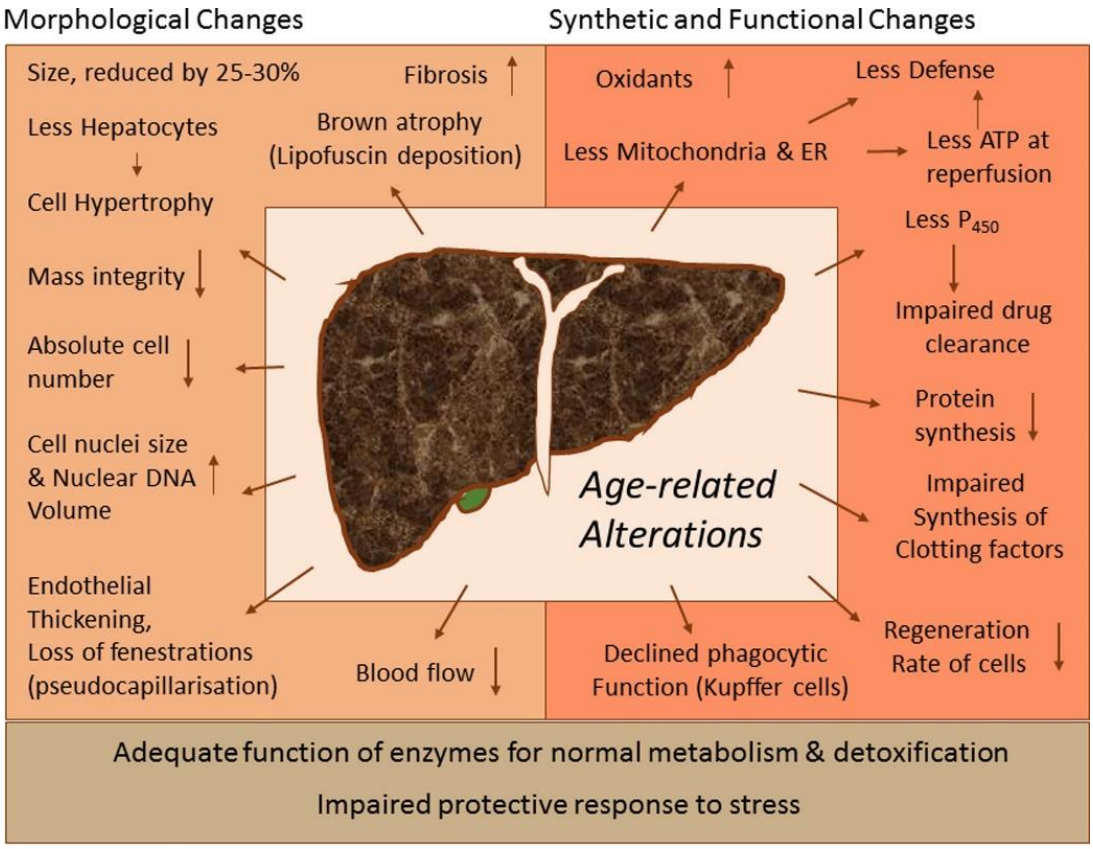



**Figure 7.** Age-related alterations in old livers. Adapted with permission from Dasari et al, The use of old donors in liver transplantation, Best Pract Res Clin Gastroenterol. 2017 Apr;31(2):211-217

#### 3.6.1.2 Outcome in LT using old livers

Even though the age-related changes in older livers mentioned above theoretically could have impact on outcome after LT, there is mounting evidence that using senescent donors, even beyond 80 years of age, can have very good outcomes almost comparable or equivalent to younger donors. Early studies on the use of geriatric liver donors showed divergent results regarding GS, and most of these studies were confined to relatively short follow-up periods with 1- and 3-year survival-figures[176,181,184-194]. In a study from 2002, Cuende et al published 5-year GS as low as 51%[195]. Similar 5-year results with a GS of 58% were documented in a paper by Jimenez-Romero et al from 2013[196], while Lai et al found a 5-year GS as low as 41% in a study from 2011[197]. On the contrary, Borchert et al achieved a corresponding GS of 75% in their study from 2005[198], similar to what Darius et al published in 2012 with a 78% 5-year GS[199]. In a study from 2008, Cescon et al even observed an 81% GS-rate using highly selected donors above 80 years when these livers were transplanted into recipients in good clinical preoperative condition[200].

Over the last few years there have been numerous publications documenting satisfactory, and sometimes excellent, results utilizing old liver donors. In 2014 Ghinolfi et al published a series of 85 LT using donors above 80 years achieving 77% GS at 5 years in the setting of proper donor-selection and donor-recipient matching[201]. The same group reported favorable results when using donors above 90 years in a small series from 2016[202], although the follow-up was very short. In a study from the same year, Barbier et al documented similar 5-year GS for donors above 75 years compared to donors below 60 years of age (65% vs 64% respectively)[203], while Paterno et al found slightly worse long term GS for donors above 70 years compared to donors below 60 in an analysis of the US Scientific Registry of Transplant Recipients (SRTR)[204]. However, when excluding HCV-positive recipients, GS was equal between the two groups.

Bertuzzo et al found no difference in 5-year GS between recipients of donor-livers above and below 70 years (67% vs 71% respectively), even without specific matching criteria in use[205]. Similar results were published by Jimenez-Romero et al in 2017, with 64% 5-year GS for recipients from donors above 80 years. When HCV-positive recipients were excluded, 5-year GS rose to 77%[206]. Dasari et al published a meta-analysis the same year comparing LT using grafts from donors >70 years vs donors <70 years including 7 different studies. Surprisingly, they found better 5-year GS in patients receiving the older livers, probably reflecting different patient-selection between the groups.

In a recent paper by Halazun et al from 2018 based on the United Network for Organ Sharing (UNOS) database investigating more than 3000 patients receiving livers from donors above 70 years, the unadjusted PS was significantly worse in recipients of the older grafts. Significant factors for poor outcome were recipient over 60 years, HCV+ status, ICU-stay, pre-transplant hospitalization and previous surgery. The only donor risk factor in the group receiving older grafts was CIT exceeding 8 hours. Interestingly, when adjusting for these risk factors and then comparing recipients of donor livers above 70 years with no risk factors versus all recipients of younger aged donors, the found no difference regarding neither PS nor GS[207].

In a different study analyzing data from the same UNOS-database, Haugen et al compared recipients of old livers (> 70 years, n=1861) to recipients of younger grafts (18-69, n=37030) and found no difference in 5-year-all-cause graft loss when older livers were given to so-called preferred recipients, defined as first-time, non-urgent patients older than 45 years with BMI below 35 and indication for LT other than HVC and CIT under 8 hours[208].

Outcomes in LT are measured by more parameters than GS alone. There has been concerns that high donor age is a risk factor for increased risk of biliary complications[177], HAT[175] and delayed/poor graft function[209]. Other studies have concluded with no correlation between high donor age and these complications[174,182,210,211]. However, many of these complications will be

possible to solve either by endoscopic treatment or re-operation and will therefore not necessarily affect long term results. Thus, GS stands out as the single most important parameter for evaluating the true impact of donor age on the outcome of LT and has consequently been given main attention on this chapter.

### 3.6.2 ABO-incompatible LT

#### 3.6.2.1 A brief overview of the ABO system

The ABO blood types were discovered by Karl Landstein in 1901, for which he won the Nobel Prize in Physiology and Medicine in 1930. Landsteiner called the antigens A and B. Depending of which antigen the red blood cells (RBC) expressed, it belonged to either blood group A or B. A third group of RBCs did not have the properties of either A or B and was therefore named "O" after the German word "ohne". Later he discovered a new group of blood cells that expressed both A and B antigens, which was named group AB[60]. The ABO blood group antigens consist of oligosaccharides and are mainly expressed on the surface of RBCs (Figure 8). However, ABO antigens are also found on a wide variety of human tissues and are present on most epithelial and endothelial cells[61].

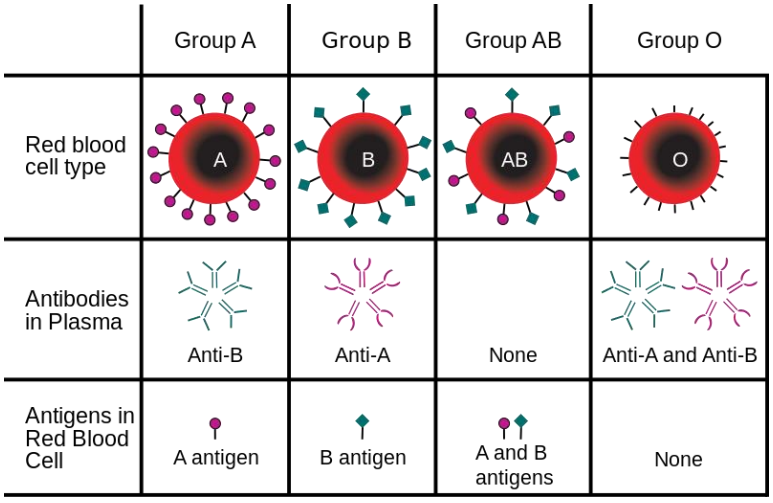



**Figure 8.** Illustration of the ABO blood group with antigens present on red blood cells and antibodies present in the serum

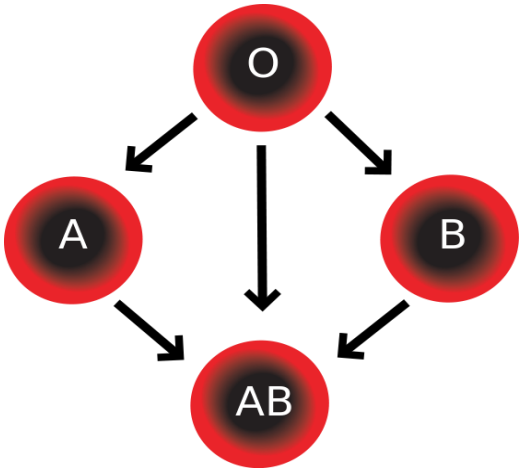



**Figure 9.** Individuals of blood type O are only compatible with donors of the identical blood type, type A and B individuals are compatible with itself and type O, while individuals with type AB are compatible with all blood types. Hence, type O persons are universal donors, while type AB are universal recipients.

The distribution of the blood groups A, B, AB and O varies around the world according to the population. Blood group O is the most common type in most populations, occurring in 44% of Caucasians, 49% of Blacks and 43% of Asians. In Europe, there is a higher frequency of type A, reflected in the following distribution of blood types in Norway; A: 48%, O: 40%, B: 8% and AB: 4%. (www.giblod.no). Group A is further divided into two main phenotypes, called A1 and A2. RBCs of subtype A2 have a low expression of A antigens on the surface, which is believed to be responsible for the lower immunogenicity of organs of type A2 donors with its clinical implication in the setting of organ transplantation[212,213]. A2 is in fact divided into many different subtypes and is often referred to as blood group A1-minus. Among Caucasians, type A1 is found in approximately 80% of persons with blood group A, while A2 (or A1-minus) comprises around 20%. In Asians however, group A2 is very rare with a frequency of less than 1-2%[214,215].

The immune system produces highly reactive antibodies towards the ABO blood group antigens which are *not* found on the individual's red blood cells. Hence, a person with group A will have anti-B antibodies, and a group B individual will have anti-A antibodies. Individuals with group O type will have both anti-A and anti-B in their serum, while group AB individuals will have

neither anti-A nor anti-B. These antibodies are mostly of the IgM class, but can also consist of IgG and IgA[61]. Interestingly, a person's ABO type can be temporary altered by several illnesses. For instance, an A-individual with an infection can acquire the B-antigen because the infecting bacteria release an enzyme that converts the A antigen into a B-like antigen, thus stimulating production of anti-B into the serum. After treatment of the infection, the individual's blood group returns to normal[216,217]. Furthermore, it is known that an individual's level of antibodies will fluctuate with time and that infection with certain bacteria can stimulate their production[60]. In addition, neo-expression or aberrant expression of A or B substances in malignant cells can also boost the production of antibodies, implying that patients undergoing ABO incompatible LT due to malignancy might be more exposed to antibody-mediated insults[218].

#### 3.6.2.2 Clinical experience with ABO-incompatible LT

##### 3.6.2.2.1 History of ABOi LT

The current experiences utilizing ABO-incompatible liver donors for transplantation can broadly be divided into two main categories. ABOi-LT with deceased donors, first introduced in the early 1970ies by Thomas Starzl and coworkers, have mainly been performed in urgent cases in Western countries. There are relatively few published reports, and the results have varied from almost equally good as ABO compatible transplantations to poor outcomes with significantly inferior patient and graft survival. On the contrary, multiple large-scale reports from experiences in Asia, mainly from Japanese and Korean transplant centers, currently show results from ABOi-LT that are in line with what is achieved with ABO compatible transplantation. However, the majority of the transplantations performed in Asia have been performed during the last 15 years using living donors and, in an elective, thoroughly prepared setting with few urgent cases.

Results from early animal studies performed by Starz et al suggested that the liver is a "immunologic privileged organ" less susceptible to rejection than kidney and heart[86]. In 1979 the same group reported a series of 11 patients without any evidence of developing acute rejections after ABO incompatible LT. Although graft survival rates were worse than in the group with ABO compatible transplantations, six of the 11 patients survived more than 6 months and the outcome were considered acceptable for use in adults in emergency situations[219]. Later Starzl and coworkers published their experience with 31 adult ABOi LT using a more developed immunosuppressive regimen with cyclosporine and prednisolone and found that results were "surprisingly successful", albeit clearly inferior to the ABO compatible comparison group[220]. As the worldwide experience with liver transplantation grew, more groups found that crossing the ABO barrier was associated with high risk for hyperacute and antibody-mediated rejection resulting in cholangitis, arterial thrombosis, necrosis and often eventually graft loss with 2-year graft survival as low as 30%[18,221-227]. Patient survival were in several studies in line with results from ABO compatible transplantations, however at the price of a high rates of retransplantations. Because of these inferior results with often severe complications ABOi LT using deceased donors became unfavorable and was reserved for highly urgent cases only.

#### 3.6.2.2.2 Living donor ABOi LT

Inspired by new advances in immunosuppressive regimens and successful experiences from ABO-incompatible living donor renal transplantations[61], ABO incompatible living donor liver transplantation (ABOi LDLT) was initiated in Japan at the beginning of the 1990ies. However, the outcomes of early ABOi LDLT were also poor, often resulting in acute rejections and graft loss[228]. Two typical reasons for graft failure were described: Firstly, liver necrosis occurring acutely within 1-2 weeks, leading to graft loss within a month. Secondly, a type of intrahepatic

bile-duct injury typically manifested more slowly 2-3 months after transplantation, often resulting in need for retransplantatition after some time[229]. Although use of pre- and postoperative plasma exchange, splenectomy and high doses of immunosuppressive drugs seemed to improve the results, the major breakthrough in ABOi LDLT came after the introduction of the drug Rituximab (RIT) for clinical use in liver transplantation in 2003/2004. Rituximab is a monoclonal antibody against the CD20 receptor on the surface of B-cells, inducing cell death upon binding to the receptor through two main mechanisms (Figure 10). Although RIT does not affect B-stem cells or already produced antibody secreting plasma cells, the drug prevents clonal expansion of activated B cells and thereby effectively attenuates antibody production.

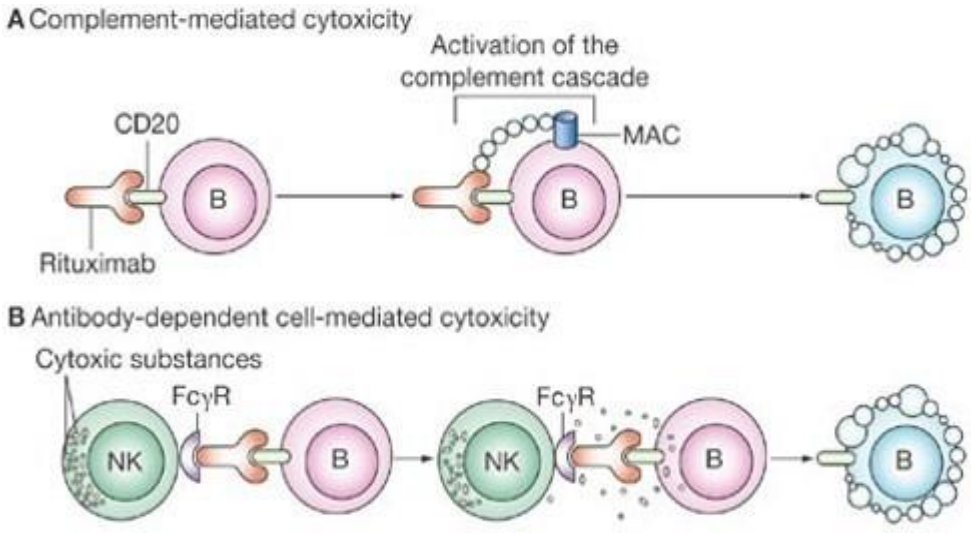



**Figure 10.** Illustration of the two main pathways for rituximab to induce B-cell death. Adapted with permission from Bayry J *et al.* (2007) Monoclonal antibody and intravenous immunoglobulin therapy for rheumatic diseases: rationale and mechanisms of action *Nat Clin Pract Rheumatol* **3:** 262–272

In a study from 2008, Egawa et al documented that 3-year patient survival had increased from 30% to 80% after systematic use of RIT in ABOi LDLT in Japan[215]. Since then, further refinements in desensitization-protocols have further improved the results, and numerous

reports from especially Korean and Japanese transplant centers shows that current outcomes from ABOi LDLT are almost equal to what is achieved in the ABO compatible setting[59,62,230-234]. A recent report from Lee et al indicates that RIT alone together with immunoglobulins given in the postoperative course is sufficient as desensitization (DZT), suggesting that plasmapheresis can be omitted in ABOi LT[233]. Plasmapheresis is hampered with several potential adverse effects like cardiovascular complications, water-electrolyte disturbances and risk for infections[235]. The correct timing Rituximab-administration is debated, most groups have given the recipients a single dose in the period from 2-4 weeks before transplantation. Several studies have revealed that the effect of RIT on B cells occur rapidly, eliminating cells within 48-72 hours and persisting for several months after administration[59,236]. Interestingly, some reports have shown equally good results in patients receiving RIT within the last 7 days before transplantation[231,232,237], which indicates that RIT can have impact also in the acute setting with emergent transplantation. The concept of RIT in acute cases was investigated in a single center study by Shen et al from 2014, utilizing deceased donors. 35 patients were given RIT on the day of transplant, and their 3-year graft survival of 80% did not differ from the ABO compatible group[238]. Multiple doses of RIT do not produce better results. On the contrary, it seems to be associated with significantly increased risk for fungal and CMV infections post tx[231]. The existing literature is inconclusive regarding the association between levels of pre- and postoperative anti A/B titers, the cut-off values and the effects on outcome. It is a general belief that high titer-levels in the postoperative course is related to increased risk for AMR and graft failure. However, some reports show a correlation[231,238], other conclude with no association[59,62,230] while many more are inconclusive to whether titer-levels are of importance or not.

#### 3.6.2.2.3 Deceased donor ABOi LT

During the same time period as ABOi LDLT has flourished in Asia, ABOi LT with deceased donors (ABOi DDLT) have continued to be occasionally performed at centers in the Western World. The same principles for immunosuppression and DZT as used in Asia have to different degrees been adopted, but not in the same methodological and systematic manner like in the setting of ABOi LDLT. Most of these transplantations have been undertaken in urgent situations where no ABO compatible graft has been available, and time for preoperative desensitization have been non-existent or at least very limited. Most of the relatively few published studies are biased by very heterogeneous patient populations involved in combination with often inconsistent protocols for immunosuppression in use, making it rather difficult to draw clear conclusions. In a small sample study (n=14) by Toso et al from 2007 they found a 5-year GS of 56% among ABOi LT patients, not different from the ABO compatible group[16]. Plasmapheresis was used as adjuvant treatment for rejections only in the first 12 patient and as prophylaxis for high or rising isoagglutinin titers in the subsequent two. Stewart et al published results from a large UNOS-based study in 2009 including 667 adult ABOi LT patients finding that although graft survival had improved in the recent era, outcome was still clearly inferior compared to ABO compatible LT[15]. In a meta-analysis by Wu et al from 2011 including a total of 811 ABOi LT-patients, the main conclusion was that graft survival was significantly inferior compared to ABO compatible control groups[239]. However, details regarding immunosuppression were not provided and most included studies did only have short term follow-up. Zou et al reported clearly inferior results in 22 ABO incompatible LT performed on patients suffering from HBV-related acute liver failure accomplishing a 5-year GS of only 21%[240]. None of the patients received RIT, and only two of 22 were treated with plasmapheresis. These disappointed results are in contrast to what was achieved in the above-mentioned study by Shen et al, where patients in similar settings as those Zhou-study achieved

excellent outcome when given RIT at day of transplant[238]. Results from several small studies have indicated that adjuvant treatment with RIT, plasmapheresis and immunoglobulins, or different combinations of these, can produce acceptable results in deceased-donor ABOi LT[13,14,17,241]. A recent meta-analysis from 2017 by Lee et al including 21 studies and more than 8200 patients from both deceased donor and living donor transplants confirmed earlier findings in large-scale studies, concluding that ABO incompatible liver transplantations in general are clearly associated with worse graft survival and higher incidence of complications[242]. However, and in line with multiple other studies, patient survival seems not to be inferior and in those patients that received rituximab graft survival was comparable to that of ABO compatible LT.

A major limitation affecting most of the published studies is the lack of discrimination between A1 and A2 donors, which undoubtedly will bias the results since it's a well-established fact that A2 livers given to blood type O recipients produce very good results[243-246].

## 3.7 Allocation of organs

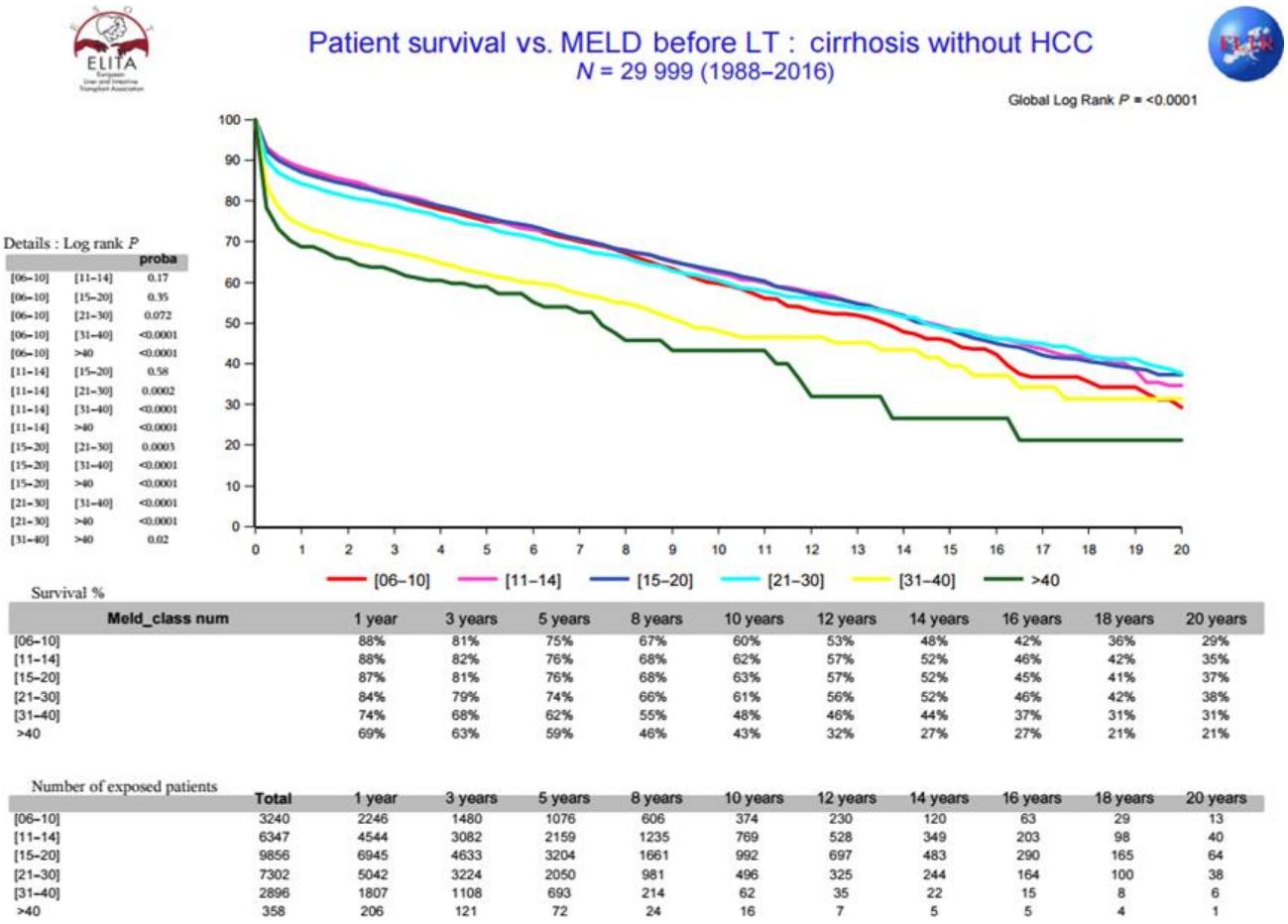
### 3.7.2 Organ allocation and the MELD-score

Worldwide, the fundamental challenge in organ transplantation is the overwhelming gap between the supply of donor organs and the number of patients with end-stage organ failure expected to benefit from being transplanted. When designing a fair as possible allocation system, three main ethical principles should be utilized. *Justice* means that the allocation system should be able to distribute organs according to objective and uniformly applied rules. *Equity* refers to the principle that allocation should be based on the severity of disease rather than the type of disease and the patient's waiting time, meaning that individuals with similar risk of death should have similar access to a transplant. *Utility* means that the allocation system should prioritize patients that are most likely to benefit the most from being transplanted defined by

incremental life-years gained compared to waiting list survival[66,247]. Taken together, this means that the goal should be to rank patients on the waiting list according to medical need, with priority to patients that are believed to benefit the most from the transplant.

Obviously, fair distribution of organs is a much greater challenge in areas with long waiting lists compared to the fortunate situation in Norway, where the current waiting-list typically is comprised of only around 30 patients. Until 2002 the potential candidates for LT in the US were prioritized according the patient's United Network of Organ Sharing (UNOS)-status in combination with time on the waiting list, which basically gave big benefits to patients that were listed for transplant at an early stage of their disease[248]. After 2002, the Model for End-stage Liver Disease (MELD[249]) have been used for ranking patients on the waiting lists in the US, adopting the urgency principle for allocation of livers. This is a mathematical model that defines the risk of death using three laboratory values: international normalized ratio (INR), total bilirubin and creatinine. It also takes into account whether the patient has been on renal replacement therapy during the last week or not. The MELD-score will range between 6 and 40, where the maximum score of 40 is associated with 3-months mortality as high as 70 %. Interestingly, studies have shown that patients with MELD scores below 15 incur more risk by the LT-procedure itself then the possible benefit by being transplanted, and even more so if they are given organs of suboptimal quality[250,251]. One year after the introduction of this system in the US, the on-list mortality was reduced with 3,5% and the median waiting time was decreased by over 200 days. At the same time, the number of patients on the waiting list was reduced by 12%, mainly due to fewer patients with low MELD-score being listed[252,253]. The MELD-score has been further refined by incorporating sodium in the equation and by granting extra MELD-points for certain diseases, like for instance patients with HCC with a well-functioning liver and hence low MELD score, but with a with high risk of death due to cancer due to the malignancy. The limitation and main disadvantage to the MELD-model is that although it has

demonstrated high accuracy in predicting short-term mortality for patients on the wait-list, is has not proven accurate in predicting long-term outcome and the survival benefit after transplantation[253]. In addition, the MELD-system does not take special consideration in patients who in spite of low MELD-scores still need a liver transplant due to portal hypertension or other complications to liver cirrhosis[254,255]. However, despite its limitations and in lack of better and more robust alternatives, MELD-based allocation systems have gained considerable acceptance also outside the US and is today used in most parts of Europe and South-America. Since most European countries started to use MELD-score for allocation in 2006-2007 the proportion of patients with MELD score more than at the time of LT has almost doubled. However, the survival of this group is inferior, especially for those with MELD more than 40[159] (Figure 11).




Survival %

| Meld_class num | 1 year | 3 years | 5 years | 8 years | 10 years | 12 years | 14 years | 16 years | 18 years | 20 years |
|---|---|---|---|---|---|---|---|---|---|---|
| [06–10] | 88% | 81% | 75% | 67% | 60% | 53% | 48% | 42% | 36% | 29% |
| [11–14] | 88% | 82% | 76% | 68% | 62% | 57% | 52% | 46% | 42% | 35% |
| [15–20] | 87% | 81% | 76% | 68% | 63% | 57% | 52% | 45% | 41% | 37% |
| [21–30] | 84% | 79% | 74% | 66% | 61% | 56% | 52% | 46% | 42% | 38% |
| [31–40] | 74% | 68% | 62% | 55% | 48% | 46% | 44% | 37% | 31% | 31% |
| >40 | 69% | 63% | 59% | 46% | 43% | 32% | 27% | 27% | 21% | 21% |

Number of exposed patients

| | Total | 1 year | 3 years | 5 years | 8 years | 10 years | 12 years | 14 years | 16 years | 18 years | 20 years |
|---|---|---|---|---|---|---|---|---|---|---|---|
| [06–10] | 3240 | 2246 | 1480 | 1076 | 606 | 374 | 230 | 120 | 63 | 29 | 13 |
| [11–14] | 6347 | 4544 | 3082 | 2159 | 1235 | 769 | 528 | 349 | 203 | 98 | 40 |
| [15–20] | 9856 | 6945 | 4633 | 3204 | 1661 | 992 | 697 | 483 | 290 | 165 | 64 |
| [21–30] | 7302 | 5042 | 3224 | 2050 | 981 | 496 | 325 | 244 | 164 | 100 | 38 |
| [31–40] | 2896 | 1807 | 1108 | 693 | 214 | 62 | 35 | 22 | 15 | 8 | 6 |
| >40 | 358 | 206 | 121 | 72 | 24 | 16 | 7 | 5 | 5 | 4 | 1 |

**Figure 11.** Patient survival versus MELD-score at time of LT for patients with cirrhosis without HCC. Adapted with permission from Adam, 2018 Annual Report of the European Liver Transplant Registry (ELTR) - 50-year evolution of liver transplantation, Transpl Int. 2018 Dec;31(12):1293-1317

In the Scandinavian countries the waiting lists are relatively short, and allocation of liver grafts are performed based on center-wise allocation, meaning that each center has its own waiting-list and the right to determine the prioritization among patients and use of donor livers within a defined geographical area. Instead of allocating liver grafts based on a mathematical formula like the MELD-system, prioritization can be based on clinical assessment, considering both donor and recipient risk factors. However, for very sick patients in urgent need for a liver graft, the Scandiatransplant organization has developed a system that requires the first available liver graft in the entire Scandinavia to be given to this patient. This highly-urgent status does also include patients in need for a re-transplantation within 14 days of the primary LT due to a non-

functioning first liver graft or severe complications resulting in graft failure. Donor livers that are sent out due to the urgent-call system are required to be paid back later to the procurement-center. There is also a system for sharing organs between the Scandinavian transplant centers through a "rotation-list". If the procuring center does not have a suitable recipient for a liver graft, this liver will be offered to the other centers in Scandinavia according to a rotation system, securing that every procured donor liver can be utilized. In addition, Scandiatransplant has since 2015 established a common waiting list for children, meaning that each center is obligated to offer liver grafts fulfilling certain criteria to centers having children on the joint waiting list. After the introduction of this shared waiting list the waiting time for children in need of a liver graft has been significantly reduced in all the Scandinavian countries.

### 3.7.3 Donor and recipient matching

Ideally, every liver transplant patient should receive an optimal graft. However, due to the disparity between the demand and the number of liver grafts available, this is not possible to achieve. In the setting of limited organ availability and increasing use of extended criteria donors it is of importance to match the right organ to the right patient. Every patient will naturally be best off with an ideal organ, so for practical purposes the concept of organ-matching in LT comes down to allocating livers from older donors and other types of extended criteria donors to recipients while still getting acceptable results.

Satisfactory results have definitively been achieved when allocating old donor grafts to suitable recipients as described in section **3.6.1.2** (Outcome in LT using old livers). However, multiple studies conclude that one should refrain from allocating these grafts to candidates with high MELD-scores and prolonged ICU-stay, avoid patients with untreated HCV-infection and stress the importance of keeping cold ischemia time as short as possible[10,200,256,257].

Several research groups have tried to develop models for aiding decision-making when it comes to identifying favorable donor-recipient pairs. In 2006 Feng et al introduced the concept of donor risk index[8], identifying seven risk factors associated with an increased risk for graft failure: donor age above 60 years, DCD-donor, split grafts, African-American race, low height and cerebrovascular incident as reason for death. The model also included cold ischemia time and was based on calculations from more than 20 000 LT performed in the US between 1998 to 2002, before the introduction of MELD score. DRI provides useful data on certain donor-parameters and has served as a foundation for several later tools for organ risk assessment. However, it does not consider recipient characteristics and has not come into widespread use in the transplant community[11]. Several studies have shown that livers from donors with high DRI-score (meaning a high-risk donor) most likely have been given to patients with low disease severity (MELD score 10-14) and least likely to very sick patients with high MELD-score, resulting in inferior and sometimes detrimental outcome for low-MELD patients receiving suboptimal grafts[251,258]. Schaubel et al and Rauchfuss et al concluded that livers from high-DRI donors are effective for high but not for low-MELD candidates, suggesting that earlier transplantation for very sick patients is more important than waiting for a more optimal organ[251,259]. Pairing of high-DRI livers with lower-MELD patients fails to maximize survival benefit and may deny life-saving organs to high-MELD candidates who are at high risk of dying without rapid transplantation[251].

After the introduction of DRI several other models have been proposed for more accurate predication of post-transplant outcomes, taking into consideration both donor factors, recipient factors and to some extent operative factors. The Donor Age and Recipient Model for End-Stage Liver Disease (D-MELD) is the mathematical product of donor age and preoperative MELD-score. Utilizing this model, avoiding a D-MELD score above 1600 improved patient- and graft survival in a study by Halldorson et al[260]. This model has been criticized for lacking

important donor and recipient factors and further risk assessments for older donor livers[261]. The Survival Outcomes Following Liver Transplantation (SOFT) score uses 18 different risk factors to predict 3-month survival after LT[262]. However, it includes complex modelling, has focus on short-term survival and often all donor factors needed for computation are not available at time of the donor offer, hence its use is of limited value[11]. The Balance of Risk (BAR) score includes 6 predicators of post-transplant survival: recipient age, MELD, re-transplant, dependence on life support prior to LT, donor age and CIT [263]. A BAR-score was calculated between 0 and 28, where a score above 18 was associated with inferior outcome. However, the authors did not have focus on long-term outcome, and only 3% of the patients in this study had a score of 18 or more, resulting in limited applicability of this model. The Eurotransplant Donor Risk Index (ET-DRI) includes 5 donor factors (age, cause of death, gamma-glutamyl transferase, DCD-donor, split-liver) and 3 transplant factors (CIT, regional/national share, rescue-allocation)[264]. However, the accuracy in predicting outcome after LT has been shown to be relatively low, with C-statistics around 0,5 when validated externally[265]. It has later been refined and combined with a simplified recipient risk index (sRII) achieving a somewhat better c-statistic of 0.62[266].

There are basically two main problems affecting all the above-mentioned models and preventing their wide-spread clinical use. Firstly, none of them have proven to be able to predict the outcome after LT with sufficient degree of accuracy. Secondly, with all these models it is necessary to calculate a score for each potential donor-recipient pair, making them time-consuming in situations with many possible candidates on the waiting-list. Although these scoring-systems may be helpful in guiding clinicians choosing the right patient for the right liver graft, no single statistical model can accurately capture, characterize or predict the clinical donor-recipient matching that is undertaken at the time of organ acceptance[267]. Hence, the ideal scoring system for identifying the best donor-recipient match does not yet exist, as the systems currently available are not statistically robust enough. Scoring-systems for the future should

probably be based on not just a list of certain donor and recipient variables, but also take into consideration probability of death on the waiting list, post-TX survival, global survival benefit and even cost-effectiveness[261].

Table 4 summarizes selected models for predicting outcome after LT.

**TABLE 2. Summary of Selected Models to Predict Posttransplant Outcomes in the MELD Era**

| | DRI | ET-DRI | SOFT | BAR | D-MELD | LDLT DRI |
|---|---|---|---|---|---|---|
| Variables included | Donor factors: age, COD, DCD, partial/split, race, height, regional/national share<br>Operative factor: CIT | Donor factors: LDRI (age, COD, DCD, partial/split, regional/national share, GGT, rescue allocation<br>Operative factor: CIT | Donor factors: age, COD, creatinine, national share<br>Recipient factors: age, BMI, previous transplants, abdominal surgery, albumin, dialysis, ICU hospital admission, MELD > 30, life support, encephalopathy, portal vein thrombosis, portal bleed, ascites<br>Operative factor: CIT | Donor factor: age<br>Recipient factors: age, MELD, retransplantation, life support<br>Operative factor: CIT | Donor factor: age<br>Recipient factor: MELD | Donor factors: age, weight, graft type<br>Recipient factors: age, weight, albumin, diagnosis |
| Available online calculators | https://gastro.cchmc.org/calculators/donor-risk-index/ | | | http://www.assessurgery.com/bar-score/bar-score-calculator/ | http://www.d-meld.com/calc.php | |
| Advantages | Readily available variables at the time of transplant, most widely recognized and studied model, well-validated in selected recipients | Readily available variables at the time of transplant | Multiple levels of risk, identified futility | Readily available variables at the time of transplant, included relevant recipient factors | Simple to use | Parsimonious, all factors available at time of transplant |
| Limitations | Included factors that lacked biological plausibility, low C-statistic, exclusion of salient donor factors, needs validation in current donor/recipient pools, center impact unaccounted, donor factors not all available at time of offer | Lower C-statistic in validation studies, donor factors not all available at time of offer | Multiple factors, complex statistical modeling, emphasis on short-term survival, donor factors not all available at time of offer | Exclusion of salient donor factors, lack of longterm data, majority of liver transplants would fall in favorable category (<18), donor factors not all available at time of offer | Many significant factors excluded, center and geography unaccounted, lacks further risk assessment for older donor livers | C-statistic is only comparable to models with deceased donor, which is rather low |

**Table 4.** Summary of selected models for predicting outcomes in LT. Adapted with permission from Flores, The Donor Risk Index: A Decade of Experience, Liver Transplantation, Vol 23, Issue 9, Sept 2017

# 4 AIMS OF THE PROJECT

The aims of this thesis were related to three different aspects of liver transplantation:

## 4.1 Paper 1

The aim of this study was to investigate whether the outcome of the use of liver donors above the age of 75 years in the Scandinavian countries during the period 2001 to 2011 gave acceptable results compared to a matched group of recipients receiving liver grafts from donors aged 20-49 years.

## 4.2 Paper 2

In this study, we analyzed the outcome of all ABO incompatible liver transplantations performed in Gothenburg and Oslo between 1996 and 2011. The aim was to compare these results to the patient- and graft survival of all other LT performed at the two centers during the same period in order to assess whether the ABO-I transplantations gave acceptable results. In addition, we also investigated whether the rate of complications including cellular and antibody-mediated rejections rate in the ABO-I group was higher than in the ABO compatible group.

## 4.3 Paper 3

In this study, we analyzed our institutional experience with rescue liver transplantations in patients diagnosed with acute liver failure due to either iatrogenic injuries to the portal vein or

hepatic artery or following liver resection in the period 2003 to 2016. The aim was to elucidate whether it is feasible and justifiable to offer these patients LT compared to patients with standard indications.

# 5 SUMMARY OF THE RESULTS

## 5.1 Paper 1

During the period from 2001 to 2011 a total of 54 patients in Scandinavia received a liver graft from donors above 75 years of age (D75 group). The control group consisted of 54 matched patients that were transplanted with livers from donors aged 20-49 years in the same period (D20-49 group). Median donor age was 77 years (range 75-86) in the D75 group and 41 years in the 20-49 group. We did not find any significant differences in either patient- or graft survival between the two groups at 1, 3- and 5-year follow-up after LT. The 1-, 3- and 5-year patient/graft survival were 87/87%, 81/81% and 71/67% in the D75 group, versus 88/87%, 75/73% and 75/73% in the D20-49 group. When we analyzed only patients who were transplanted due to non-malignant diseases (n=28 in both groups), we also found no significant difference in patient- or graft survival. Further, there we no difference in vascular complications rates between the groups, and we found comparable rates of rejections in both cohorts. Two patients in the D75 group and one patient in the D20-49 group needed a re-transplant. Importantly, there were no incidents of primary non-function in any of the groups. However, we did find a significantly higher incidence of biliary complications (30% vs 13%, p=0,03) among patients receiving livers from donors above 75 years, indicating that older livers are more vulnerable to the donation process and the transplant procedure itself.

## 5.2 Paper 2

From 1996 to 2011 a total of 61 ABO incompatible LT were performed in Oslo and Gothenburg. Most of the patients were transplanted du to urgent indications (n=33) or hepatic malignancies (n=13). The transplantations were performed over a relative long time-span and at two different centers, hence the immunosuppressive regimens used and the indications for LT were heterogeneous. The 1-, 3-, 5- and 10-year graft survival for the whole ABOi-group were 71/57/55/51% respectively. This was significantly worse than the corresponding GS of all other LT-recipients (control group) during the same time period at 87/79/73/60%, respectively. However, recipients of blood type A2-grafts did not have inferior GS compared to the control group. The patient survival for the ABOi-group was not significantly worse than for the control group and this could be attributed to the high rate of re-transplantation (23%). There were high rates of vascular (34.5%) and biliary (35%) complications in the non-A2 group, while the corresponding rates for the A2-group were 19% and 22%, respectively. Twenty-eight patients (46%) were treated for acute rejection during the first postoperative month, of which 4 patients were diagnosed with AMR. The total rate of AMR was 6.5%, and the overall rejection rate was 37.5% in the A2-group and 55% in the non-A2-group. There seems to be a correlation between the levels of anti A/anti B titers measured after LT and risk for rejection, but our data were too limited to draw any clear conclusions on this issue. We conclude that ABO incompatible LT with non-A2 grafts is associated with inferior graft survival and increased risk of rejection, vascular and biliary complications, and should probably be reserved to very acute situations where no ABO compatible graft is available. ABOi LT with A2 grafts seems to give good long-term graft survival and can safely be used in urgent situations, and probably in elective settings where long waiting times are to be expected.

## 5.3 Paper 3

In the period from 2003 to the end of 2016, a total of thirteen patients at our institution underwent liver transplantation due to either severe iatrogenic injuries of the liver vasculature or due to remnant liver failure after prior resection. Both conditions are associated with high mortality. The fortunate donor situation in Norway combined with a short waiting list has made it possible to perform LT also in patients beyond conventional criteria. Seven of these 13 patients had been operated with radical surgery for cancer before LT, and six patients were diagnosed with non-malignant disease. Three of the patients with malignant disease did not experience disease recurrence. However, four patients had cancer recurrence and died 7, 24, 45 and 78 months after transplantation. Five of six patients with non-malignant disease fully recovered, but one patient died after 9 months due to infectious complications. We conclude that LT for patients suffering liver failure due to portal vein and hepatic artery injury in patients with non-malignant disease seems justified. However, it is doubtful whether patients with proven malignant disease beyond established criteria for LT should be offered liver transplantation in settings where the primary operation has resulted in irreversible liver failure.

# 6 METHODOLOGICAL CONSIDERATIONS

## 6.1 Patient populations and study design

The first study is a retrospective case-control study on the 54 patients in the Scandinavian countries that were transplanted with donors aged 75 years and above (D75 group) between 2001 and 2011. Outcome were compared to a control group of 54 patients that were transplanted with donors aged 20-49 years (D20-49 group) during the same time period. Data on the patients and corresponding donors were extracted both from the medical files at each participating center and from the NLTR-database. Primary end-points of the study were PS and GS after 1, 3 and 5 years follow-up. In addition, we analyzed the most common complications occurring after the LT procedure. For further analysis the GS of the patients in the D75 group with benign diagnosis was compared to the GS of all other LT-patients registered in the NLTR from 2001 to 2011 that underwent LT due to benign diagnosis with donors aged 20-49 years (NLTR group, n=802). The primary end-points were calculated from the day of LT to end of study at February 1st ,2012, or to patient death or graft loss. The Student's t-test and Mann-Whitney U-test were used to compare continues variables between the group, while categorical variables were compared by using the chi-squared test. Survival analysis was performed using GraphPad Prism version 5 (GraphPad Software, Inc, La Jolla, CA) and data from the groups were compared by using the log-rank test. P-values less than 0.05 were considered statistically significant.

The NLRT comprises complete data files of all LTs in Denmark, Sweden, Norway and Finland from 1982. Since January 1st ,1990, all patients that have been listed for LT have been registered prospectively and all data are stored at Scandiatransplant in Aarhus, Denmark. Participation in the registry is mandatory to be put on the waiting lists for a transplant and is based on informed consent by the patients. The data in the NLTR and Scandiatransplant system is managed within

a data agreement between "Nordisk Lever Transplantasjonsregister" (CVR 29763011) and OUH-R (dated February 1st ,2007).

The second study is a retrospective uncontrolled study on 61 patients transplanted with ABOi liver grafts in Gothenburg and Oslo from 1996 to 2011 (ABOi group). However, the primary endpoints of the study, PS and GS, were compared to a control group consisting of all other LT transplants performed at the two centers over the same period of time (NLTR group, n=1372). Data on the ABOi group were extracted from center-specific medical records and survival data on the NLTR group was collected from the NLTR database under the same juristic and technical considerations as mentioned in the previous section handling study 1. Details regarding testing methods for determination of anti A/B titers are described in Paper 2. The main endpoints (PS and GS) were calculated from the day of LT until September 1st ,2012, or to patient death or graft loss. Kaplan Meier survival analysis was performed by using the same software as in study 1 (GraphPad® ver. 5) and survival rates were compared by using the log-rank test. P-values $<0.05$ were considered as statistically significant. When comparing the group of patients that received livers from A2 donors (n=32) and those who were transplanted with non-A2 donors (n=29), the Chi-squared test was used was categorical variables while The Student's t-test and Mann-Whitney U-test were used for continuous variables.

The third study is a retrospective, uncontrolled study analyzing the outcome of 13 patients transplanted at our center due to either to liver failure caused by severe iatrogenic injuries of the liver vasculature or due remnant liver failure after prior resection. Complete medical files for all the included patients were available for data-extraction, while the corresponding donor-data was collected from our local donor-registry. The primary endpoints in the study (PS and GS) were estimated by Kaplan Meier analysis using the same software and methods as described for study 1 and 2. Further, we compared the survival differences between the group

of patients with benign diagnosis (n=6) and for those with malignant conditions (n=7) by same methods.

## 6.2 Ethical considerations

All three studies were approved by the local institutional review board (approval number 2012/4059 for study 1, 2011/582 for study 2 and 2015/1442 for study 3) and all were performed according to the Declaration of Helsinki. All included patients have given written consent to be included in "Norsk levertransplantasjonsregister" and "Nordisk levertransplantasjonsregister" (NLTR). In addition, all patients have given written consent to be included in research on hepato-biliary diseases. Data management and processing were done at secure systems according to institutional data protection clearances. The juristic aspects regarding management and handling of data from the NLTR is described above. With respect to the specific ethical considerations related to the three different studies included, these aspects are addressed in the Discussion section of this thesis.

# 7 DISCUSSION

This thesis is based on three original research articles that all describes liver transplantations performed in the settings were either donor- or recipient criteria are at the limit or even beyond what are regarded as internationally accepted. In paper 1 we describe the common Scandinavian experience with the use of liver donors above 75 years in the period 2001 to 2011 and compared the results with a control group that received so-called ideal liver grafts from donors aged 20-49 year. In paper 2 we have analyzed the results from Gothenburg and Oslo when using ABO-incompatible liver donors between 1996 and 2011 and compared the main outcomes (patient- and graft survival) to all other liver transplantations performed at the two centers during that period. In paper 3 we describe our institutional experience with rescue liver transplantations to patients that have developed acute liver failure after iatrogenic injuries to the liver or after liver resections, a patient group that is rarely described in current medical literature.

## 7.1 Paper 1

### 7.1.1 Patient- and graft survival

In our study the patients that received livers from donors above 75 years did not have inferior PS nor GS compared to those that were transplanted with livers from donors aged 20-49 years. One-, 3- and 5-year PS were 87%, 81% and 71% respectively for the D75 group, compared to 88%, 75% and 75% respectively, for the control group. The corresponding 1-, 3- and 5-year GS were 87%, 81% and 67% for the D75 group, and 87%, 73% and 73% for the control group. Prior to our work only one earlier study had published data on 5-year GS in LT using exclusively donor grafts above 75 years, reporting a 5-y GS of 51% in 19 patients. Despite several earlier studies documenting good results using livers from donors >70 years starting in

in the mid 1990's[181,184,185], later reports showed inferior patient outcomes in the setting of old grafts. Petridis et al documented a 3-y GS as low as 20% in patients with donors aged 80-93 years in a study from 2008[190], while Lai et al reported somewhat better, but still inferior, results with 41% 5-y GS in recipients of donors from 70 to 89 years of age. Several other reports also showed disappointing results in the past with elderly grafts[187,268,269]. In the original DRI-paper by Feng et al from 2006, donor age was reported to be the strongest factor associated with graft failure[8]. However, contrary to this, other groups have achieved very good results when using grafts from donors above 70[188,196,198-200,270,271] and 80[191,200,201,272] years of age. Even livers from donors more than 90 years old seem able to produce acceptable outcome[202] given the right patient selection.

Our findings were in line with those published in a concurrent study on 85 patients by Ghinolfi et al from 2014[201], and results from several other studies published during the last few years supports the notion that old livers can give excellent results in right patients. Ghinolfi et al published another study in 2016 demonstrating a 5-year GS as high as 81% in 88 patients with donors above 80 years. In another study from 2016, Barbier et al published results from 157 LT using donors above 75 years with a 5-year GS of 65% compared to 64% in a control group with donors aged below 60 years[203]. The same year Paterno et al published results from US SRTR/UNOS database on 540 patients receiving livers from donors above 70 years. Patient survival was equally good compared to a group of patients with donors below 60 years, but unadjusted GS was slightly worse. However, when recipient HCV status was not included in the analysis, the difference in GS was not statistically different. Jiménez-Romero et al documented a 5-year GS of 64% in 51 patients transplanted with livers from donors aged above 80 years in a study from 2017, not statistically different from 74% in a control group with donors below 65 years[206]. When excluding the 10 patients with HCV+ status, 5-year GS rose to 77%. In another study from 2017, Bertuzzo et al found a 5-year GS of 68% in 190 patients

receiving grafts from donors above 70 years, not different from a corresponding 71% for those with donors below 70[205]. Also in 2017, Dasari et al published a review and meta-analysis including 8 studies with a total of 879 patients with donors above 70 years[273]. Results were compared to recipients of donor livers less than 70 years. Surprisingly, at 5-year follow-up, both PS (OR 1.52, p=0.001)) and GS (OR 1.46, p= 0.001) were better among the recipients of the older grafts. The authors concluded that these unexpected results were due to highly selective use of the old grafts in the included studies. In a large UNOS-based registry study from 2018 Halazun et al analyzed the outcome of more than 3100 patients that were transplanted with livers from donors above 70 years[207]. The overall unadjusted PS and GS for this group was clearly inferior when compared to all patients transplanted with donors under 70 years with 3-year GS of 67% and 76%, respectively. However, when adjusting for 7 identified risk factors (HCV+, pretx- hospitalization/ICU/dialysis/previous transplant/previous abdominal surgery and CIT>8 hours), outcome was equally good among patients that received the old livers with 3-year GS of 79% in patients with no risk factors. Another large-scale study originating from UNOS-data published by Haugen et al[208] in 2018 validated the concept of "preferred recipients" produced by the same group in 2007[188] by examining 1861 cases with liver donors above 70 years of age. The Results were compared to a group of patients with donors age 18-39 (ideal liver donors) and a group of 40-69 years (average liver donors). 5-year GS for "preferred recipients of old donor livers", defined as first-time TX, non-urgent patients older than 45 years with BMI below 35, CIT under 8 hours and indication for LT other than HCV, was 75% and not statistically different from patients with "average" or "ideal" grafts. Very recently Ghinolfi et al published a study on 515 patients transplanted with donors above 70 years and compared the outcome to a propensity-matched control group of 448 patients with donors aged 18-69 years[274]. For recipients of old donor livers with no risk factors, defined as donor with DM,

HCV+ recipient and CIT>8 hours, the 5-year GS was 77% and not significantly different from the control group. However, with these risk factors present, 5-year GS fell to 70%.

Almost 50% of our patients was diagnosed with malignancy prior to transplantation, a greater proportion than in most of the comparing studies mentioned above. Although improved during recent eras, the latest ELTR-report from 2018 clearly show inferior survival among patients with HCC compared to LT performed for all indications with 5-year PS of 67% vs 71%, respectively[159]. When restricting the analysis to only patients with non-malignant disease (n=28/54) we achieved 5-year PS/GS of 80/76%, respectively, which should be considered acceptable in the setting of donors above 75 years of age.

### 7.1.2 Biliary complications

We found a rate of biliary complications of almost 30% in the D75 group, significantly higher than 13% in the D20-49 group. The reported incidence of biliary complications (BC) after LT varies widely but are reported to be somewhere between 10 and 30% in most studies[104-106,275,276]. In a recent review of more than 14,000 transplanted patients the total incidence of BCs was 23%, with leakage occurring in 8.5% and strictures in 14.7%[277]. In our D75 group eight patients (14.8%) were diagnosed with leakage and six (11.1%) with strictures. In addition, 2 patients were treated for sludge/stones in the biliary tree, conditions that in many reports are not considered as biliary complications related to the LT-procedure. In 15 of these 16 patients the biliary complications derived from the common bile duct. Earlier studies on the use of old liver donors have raised concerns regarding increased risk for biliary complications, but the available data are conflicting as some studies show increased risk while other reports do not show any association[177,178,205,206,273,278,279]. Some studies have highlighted that old donor age is an independent risk factor for ischemic-type biliary lesions (ITBL) with incidence ranging from

4% to 25%[178,279]. In a study by Ghinolfi et al from 2016 on 88 patients receiving livers from donors aged 80+, the total incidence of biliary complications was 24% and the incidence of ITBL 17%[178]. Three of these 15 patients (20%) had to be listed for re-transplantation after failure of multiple endoscopic interventions to solve the biliary problems. ITBL are postulated to be caused by three main mechanisms: ischemia reperfusion injury (IRI), bile salts and immune-mediated mechanisms[278,280]. Ghinolfi et al identified 3 distinct risk factors for ITBL in their study: donor instability, donor diabetes mellitus and high MELD score in the recipient. Old donors are more sensitive to hemodynamic instability, and hemodynamic factors might contribute to increasing risk for IRI and hence ITBL. Diabetes mellitus in the donor may have impact on the biliary vasculature and further aggravate both biliary ischemia and development of IRI. Unfortunately, we did not collect data on the presence of donor-diabetes in our study. It is however likely that the increased incidence of biliary complications among recipients of old donor grafts in our study in part can be explained by ischemic injury caused by inadequate blood supply to the distal bile ducts. In addition, studies have revealed that there is a general reduction in hepatic blood flow with age[173,182], augmenting the risk for biliary complications. Importantly, all of the biliary complications could be managed by either endoscopic intervention or early reoperation, and none of the patients in our D75 group were re-transplanted due to postoperative biliary complications. Delayed diagnosis of biliary problems can lead to irreversible damage and prompt intervention is of vital importance in recipients of elderly grafts[281].

### 7.1.4 Vascular complications

The total rate of vascular complications in the D75 group was 7.3% and comparable to that of the control group (9.3%). Although the hepatic arterial tree often remains unaffected by donor atherosclerosis[174], in theory it might negatively impact vascular reconstruction and there has

been concerns regarding higher frequency of hepatic artery thrombosis and stenosis in grafts from older donors[175,282]. The reported incidence of HAT lies between 1.6% and 4% in adult recipients[101,102,283]. Two patients (3,7%) both in the D75 group and the control group experienced HAT or stenosis, which is in line with other recent studies showing no increased occurrence of arterial complications in grafts from older compared to younger donors[203,205,206,273,274].

### 7.1.5 Primary nonfunction

According to some studies, older donor livers are perceived to be associated with increased risk for primary nonfunction[209,284,285]. The incidence of PNF after LT in general is reported to be between 1 to 7%[100]. However, we did not experience any incidents of PNF, neither in the D75 group nor in the control group. Furthermore, multiple recent studies have not revealed any difference in PNF between old and younger donors[201,203,205,206,273,274], illustrating that in cases where PNF occurs, it is probably not directly related to the age of the liver graft.

### 7.1.6 Rejection

The rates of T-cell mediated acute rejections are given to be from 10% to 40% and even up 65% in some series[29,107-109,286]. The observed rates in our study with 44% (37% biopsy proven) in the D75 group and 30% (17% biopsy proven) in the control group. These were not statistically significantly different and are within normal ranges for rejection in the current literature. Our findings do not indicate that old donor livers are more prone to develop rejection-episodes, an observation that are in line with earlier findings[199,206,274]

### 7.1.7 Hepatitis C

The observed 1-, 3- and 5-year GS for the 19 patients diagnosed with HCV in the D75 group were 89%, 82% and 82%, respectively, and not inferior to the 17 patients diagnosed with HCV in the control group (p=0.12). These findings are in stark contrast to the substantial body of literature showing increased risk of HCV-relapse and inferior outcome when old liver grafts are given to HCV positive recipients[165,186,188,200,206,257,272,287,288]. Our good results with the combination of HCV positive recipients and donors above 75 years might be due to relatively small sample size and a heterogeneous group of donors. With the advent of the highly effective treatment using direct acting antivirals (DAAs) the negative effect of allocating elderly grafts to HCV positive recipients is likely to disappear[282].

### 7.1.8 Cold ischemia time

Median cold ischemia time in the D75 group was 7.8 hours compared to 8.5 hours in the control group (p=0.55). In general, long CIT is an independent risk factor for delayed graft function and primary nonfunction[289], and large studies have demonstrated that patient survival is negatively affected by CIT over 10-12 hours[285,290]. However, when transplanting livers from elderly donors, multiple studies points to the paramount importance of keeping the CIT as short as possible and at least below 8-9 hours[180,183,186,207,208,274,291]. Old livers are probably more susceptible to cell damage by prolonged ischemia times and the following increased ischemic reperfusion injury[174]. We analyzed if longer CIT times could have impact on PS or GS in the D75 group by stratifying patients into CIT <8 hours (n=30/51) and CIT > 8 hours (n=21/51) and did not find any difference in neither PS nor GS between the two groups. Likewise, we analyzed if there were differences in biliary complications in relation to CIT in both the D75 group and the control group. We observed no differences in CIT between those who developed

biliary complications and those who did not in neither of the groups. Again, these somewhat unexpected results may be caused by the small number of patients in our cohort combined with the fact that CIT were kept below 10 hours in 43 of 54 patients (80%) in the D75 group and in 38 of 54 patients (70%) in the control group.

### 7.1.9 Recipient and donor selection criteria

Although good outcome using old livers without utilizing any specific allocation criteria recently was published by Bertuzzo et al[205], most studies and reviews in the field of old liver donors strongly advocate the importance of matching the right recipient to elderly liver grafts.

In contrast to other organs like heart and kidney, age-related morphological and functional changes seem to be significantly less pronounced in the liver[191,292], probably due to its regenerative capacity, the large functional reserve and dual blood supply. Older donors are undoubtedly extremely heterogeneous as a group. Among donors of the same age, general health status and physiologic reserve including liver status will vary markedly, making it impossible to accept or exclude an elderly donor based on the individual's age alone. Several groups have proposed different sets of criteria for identifying donor/recipient matches likely to produce good outcome[174,188,201,206,208]. Nardo et al[186] suggested the following criteria for accepting an elderly liver: Normal gross appearance and consistency, normal or almost normal liver tests, no relevant histological abnormalities if biopsy is performed and pre-procurement hemodynamic stability with minimal need for vasopressors. Jiménez-Romero et al[206] recommended absence of atherosclerosis in the hepatic and gastroduodenal arteries and donor ICU-stay below 3 days to the above notions by Nardo et al. Routine biopsy before procurement of livers from donors above 80 years is recommended by several authors[186,191,200,201]. In a study from 2007 Segev et al[188] proposed seven criteria for "preferred" recipients who did not incur

additional risk of graft failure or death when transplanted with elderly grafts: First-time LT, non-urgent, above 45 years, BMI < 35, non-HCV indication and CIT < 8 hours. These criteria have recently been validated and found robust by the same group[208]. High MELD-score have proven to be associated with worse outcome in multiple studies. Paterno et al[204] found inferior outcome in recipients with MELD score above 27 receiving grafts from donors above 70 years, an observation that was reinforced by two other studies where a MELD-score above 20[287] or D-MELD above 1400[288] were associated with negative impact on the outcome for recipients of older grafts. Ghinolfi et al[201] suggested that livers above 80 years should be allocated to recipients with MELD score less than 25. Grafts from older donors are often allocated to patients with HCC, however, high donor age has been suggested as a possible contributor to post-LT HCC recurrence due to the increased oxidative stress caused by IRI[293]. Table 5 summarizes prognostic factors for donors and recipients associated with inferior outcome when using old liver grafts.

In terms of accepting donors above 75 years in our study, the general criteria included the presence of normal or slightly elevated liver enzymes before procurement together with normal gross appearance and absence of obvious steatosis judged by the procurement surgeon. No routine biopsies were taken, but zero-biopsies were obtained from 32 of 54 donors (59%) of which 3 biopsies showed moderate (30-60%) steatosis. As shown in Table 3 in Paper 1, the median values for transaminases, bilirubin and INR were within normal ranges among the old donors and the median ICU-stay was 1.5 day. Almost all of the donors were on low-dose vasopressors at time of procurement. However, this was mainly due to protocol-based algorithm for standard treatment of all organ donors, and hence does not indicate that these donors were considered to be hemodynamic unstable. Taken together, most of the donors in the D75 group fulfilled the criteria given by Nardo et al and Jiménez-Romero et al mentioned above.

**TABLE 5. Prognostic factors of poor LT outcome when using elderly donors**

| Risk Factor | Strength of association | Potential means to minimize its impact | Reference |
|---|---|---|---|
| Macrosteatosis <30% | +++ | Liver biopsy | Darius et al (2012)[199]<br>Nardo et al (2004)[186]<br>Ghinolfi et al (2014)[201] |
| CIT > 8-9 hours | ++++ | Shorten ischemia time | Briceno et al (2002)[180]<br>Nardo et al (2004)[186] |
| Abnormal gross appearance | ++ | Clinical judgment performed by the surgeon | Petridis et al (2008)[190]<br>Singhal et al (2010)[191] |
| HCV as an indication | +++++++ | Antiviral therapy with DAA | Berenguer et al (2002)[257]<br>Nardo et al (2004)[186]<br>Cescon et al (2008[200])<br>Kim et al (2011)[287]<br>Franco et al (2013)[288]<br>Chedid et al (2014)[272] |
| MELD-score > 20 | +++++ | Avoid older donors in severely ill patients | Burroughs et al (2009)[294]<br>Kim et al (2011)[287]<br>Darius et al (2012)[199]<br>Ghinolfi et al (2014)[201] |
| ICU-stay > 24 hours | +++ | Avoid older donors in critically ill patients | Busquets et al (2001)[295]<br>Jimènez-Romero et al (2017)[206]<br>Nardo et al (2004[186]) |
| S-glucose level of donor > 11 mmol/l | ++ | Correction of hyperglycemia before procurement | Kim et al (2011)[287]<br>Busuttil et al (2003)[268] |
| Atherosclerosis in hepatic artery | +++ | Surgeon expertise | Nardo et al (2004)[186]<br>Fiel et al (2011)[296]<br>Jimènez-Romero et al (2017)[206] |

**Table 5.** Showing different prognostic factors associated with worse outcome in LT using old donors. Adapted from Berenguer et al, Pushing the donor limits: Deceased donor liver transplantation using organs from octogenarian donors, Liver Transpl. 2017 Oct;23(S1): S22-S26.

All of the patients in the D75 group were first-time transplants. The main criteria for selecting recipients of old grafts were low MELD score or malignant liver disease. However, at times of long waiting lists and in a few cases with high urgency with no other graft available, old liver grafts were also allocated to younger recipients or patients with high MELD-score. Importantly, these transplantations took place over an 11-year period of time, during which allocation

policies evolved. As shown in Table 2 in Paper 1 the median MELD-score in the D75 group was 12, and only 7 patients (13%) had a score above 27. The median patient age was 58.5 years, with 9 patients (17%) aged below 45 years. At the same time median BMI was low at 25.4 and median CIT was 7.8 hours. With the exception of HCV-positivity in 19 of the 54 patients (35%), we can conclude that most of the patients in the D75 group were within the criteria defined by Segev et al as "preferred recipients" of old liver grafts.

### 7.1.10 Limitations of the study

As for all retrospective studies, results from this study should be interpreted with some caution. In addition, it reports data from six different centers for transplantations performed in the period 2001 to 2011 and both clinical practice regarding use of elderly liver donors as well as allocation policies have varied within and between all participating centers. During this time, there has also been improvements in the surgical procedures and peri-operative care. Further, the majority of these old donors come from a Scandinavian population with generally good health-status and low BMI, implicating that our results might not be transferable to regions with different demographics.

## 7.2 Paper 2

### 7.2.1 Patient survival, graft survival and desensitization-techniques

For the whole study group of ABO incompatible transplantations, the 1-, 3-, 5- and 10-year patient survival rates were 85%, 79%, 75% and 59%, respectively. This was not statically inferior to all other LT performed at the two participating centers during the same time period (NLTR group) with the corresponding results of 90%, 84%, 79% and 65%, respectively (p=0.27). For comparison, the overall 1-, 5- and 10-year PS for all registered LTs performed in Europe from 2000 to 2009 were 85%, 73% and 62%, respectively[159]. In addition, 8 of the ABOi LT in our group were re-transplantations due to failure of a first ABOc graft. It is well established that survival after re-transplantation is significantly inferior compared to primary LT[108,159]. Further, 46 of the 61 patients (75%) were transplanted due to either urgent indications or cancer, both conditions that clearly are linked to inferior PS after LT[114,159]. When taking this into account, the overall patient survival among our ABOi patients must be acknowledged as remarkably good. The fact that ABOi LT with deceased donors can be performed with good patient survival was demonstrated long ago by several groups[18,219,220,225]. However, and as demonstrated in many earlier series, the excellent PS among our patients came at the cost of a high rate of re-transplantations, which is evident by the inferior graft survival observed in the ABOi-group: The 1-, 3, 5- and 10-year GS were 71%, 57%, 55% and 51%, respectively, compared to the corresponding GS of 87%, 79%, 73% and 60% in the NLTR group (p=0.0003). Among the non-A2-ABOi patients the GS was as expected even lower with 60% at 1 year and 48% at 3, 5 and 10- year post-LT. Among the recipients that were given A2 grafts the GS was 81%, 67%, 62% and 57% at 1, 3, 5 and 10 years, confirming an already well-known fact that A2 livers given to blood type O recipients produce almost equivalent GS as ABO compatible grafts[243,245]. We could not demonstrate any significant difference in GS between recipients of A2 (n=32) and non-A2 grafts (n=29) in our study, probably related to the relatively small

sample-size. One would expect the GS to be lower among the patients transplanted due to urgent indications (n=33) compared to those with non-urgent conditions. Although GS seemed to be inferior it did not reach statistical significance, also probably due to limited number of patients in each group.

Most other studies published on deceased donor ABOi LT have not distinguished between A2 and non-A2 grafts, making them difficult to interpret in relation to our findings. Results from studies performed in the 80'ies and 90'ies, undertaken when current immunosuppressive agents were unavailable and also less sophisticated techniques in LT were used, makes comparison further difficult. Results after LT have steadily improved over time, although the general improvement in survival appears to be relatively steady since the millennium-change[159]. For instance, in a study from 1995 Farges et al noted a 5-year GS as low as 20% in 43 patients transplanted between 1986 and 1992[18], and Gugenheim et al[225] found that only 40% of 17 ABOi grafts were functioning 2 months after LT. As a consequence of reports showing inferior GS, ABOi LT at many centers was considered inappropriate even in urgent cases and this opinion was applied as an official policy by Eurotransplant during the 1990'ies[297]. However, some centers still occasionally performed ABOi transplantations with acceptable results. Toso et al[16] documented a 5 -year GS of 56% in 14 patients transplanted due to ALF in a study from 2007, not significantly inferior to the ABOc comparison group. In a large UNOS-based study from 2009 Stewart et al[175] on 667 adult ABOi LTs the authors found significantly lower GS compared to ABO compatible transplantations, although the results improved during the latest era. Similar conclusions were drawn in two different meta-analysis on ABOi LT from 2011[239] and 2017[242]. As increasingly good results were published after ABOi kidney transplantation and thereafter living donor ABOi LT in Asia, some centers have utilized the same techniques for desensitization including plasmapheresis, selective immunoadsorption, immunoglobulins

and rituximab in deceased donor ABOi LT. However, there are currently few available reports and all with small patient-cohorts with conflicting results[13,14,17,241].

In Asia, where deceased donors rarely are used due to cultural reasons, protocols for ABOi living donor liver transplantation have been successfully developed with excellent graft survival comparable to what is achieved with ABO compatible LDLT[59,231-233], although hampered with increased risk for biliary strictures[62]. Different techniques for desensitization (DZT) have been used with the goal of removing pre-formed ABO-antibodies and depleting antibody-producing B-cells before the LT procedure, like plasmapheresis, immunoglobulins (IVIG) and eventually rituximab, which significantly improved PS and GS after the introduction in 2003/2004[215,229,231]. Heavy immunosuppressive regimens resulted in increased rate of adverse effects, and use of plasmapheresis is hampered with risks for infections, arrhythmias and water-electrolyte imbalance[232]. Recent studies from South-Korea suggests that excellent results can be achieved with RIT in combination with immunoglobulins eliminating the need for plasmapheresis[232,233].

Regarding use of DZT in our study, the findings are complicated by the fact that these transplants were performed over a long period time (1996-2011) and at two centers with different immunosuppressive protocols in use during the study-period. In total, 28 patients (46%) received IVIG, 18 (30%) received plasmapheresis, 19 (31%) selective immuno-adsorption with glycosorb columns (GSC) and 8 (13%) were treated with both plasmapheresis and GSC at some time during the perioperative course. Specifically, 30 of the patients (49%) received treatment with RIT, given at the day of LT. This means that at least 50% of the patients were not treated according to what we today would consider as adequate pre-ABOi LT conditioning. Most centers performing ABOi LDLT have advocated administration of RIT at least 6-7 days prior to LT, which of course is difficult to achieve in urgent settings. In theory, the effects of RIT on B-cells occur already within 48-72 hours and the effect persists for several

months in transplant recipients[236]. Interestingly, Egawa et el[231] found no correlation between time for RIT administration and outcome in multi-center study on ABOi LDLT from 2014. Specifically, a small subset of urgent patients that received RIT at the day of LT did achieve comparable results to the control group. These findings were confirmed in a single center retrospective study by Shen et al[238] in the setting of deceased donor ABOi LT on 35 patients with acute liver failure that were given RIT at time of transplant in combination with IVIG for 10 days consecutive days postop. They reported an excellent 3-year GS of 80% in this group compared to 86% in the ABOc control group. In contrast, a study on urgent ABOi LT on 22 patients that received no DZT, Zhou et al[240] reported a 5-year GS as low as 21%, almost equivalent to the findings of Farges et al[298] in 1995. Further, the feasibility of ABOi LDLT with urgent patients receiving RIT at time of LT in combination with PP was recently demonstrated in a small series by Kim et al[237]. These findings indicate that there is a potential for improving the outcome after deceased donor ABOi liver transplantation when modern and adequate immunosuppressive treatment with focus on sufficient DZT are utilized. Based on these observations we have now implemented a protocol for ABOi LT using B-cell depletion with RIT in combination with IVIG and standard immunosuppression at both centers (Gothenburg and Oslo).

### 7.2.2 Biliary and vascular complications and re-transplantations

In the whole ABOi cohort, 16 patients (26%) experienced vascular complications, of which 15% was HAT (12.5% in the A2-group and 17% in non-A2 group, p=0.16). This is in line with earlier studies on ABOi LT[15,242] and clearly higher than what is reported as expected liver transplantation[101,102]. In addition, 5 patients (17%) in the non-A group and 1 patient (3%) in the A2 group had portal vein thrombus (PVT), a complication that is reported to occur on only 1-2% after LT[102]. Biliary complications is a well-known problem after ABOi LT described both

in earlier series with deceased donors[221,239,299] as well as in recent studies on ABOi LDLT using modern DZT[62,232,233]. The incidence of biliary complications after LT in general varies widely but are reported to be somewhere between 10 and 30%[104-106,275,276]. BC are known to be more prevalent in LDLT due to often small duct size, often multiple ducts and cutting liver parenchyma[242]. In our study we found a total of 17 patients (28%) suffering from some form of BC, with 7 seven patients (22%) in the A2-group vs 10 patients (34.5%) in the non-A2 group (p=0.27). Of these 17 patients with BC, only 2 (3%) were diagnosed with diffuse intrahepatic biliary strictures (DIBS), which is described as the most common BC related to modern ABOi LDLT[62,231]. We observed 8 patients with leakage and 4 with stenosis of the common biliary duct (CBD), and one patient was diagnosed with both. In addition, 3 patients developed necrosis of the CBD without signs of leakage. It appears that the BC observed in our group of deceased donor ABOi LT differ in nature from what is typically observed in LDLT cases. Notably, 10 of these17 patients were also diagnosed with vascular complications, of which 70% were arterial and it is likely that a vascular component rather than immunological mechanisms have been a contributing factor and perhaps the main cause of the BC seen in our cohort as the bile ducts receive their blood supply solely from the hepatic artery[104,275].

In general, 5-10 of LT-patients will at some time need a new transplant[159]. The total rate of re-tx in our cohort was 23% (14 patients), with five patients (16%) in the A2 group vs nine (31%) in the non-A2 group (p=0.15). The difference was not statistically different, probably due to a relatively small number of patients in each group. Ten of these retransplantations occurred within four months, which is typically earlier than what is observed in LDLT ABOi LT. Biliary complications appeared to be the major reason for re-LT with seven of the 14 patients falling into this category, followed by vascular complications (n=3), acute rejection (n=3) and relapse of HCV (n=1). However, and as mentioned above, it is important to recognize that most of the patients with BC also were diagnosed with vascular problems.

### 7.2.3 Rejections and A/B antibody titers

Twenty-eight (46%) of the patients were treated for acute rejections (including both T-cell mediated and AMR) during the first four weeks after LT (86% (n=24) were biopsy-proven). We could not demonstrate any significant difference in total rejections rates between the A2 group (37.5%) and the non-A2 group (55%, p=0.17). Three patients were diagnosed with biopsy-proven AMR (A1→O, A2→O and B→O) and one was regarded to have AMR on clinical suspicion (B→O), thus the total rate of AMR in our study was 6.5%. The A2 patient with AMR had high levels of pre-transplant DSA and a positive lymphocyte cross-match. Although early reports found that positive cross-match was not associated with impaired outcome, recent studies show that preformed DSA significantly increase the risk for acute rejection and graft loss[300,301]. Since the concept of AMR has evolved considerably during the recent years, it is difficult to compare and relate our AMR-rate of 6.5% to previous studies on deceased donor ABOi. It is reasonable to believe that some of the patients they categorized as having "steroid-resistant" rejections in fact had developed AMR. When considering the published studies on ABOi LDLT from Asia, the reported incidences on AMR are very divergent, ranging from 0% in some series to 23% in others[62,230-233]. It is important to recognize that AMR in LT has been difficult to diagnose, both in ABOi and ABOc cases[302], and that the Banff criteria for rejection-diagnosis and classification did not include specific criteria for AMR until the 2016-version[44]. Some centers in Asia have diagnosed AMR in ABOi LT on the basis of radiological signs (DIBS) in combination with clinical findings (increased liver enzymes, signs of cholangitis) without histological confirmation[215,229] and have reported higher frequencies of AMR although their rates of BC have been in line with other studies reporting much lower rates of AMR. If the biopsies taken from our patients diagnosed with rejection had been re-analyzed using the latest diagnostic criteria, the results regarding AMR might come out

different. AMR after ABOi LDLT is clearly linked to inferior GS[231], and it is no reason to believe that the implications of AMR in deceased donor ABOi LT should be different.

Several reports have failed to show any association between levels of preoperative A/B antibody titers and frequency of AMR[215,238,244]. When PP/selective immunoadsorption is used to lower levels of antibodies prior to LT, the potential for detecting increased titer-levels after LT is by logical means higher than in settings were such treatment is not used. High titer-levels of A/B antibodies in the post-operative course are generally believed to be associated with increased risk for rejection and inferior results[232]. However, results from different studies are inconclusive[16,59,62,231-233,238,241]. Cut-off values being used in ABOi kidney transplantation[61] are probably not transferable to the setting of ABOi LT due to the liver's inherent capability to absorb ABO antibodies and its much larger endothelial surface, illustrated by the fact that combined liver-kidney transplantation has be shown protect the kidney against antibody mediated rejection[303,304]. Egawa et al[231] noted higher frequency of AMR in patients that developed titers above 1/64, which has also been indicated as a possible cut-off level in other studies on ABOi LDLT[232,305]. It is unclear whether these findings can be transferred to DBD ABOi LT were the donor liver has been exposed to the immunological activation cascade associated with the cerebral herniation process Despite a possible correlation between occurrence of rejections and high titer-levels on in our study (Table 7, paper 2), we could not demonstrate any statistically significant association. This analysis was precluded by the fact that few patients were diagnosed with AMR, and that AMR was seen in both patients with high and low postoperative titers. It has been speculated that a partial defect in the accommodation mechanism after ABOi LT may induce an attenuated form of AMR[62], regardless of the antibody titer present in serum, and studies have shown that donor blood type antigens can be present on vascular and biliary epithelium for up to 150 days after transplant[63]. Further, it is known that levels of ABO-antigen expression in the liver grafts vary among individuals[306] making some

livers more vulnerable to high levels of circulating ABO-antibodies than others. The liver is capable of reducing the number of antibodies by Kupffer cell clearance and hence have an inherent protection against antibody-mediated insults[44]. When these factors are taken into consideration, it is not surprising that findings in the published literature diverge.

Although data are based on very few patients (n=8), it is of interest that ABOi LT performed in the presence of anti-HLA antibodies (DSA) seems to result in inferior GS (p=0,058). This may indicate that it is the presence of antibody-driven rejection per se that is the determining factor, and not whether these antibodies are directed against donor HLA or ABO antigens, implicating that ABOi LT in the setting of proven DSA could require closer follow-up and possible more intensive immunosuppressive treatment.

### 7.2.4 Ethical considerations

Concerns have been raised that deceased donor ABOi LT is ethically problematic in the context of its proven inferior graft survival combined with long waiting lists for liver transplantation at most centers around the World[222,307]. The situation with LDLT ABOi LT is of course a different entity where the procedure is justified when no ABOc living donor is available. Currently, the achieved GS in these situations is excellent, and no other candidates on the waiting lists are being disadvantaged. As shown in our study, and in line with multiple other series, satisfactory patient survival has come at a price of high rates of re-TX and therefore have the potential of increasing organ shortage. The practice of ABOi DDLT does not increase the overall donor pool, it only makes it more fluid[222]. In countries with long waiting lists it will in most situations be possible to find a suitable ABOc recipient for all liver grafts being procured.

The fortunate situation in most of the Scandinavian countries with short waiting lists makes allocation less controversial. At times during the study period the wait list in Oslo has been

comprised of very few patients, sometimes even making adequate matching of donor organs difficult. Mortality of wait list patients in Norway and Sweden is very low, especially for blood type A, B and AB recipients[119]. When an ABOi LT has been performed, it has always been carried out with consideration of the other patients on the waiting list, and both centers have been reluctant to use a ABOi graft is this has jeopardized the prognosis of another patient. Although this practice may have resulted in prolonged waiting time for some patients, we have found it ethically acceptable considering that the ABOi transplantations have been life-saving. Also to be noted is that 54 of the 61 procedures were performed in type O recipients, thereby somewhat counteracting the observed prolonged waiting times for blood type O patients in NLTR[119].

### 7.2.5 Comparability between ABOi DDLT and ABOi LDLT

In several sections of this discussion the findings in our study on deceased donor ABOi have been compared to results from the Asian experience with living donor ABOi LT, in most cases due to lack of available similar studies performed with deceased donors. However, it is important to appreciate that there are some important differences between ABOi DDLT and LDLT that may make comparison and transferability problematic. Firstly, organs taken from DBD donation are exposed to the major immunological activation linked to brain death, potentially increasing the antigenicity of the donor liver[308]. Secondly, most studies on ABOi LDLT have been performed on relatively healthy patients with low MELD-score[215,232]. In contrast, our group of patients had much higher median MELD-score where 33 of 61 patients were transplanted due to urgent indications. It is well established that high MELD-score is associated with inferior outcome. Thirdly, the age distribution in the Asian countries performing most of the ABOi LDLTs show a higher number of young LT-patients than in Scandinavia[309]. From the LDLT experience, it has been reported that advanced recipient age is a separate risk

factor for inferior outcome in ABOi LT[229]. Fourthly, in a large-scale meta-analysis from 2017 Lee et al[242] documented higher risk for T-cell mediated rejection in ABOi DDLT compared to LDLT, probably resulting from higher genetic similarities between donor and recipient. Lastly, and perhaps most prominent, the cold ischemia times are significantly shorter in LDLT, often in the range of only 1-2 hours[59]. Longer CIT-times have been associated with inferior outcomes in several studies[8,310-312]. Specifically, Lee et al[233] found that longer CIT was the only prognostic factor for overall outcome in their group of ABOi LDLT patients, suggesting that ABOi grafts are more susceptible to prolonged CIT than ABOc grafts.

### 7.2.6 Limitations of the study

This was an uncontrolled retrospective observational study where the transplantations were performed at two different centers over a period of 14 years. The patients included in the study represented a heterogeneous group in terms of diagnosis and clinical settings, and the immunosuppression in use have for natural reasons varied and evolved over time. During this period there has been improvements in both surgical technique, perioperative handling of LT patients as well as development of new immunosuppressive drugs and techniques for desensitization. There has not been a uniform, common protocol at the two participating centers, and most of the patients involved were not treated according to what we today would consider as appropriate in the setting of ABOi LT. Also, to be considered is that the epidemiology and disease panorama reflects the Scandinavian population, and that our results therefore might not be amendable to other parts of the World. Perhaps foremost, this series of ABOi liver transplantations have been performed at two centers with generally short waiting lists and relatively few high-MELD patients on the lists, making it possible to perform such procedures without risking the life other patients.

## 7.3 Paper 3

### 7.3.1 Patient- and graft survival

For the whole study-group the 1-, 5- and 10-year Kaplan Meier patient survival rates were 83%, 65% and 52%, respectively. The corresponding results for the subgroup of patients with malignant disease (n=7) were 85%, 57% and 38%, while both 1-, 5- and 10-year PS for patients in the benign group (n=6) were 80%. Three patients were re-transplanted very shortly after the primary procedure due inferior quality of the liver grafts used at the primary transplantation. When these three PNF-incidents are taken out of the equation, no other re-transplants occurred and hence survival rates for GS are equal to the results for PS given above. The number of patients in our group is low, which means that the readings from these Kaplan Meier estimates of survival must be interpreted with caution. However, as illustrated in Table 1 of the paper, 8 of the 13 patients (62%) were still alive at the end of the study period, with a median follow-up of 70.5 months (range 2.2-171).

There are very few published studies on urgent LT in the setting of posthepatectomy liver failure or LT due to iatrogenic liver injuries. This makes comparison of survival outcome in our group against a similar group of patients difficult. Earlier studies have mainly described LT after fatal bile duct injuries acquired during cholecystectomy[313-320]. In addition, smaller series or even single-case studies have been published on LT in the aftermath of iatrogenic injuries to the portal vein during bariatric surgery[321], ALF after coiling of the hepatic artery[322] and due to bleeding-complications after insertion of transjugular intrahepatic portosystemic shunts (TIPS)[323]. Urgent LT after fatal injuries to the portal vein and hepatic artery during open adrenalectomy have been described[316,324], as well as urgent LT after uncontrollable bleeding related to resection surgery[322,325]. Results from these mostly small-scale studies have been highly diverse, as illustrated in Table 4 in paper 3. However, clearly the LT-procedure have been life-saving for a number of patients despite disastrous outcomes for others.

A total of 346 patients are registered in ELTR with "Traumatic acute hepatic failure" as reason for LT during the last 15 years with a recorded 10-year GS of only 36%[159], but further details regarding this group and whether the patients are similar to ours are unknown (Table 5). According to the latest ELTR report, the overall survival after LT in Europe for the period from 2000 to 2014 (n=84 616) is approximately 85% at 1 year, 73% at 5 year and 61% at 10 year[159], similar to the results seen in NLTR[119]. Interestingly, data from ELTR shows that the most important gain in survival during the recent years are observed for primary liver tumors, liver metastases and acute liver failure. Obviously, an observed 10-year survival of 80% in our group of benign patients justify the choice of offering these patients a liver transplant. On the other end of the scale is the corresponding and clearly inferior survival rate of only 38% in the subgroup with malignant conditions. Among the patients that died after LT were one patient with duodenal cancer (#13) and two patients with CRLM (#10 and #11) that preoperatively were in a condition with large tumors and high levels of carcinoembryonic antigen (CEA) in serum, both known markers for increased risk of cancer-recurrence[19]. In a study with small number of patients, 3 deaths will naturally have a significant impact on the results, and retrospectively neither of these patients should have been offered a transplant. Of note however, is that patient # 10 survived for almost 4 years after LT, while patient #13 died after 2 years of follow-up. One other patient with CRLM (#8) died 6.5 years post-LT, while the last CRLM-patient (#9) was still alive after 72 months. Even though survival in some of our patients may be considered as unacceptable, it is worth mentioning that data show that both 1$^{st}$ and 2$^{nd}$ re-transplants for any cause in Europe are associated with equally worse or even inferior outcome (Table 6).

**Table 6. Graft survival in rescue group vs various selected groups from ELTR last 15 years**

| Patient group | 5-year PS/GS (%) | 10-year PS/GS (%) |
|---|---|---|
| Rescue group Oslo - total (n=13) | 65/65 | 52/52 |
| Rescue group Oslo - benign (n=6) | 80/80 | 80/80 |
| Rescue group Oslo - malign (n=7) | 57/57 | 38/38 |
| Acute hepatic failure ELTR (n=6240) | 69/62 | 62/55 |
| Traumatic acute hepatic failure ELTR (n=346) | 51/41 | 44/36 |
| Cholestatic disease ELTR (n=8439) | 81/74 | 73/63 |
| Cirrhosis ELTR (n=45566) | 72/68 | 59/55 |
| Primary liver tumors ELTR (n=17329) | 67/64 | 53/49 |
| Secondary liver tumors ELTR (n=395) | 61/57 | 46/44 |
| Metabolic disesase ELTR (n=5336) | 80/74 | 71/63 |
| First retransplantation ELTR (n=3653[a]/1809[b]) | na/48 | na/39 |
| Second retransplantation ELTR (n=391[a]/218[b]) | na/42 | na/34 |

ELTR, European Liver Transplant Registry; PS, patient survival; GS, graft survival

[a] no. of patients exposed at 5 years, [b] no. of patients exposed at 10 years. na, not applicable.

Data are collected from the latest ELRT-report[158]

### 7.3.2 Primary nonfunction

Three patients (23%) in our study experienced PNF or dysfunction requiring early re-TX, which is higher than the reported incidence of 1% to 7%[100]. However, all these incidents were clearly related to marginal quality of the liver graft without signs of technical issues as cause for the graft failure. Therefore, for the three patients that were re-transplanted due to PNF, the calculation of PS/GS is based on the last liver graft.

### 7.3.3 Vascular and biliary complications and rejection rate

One patient (8%) was diagnosed with HAT and one patient suffered from PVT after transplantation. These incidences are higher than what is reported to be within normal range for vascular complications (1.6-4% for HAT and 1-2% for PVT[101,102]), but further conclusions are of limited value due to the small number of patients included in the study.

A total of four patients (31%) had biliary complications (two patients with leakage from the cystic duct and two with leakage from the hepatico-jejunostomy). Reported incidence of BC in LT lies somewhere between 10% to 30% in most studies[104-106]. In a recent meta-analysis, the rate of leakage was observed in 8.5% of the cases[277], implying that urgent patients similar to those in our study might be more prone to experience problems with bile leakage post LT.

Two patients (15%) were treated for T cell-mediated rejections after LT, which is in line with reported incidences of rejection in the literature[29].

### 7.3.4 Ethical considerations and selection of appropriate patients for rescue LT

Rescue LT in situations with iatrogenic injuries and posthepatectomy liver failure leads not only to medical challenges, but potentially pose challenging ethical concerns. These concerns are of course especially prominent in situations with organ scarcity and long waiting lists, a situation that most transplant centers in the world today encounter. Most will agree to that rescue LT after iatrogenic injuries in patients with benign diagnosis is justified and clearly less controversial than LT due to surgical complications in individuals suffering from malignant conditions. However, our study shows that it is difficult to predict the outcome in both circumstances, and that even patients undergoing surgery for malignant diseases outside internationally accepted LT-criteria can achieve reasonably good (patient #8 and #10) and excellent long-term survival after rescue LT (patient #9). Results from our institutional experience on LT in the setting of CRLM shows that it is possible to achieve good long-term results with approximately 60% 5-year PS in selected patients[20]. These findings are confirmed in a second study (SECA) soon to be published. From the SECA 1 trial, four clinical factors emerged as predictive of poor survival: Diameter of the largest tumor ≥ 55mm, pre-transplant CEA level > 80 $\mu$g/l, progressive disease on chemotherapy and time from resection of the

primary tumor to transplantation less than 2 years[19]. As illustrated in Table 3 in paper 3, the two (patients #8 and #9) CRLM-patients with best survival had both small tumors, low CEA and response to preoperative chemotherapy. Conversely, the two other patients with CRLM (patient #10 and #11) who died shortly after transplantation were outside these criteria. Although further studies with larger patient samples with longer follow-up are needed in order for this diagnosis to be established as an internationally accepted indication for LT, it is of interest that data from ELTR illustrates that transplantations in patient groups with similar or even worse 5-year survival have been performed regularly. For instance, 5-year PS for LT performed in the setting of biliary tract CCA (Klatskins tumor) during the last 15 years (n=245) is 47%, while the overall 5-year GS for first re-TX with all diagnosis included (n=3653) is 48%[159]. Two patients in our group (patient #4 and #13) underwent rescue LT in the aftermath of pancreatoduodenectomy (Whipple's procedure), both experiencing short post LT survival. In addition, one patient (patient #11) had rescue LT due to complications in the setting of auto-transplantation for CRLM resulting in a very complicated postoperative course and death after 9 months. As discussed in section **7.3.1,** it is probably wise to refrain from performing rescue LT in cases like these three.

These moments taken into consideration, the choice of performing rescue LT in our patient cohort seems justified in most of the patients and information from our findings can contribute to the discussion on whether the current systems for prioritization benefit the patients with most life-gain after LT or not.

### 7.3.5 Limitations of the study

Firstly, although representing one of the largest published series on rescue LT, this is a retrospective uncontrolled study from a single center with all the inherent biases associated with

this type of study design. However, the very nature of this topic makes a randomized controlled trial impossible to perform. Secondly, the patient cohort is small and heterogeneous. Thirdly, the patients were highly selected and underwent rescue LT without any study protocol and specific criteria, making direct transferability of results to other environments potentially problematic. As earlier mentioned, the fortunate donor situation in Norway and short waiting times have facilitated the choice of performing these urgent transplantations. Thus, in situations with organ shortage or other systems for allocation the external validity of the study should be considered with caution.

# 8 CONCLUDING REMARKS AND FUTURE PERSPECTIVES

The good results obtained over the years with LT has in most parts of the developed world led to a situation with severe lack of available donor organs. This shortage of organs has forced the transplant community to seek ways of expanding the donor pool using extended criteria donors. One of the largest potential sources of such donors is the elderly population, and the magnitude of this potential is substantial. In 1993, only 1% of the livers used for LT in Europe came from donors above 70 years, while this rate had increased to 10% in 2005 and 20% by 2015[159]. Ideally, every patient should receive a perfect organ, in the past described as a liver harvested from a donor under 50 years in good clinical condition. However, analysis from large databases have revealed that in fact, every 10-year increase in donor age starting already at age 20 results in worse patient survival relative to younger grafts[207]. At the same time, average donor age seems to increase all over the Western world[158,159,182,206] along with aging populations and fewer young patients dying from diseases and accidents. Multiple studies mentioned in this thesis demonstrate the feasibility of producing good results using old donors when certain precautions regarding selection of these donors and matching with the recipients are undertaken. Together with other donor risk factors, increased donor age should be considered as a continuum of risk rather than focusing on arbitrary upper age-limits defining usable donors. This increased risk associated with elderly donors can be balanced by keeping most other risk factors related to recipient-choice and logistics low. Also to be kept in mind is that several studies show higher survival rates using old livers instead of waiting for the ideal graft[205], in coherence with the statement from prof Henry Wismuth: "The highest risk for a patient needing a new liver is the risk of never to be transplanted"[326]. Future studies should focus on further development of biomarkers and scoring-systems for identifying both old donor livers likely to produce optimal results as well as the recipients that will benefit the most. The use of machine perfusion (MP)

is a promising technique for optimizing results when using old and other types of ECD liver grafts. There are relatively few published reports on use of MP in clinical settings, but some small series show promising results[327-332] and multicenter trials are underway with currently 17 studies registered on https:/clinicaltrials.gov. MP has the potential to reduce IRI and improve hemodynamics and potentially "resuscitating" marginal organs[333-335]. The concept of real-time metabolomic profiling by high-resolution magic-angle-spinning nuclear MR (HR-MAS-NMR) is a new technique that seems able to predict early graft dysfunction and in the future might be helpful in evaluating the efficiency of graft resuscitation on MP and to objectively select old grafts suitable for LT[336].

Except from one large registry-based report from 2009[15], our study represents the largest published series of ABOi LT utilizing liver grafts from deceased donors. The nature of the topic itself makes it impossible to conduct controlled, randomized studies examining the true effect of ABO incompability vs ABO combability in DDLT. The main findings in our study were that it is possible to achieve good patient survival and reasonably good graft survival using ABOi DDLT. Although better than in several older studies on the topic, graft survival was inferior, and the procedure is associated with increased risk for rejections and postoperative complications. Hence, in some cases patient survival comes at a price of retransplantation with a new graft, meaning that one patient has been given an extra liver that might could have been used for another recipient on the waiting list. This rises ethical dilemmas and questions whether ABOi transplantations are justifiable or not. In most parts of the world liver waiting list are long, which forces the transplant communities to manage the scarce resource of donor livers in the best possibly ethical and righteous way. Naturally, ABOi transplantation does not increase the overall DDLT donor pool and as a general rule, and as stated by Dr Starlz almost 40 years ago, ABOi transplantation should be limited to those for whom an ABOc transplantation is not

an option[219]. At the same time, our study confirmed earlier findings on the safety of transplanting A2 livers to blood type O recipients. This can safely be done in acute settings with good results, although some of the same ethical dilemmas discussed above can arise if such a transplant will disadvantage a blood type A recipient waiting. In situations with short waiting list making it difficult to match an A2 liver to an appropriate type A recipient, these A2 grafts might be allocated to elective type O recipients with minimal extra risk.

In those very few cases that ABOi DDLT still will be performed, it will be in everyone's interest that the outcome is as good as possible. New immunosuppressive strategies with focus on improved desensitization-techniques through plasmapheresis, use of rituximab and immunoglobulins have radically changed outcome in ABOi LDLT. It is reasonable to believe that these measures also can produce even better outcomes in ABOi DDLT, although not to the same extent as the undisputable success with elective ABOi LDLT in Asia. Our center in Oslo has now implemented Gothenburg's protocol for ABOi LT utilizing DZT, and preliminary results seem promising.

AMR in ABOi recipients may be caused by antibody-producing plasma cells, not being removed by RIT. Consequently, plasma cell-depleting agents like the proteasome inhibitor bortezomib might play a role in future studies on ABOi LT. Another emerging concept in transplantation across the ABO barrier is the inhibition of complement activation upon binding of antibodies. Eculizumab, a monoclonal antibody and inhibitor of complement activation, could on theoretical basis play a role in future treatment in ABOi liver transplantations. Neither of these drugs have so far been tested in clinical trials on LT. A third novel strategy that could come into clinical use is reduction of blood group antigen levels in the liver graft by ex vivo infusion of the enzyme endo-beta-galactosidase[61].

The patient group that is included in the study on urgent rescue LT represents patients that from time to time will be encountered in hepatobiliary and transplant centers. The published literature on this topic is scarce, and mainly focus on iatrogenic injuries caused during laparoscopic cholecystectomies. Patient survival in some of the previous studies on urgent LT has not been encouraging, with short-term mortality up to 80%[319,322]. However, these results may at least partly be related to the fact that the patients involved have been in a dismal clinical condition at time of LT with infections, sepsis or multiorgan failure, which are predicators of inferior outcome in any LT patient regardless of the primary disease. If the choice is made of going forward with LT in cases like those presented in our study, we believe that it is of crucial importance to have these patients transplanted as quickly as possible in order to achieve an optimal outcome. As observed in our study, long time patient survival is achievable even in the setting of malignant conditions. However, in a retrospective view, some of the patients in the study suffered from a protracted postoperative course and short survival, and probably should not have be transplanted. In conclusion, we believe that LT for patients with non-malignant disease suffering liver failure due to portal vein and hepatic artery is clearly justified. In addition, highly selected patients with malignant conditions can achieve acceptable, and in some cases exceptionally good, outcome after rescue LT. However, it is doubtful whether patients with proven malignant disease beyond established criteria for LT should be offered liver transplantation in settings where the primary operation has resulted in irreversible liver failure. Thus, if rescue LT is considered after surgery for malignant conditions, a histology report should be available for confirmation of the diagnosis and for evaluation of tumor stage and prognosis before the final decision for LT is made.

Although our study includes only 13 patients, it still represents one of the largest series presented. Data from ELRT indicates that such rescue transplantations have been performed multiple times at different centers across Europe, but the results have not been published. Larger

sample sizes are clearly needed to make more robust conclusions on whether rescue transplantations should be performed and in what kind of patients the procedure is justified. Accordingly, transplant centers possessing experience with this patient group should publish their results in an effort to produce a broader fundament for decision-making in these difficult cases.

As described in this thesis, there has been major developments and refinements in both surgical technique, immunosuppression, peri-operative handling and selections of patients found suitable for LT since the start of clinical liver transplantation in humans in the early 60ies. A long with better results and less risk related to the procedure the number of diseases and indications leading to listing for LT has been expanded and stretched, resulting in a significant gap between the number of available organs and patients in need for a transplant. The use of extended criteria donors, and in particular older donors, stands out as the most important way to remedy this challenge. Not long age donors above 50 years of age was considered inappropriate for use in liver transplantation. As addressed in this thesis, there has been a clear change in this policy during the last 10-15 years, and results utilizing selected liver donors above 75 and even 80 years seems to be highly acceptable. In addition, the concept of machine perfusion might have the potential to convert liver grafts initially considered unsuitable into usable organs. At the same time, expanding the donor pool to ABO incompatible donors have been done with great success with living donors in Asia. Results with deceased donor ABOi LT have so far mostly been inferior and should probably be reserved to those few urgent cases where no ABO compatible graft is available. As seen in our last paper, certain patients with post-hepatectomy and iatrogenic liver injuries treated with urgent LT achieve very good outcomes, even in the setting of malignant disease. These findings might challenge the established guidelines for which patient groups that should be offered transplantation at the

expense of others. Although expanding the donor pool by using more ECD donors in the future can provide more liver grafts, the greatest challenge in the field of liver transplantation will still amount to balancing the availability of organs against the large number of patients on the waiting lists in the best possible and righteous way.

## ERRATA

### Paper I

**Corrections in paper text are marked in bold:**

Page 2537, right column, 1. section under "Results"

*The paper reads:* "..CIT less than 10 hours versus 38 hours (70.4%) in the D20-49 group."

*The correct should be*: "..CIT less than 10 hours versus 38 **patients** (70.4%) in the D20-49 group."

Page 2537, right column, 2. section under "Results"

*The paper reads*: ", and 9 liver grafts (9.4%) showed moderate steatosis"

*The correct should be*: ", and **3** liver grafts (9.4%) showed moderate steatosis"

Page 2540, right column, 3. section under "Discussion"

*The paper reads:* "Median CIT in the D75 group was approximately 8.2 hours.."

*The correct should be:* "Median CIT in the D75 group was approximately **7.8** hours.."

**REFERENCES**


1 Newsletter Transplant - International figures on donation and transplantation 2016. (The Council of Europe European Committe, 2017).

2 https://optn.transplant.hrsa.gov/data/ *accessed 2709 2018*.

3 Kim, W. R. *et al.* OPTN/SRTR 2011 Annual Data Report: liver. *American journal of transplantation : official journal of the American Society of Transplantation and the American Society of Transplant Surgeons* **13 Suppl 1**, 73-102, doi:10.1111/ajt.12021 (2013).

4 Wertheim, J. A., Petrowsky, H., Saab, S., Kupiec-Weglinski, J. W. & Busuttil, R. W. Major challenges limiting liver transplantation in the United States. *American journal of transplantation : official journal of the American Society of Transplantation and the American Society of Transplant Surgeons* **11**, 1773-1784, doi:10.1111/j.1600-6143.2011.03587.x (2011).

5 Ghobrial, R. M. *et al.* Donor morbidity after living donation for liver transplantation. *Gastroenterology* **135**, 468-476, doi:10.1053/j.gastro.2008.04.018 (2008).

6 Marsh, J. W., Gray, E., Ness, R. & Starzl, T. E. Complications of right lobe living donor liver transplantation. *Journal of hepatology* **51**, 715-724, doi:10.1016/j.jhep.2009.04.023 (2009).

7 Zambelli, M. *et al.* Full-right-full-left split liver transplantation: the retrospective analysis of an early multicenter experience including graft sharing. *American journal of transplantation : official journal of the American Society of Transplantation and the American Society of Transplant Surgeons* **12**, 2198-2210, doi:10.1111/j.1600-6143.2012.04071.x (2012).

8 Feng, S. *et al.* Characteristics associated with liver graft failure: the concept of a donor risk index. *American journal of transplantation : official journal of the American Society of Transplantation and the American Society of Transplant Surgeons* **6**, 783-790, doi:10.1111/j.1600-6143.2006.01242.x (2006).

9 Aseni, P. *et al.* A Prospective Policy Development to Increase Split-Liver Transplantation for 2 Adult Recipients: Results of a 12-Year Multicenter Collaborative Study. *Annals of surgery*, doi:10.1097/SLA.0b013e31827da6c9 (2013).

10 Vodkin, I. & Kuo, A. Extended Criteria Donors in Liver Transplantation. *Clinics in liver disease* **21**, 289-301, doi:10.1016/j.cld.2016.12.004 (2017).

11 Flores, A. & Asrani, S. K. The donor risk index: A decade of experience. *Liver transplantation : official publication of the American Association for the Study of Liver Diseases and the International Liver Transplantation Society* **23**, 1216-1225, doi:10.1002/lt.24799 (2017).

12 Yilmaz, S. *et al.* ABO-incompatible liver transplantation in acute and acute-on-chronic liver failure. *Hepato-gastroenterology* **60**, 1189-1193, doi:10.5754/hge11289 (2013).

13 Maitta, R. W., Choate, J., Emre, S. H., Luczycki, S. M. & Wu, Y. Emergency ABO-incompatible liver transplant secondary to fulminant hepatic failure: outcome, role of TPE and review of the literature. *Journal of clinical apheresis* **27**, 320-329, doi:10.1002/jca.21244 (2012).

14 Saliba, F. *et al.* Successful long-term outcome of ABO-incompatible liver transplantation using antigen-specific immunoadsorption columns. *Therapeutic apheresis and dialysis : official peer-reviewed journal of the International Society for Apheresis, the Japanese Society for Apheresis, the Japanese Society for Dialysis Therapy* **14**, 116-123, doi:10.1111/j.1744-9987.2009.00792.x (2010).

15 Stewart, Z. A. *et al.* ABO-incompatible deceased donor liver transplantation in the United States: a national registry analysis. *Liver transplantation : official publication of the American Association for the Study of Liver Diseases and the International Liver Transplantation Society* **15**, 883-893, doi:10.1002/lt.21723 (2009).

16 Toso, C. *et al.* ABO-incompatible liver transplantation for critically ill adult patients. *Transplant international : official journal of the European Society for Organ Transplantation* **20**, 675-681, doi:10.1111/j.1432-2277.2007.00492.x (2007).

17 Boberg, K. M., Foss, A., Midtvedt, K. & Schrumpf, E. ABO-incompatible deceased donor liver transplantation with the use of antigen-specific immunoadsorption and anti-CD20 monoclonal antibody. *Clinical transplantation* **20**, 265-268, doi:10.1111/j.1399-0012.2005.00470.x (2006).

18 Farges, O. *et al.* The use of ABO-incompatible grafts in liver transplantation: a life-saving procedure in highly selected patients. *Transplantation* **59**, 1124-1133 (1995).

19 Hagness, M., Foss, A., Egge, T. S. & Dueland, S. Patterns of recurrence after liver transplantation for nonresectable liver metastases from colorectal cancer. *Annals of surgical oncology* **21**, 1323-1329, doi:10.1245/s10434-013-3449-9 (2014).

20 Hagness, M. *et al.* Liver transplantation for nonresectable liver metastases from colorectal cancer. *Annals of surgery* **257**, 800-806, doi:10.1097/SLA.0b013e3182823957 (2013).

21 Dueland, S. *et al.* Chemotherapy or liver transplantation for nonresectable liver metastases from colorectal cancer? *Annals of surgery* **261**, 956-960, doi:10.1097/sla.0000000000000786 (2015).

22 Line, P. D., Hagness, M., Berstad, A. E., Foss, A. & Dueland, S. A Novel Concept for Partial Liver Transplantation in Nonresectable Colorectal Liver Metastases: The RAPID Concept. *Annals of surgery* **262**, e5-9, doi:10.1097/sla.0000000000001165 (2015).

23 Dueland, S., Foss, A., Solheim, J. M., Hagness, M. & Line, P. D. Survival following liver transplantation for liver-only colorectal metastases compared with hepatocellular carcinoma. *The British journal of surgery* **105**, 736-742, doi:10.1002/bjs.10769 (2018).

24 Litman, G. W., Cannon, J. P. & Dishaw, L. J. Reconstructing immune phylogeny: new perspectives. *Nature reviews. Immunology* **5**, 866-879, doi:10.1038/nri1712 (2005).

25 Marshall, J. S., Warrington, R., Watson, W. & Kim, H. L. An introduction to immunology and immunopathology. *Allergy, asthma, and clinical immunology : official journal of the Canadian Society of Allergy and Clinical Immunology* **14**, 49, doi:10.1186/s13223-018-0278-1 (2018).

26 Costantino, C. M., Spooner, E., Ploegh, H. L. & Hafler, D. A. Class II MHC self-antigen presentation in human B and T lymphocytes. *PloS one* **7**, e29805, doi:10.1371/journal.pone.0029805 (2012).

27 Kearney, E. R. *et al.* Antigen-dependent clonal expansion of a trace population of antigen-specific CD4+ T cells in vivo is dependent on CD28 costimulation and inhibited by CTLA-4. *Journal of immunology (Baltimore, Md. : 1950)* **155**, 1032-1036 (1995).

28 Ford, M. L., Adams, A. B. & Pearson, T. C. Targeting co-stimulatory pathways: transplantation and autoimmunity. *Nature reviews. Nephrology* **10**, 14-24, doi:10.1038/nrneph.2013.183 (2014).

29 Charlton, M. *et al.* International Liver Transplantation Society Consensus Statement on Immunosuppression in Liver Transplant Recipients. *Transplantation* **102**, 727-743, doi:10.1097/tp.0000000000002147 (2018).

30 Rosen, H. R. Transplantation immunology: what the clinician needs to know for immunotherapy. *Gastroenterology* **134**, 1789-1801, doi:10.1053/j.gastro.2008.02.062 (2008).

31 Knechtle, S. J. & Kwun, J. Unique aspects of rejection and tolerance in liver transplantation. *Seminars in liver disease* **29**, 91-101, doi:10.1055/s-0029-1192058 (2009).

32 Alex Bishop, G., Bertolino, P. D., Bowen, D. G. & McCaughan, G. W. Tolerance in liver transplantation. *Best practice & research. Clinical gastroenterology* **26**, 73-84, doi:10.1016/j.bpg.2012.01.003 (2012).

33 Adams, D. H. & Neuberger, J. M. Patterns of graft rejection following liver transplantation. *Journal of hepatology* **10**, 113-119 (1990).

34 Levitsky, J. *et al.* Acute Rejection Increases Risk of Graft Failure and Death in Recent Liver Transplant Recipients. *Clinical gastroenterology and hepatology : the official clinical practice journal of the American Gastroenterological Association* **15**, 584-593.e582, doi:10.1016/j.cgh.2016.07.035 (2017).

35 Mor, E., Gonwa, T. A., Husberg, B. S., Goldstein, R. M. & Klintmalm, G. B. Late-onset acute rejection in orthotopic liver transplantation--associated risk factors and outcome. *Transplantation* **54**, 821-824 (1992).

36 Thurairajah, P. H. *et al.* Late acute liver allograft rejection; a study of its natural history and graft survival in the current era. *Transplantation* **95**, 955-959, doi:10.1097/TP.0b013e3182845f6c (2013).

37 Demetris, A. J. Spectrum of chronic hepatic allograft rejection and arteriopathy and the controversy of centrilobular necrosis. *Liver transplantation : official publication of the American Association for the Study of Liver Diseases and the International Liver Transplantation Society* **6**, 102-103, doi:10.1002/lt.500060106 (2000).

38 Blakolmer, K. *et al.* Chronic liver allograft rejection in a population treated primarily with tacrolimus as baseline immunosuppression: long-term follow-up and evaluation of features for histopathological staging. *Transplantation* **69**, 2330-2336 (2000).

39 Gupta, P. *et al.* Risk factors for chronic rejection after pediatric liver transplantation. *Transplantation* **72**, 1098-1102 (2001).

40 Demetris, A. J., Zeevi, A. & O'Leary, J. G. ABO-compatible liver allograft antibody-mediated rejection: an update. *Current opinion in organ transplantation* **20**, 314-324, doi:10.1097/mot.0000000000000194 (2015).

41 Demetris, A. J. *et al.* Immunopathology of antibodies as effectors of orthotopic liver allograft rejection. *Seminars in liver disease* **12**, 51-59, doi:10.1055/s-2007-1007376 (1992).

42 Wesson, R. N., Etchill, E. W. & Garonzik-Wang, J. Application and interpretation of histocompatibility data in liver transplantation. *Current opinion in organ transplantation* **22**, 499-504, doi:10.1097/mot.0000000000000450 (2017).

43 Hogen, R., DiNorcia, J. & Dhanireddy, K. Antibody-mediated rejection: what is the clinical relevance? *Current opinion in organ transplantation* **22**, 97-104, doi:10.1097/mot.0000000000000391 (2017).

44 Demetris, A. J. *et al.* 2016 Comprehensive Update of the Banff Working Group on Liver Allograft Pathology: Introduction of Antibody-Mediated Rejection. *American journal of*

*transplantation : official journal of the American Society of Transplantation and the American Society of Transplant Surgeons* **16**, 2816-2835, doi:10.1111/ajt.13909 (2016).

45 O'Leary, J. G. *et al.* Antibody-mediated rejection as a contributor to previously unexplained early liver allograft loss. *Liver transplantation : official publication of the American Association for the Study of Liver Diseases and the International Liver Transplantation Society* **20**, 218-227, doi:10.1002/lt.23788 (2014).

46 Kim, P. T., Demetris, A. J. & O'Leary, J. G. Prevention and treatment of liver allograft antibody-mediated rejection and the role of the 'two-hit hypothesis'. *Current opinion in organ transplantation* **21**, 209-218, doi:10.1097/mot.0000000000000275 (2016).

47 Levitsky, J. *et al.* Donor-Specific HLA Antibodies in Living Versus Deceased Donor Liver Transplant Recipients. *American journal of transplantation : official journal of the American Society of Transplantation and the American Society of Transplant Surgeons* **16**, 2437-2444, doi:10.1111/ajt.13757 (2016).

48 Musat, A. I. *et al.* Pretransplant donor-specific anti-HLA antibodies as predictors of early allograft rejection in ABO-compatible liver transplantation. *Liver transplantation : official publication of the American Association for the Study of Liver Diseases and the International Liver Transplantation Society* **19**, 1132-1141, doi:10.1002/lt.23707 (2013).

49 Kaneku, H. *et al.* De novo donor-specific HLA antibodies decrease patient and graft survival in liver transplant recipients. *American journal of transplantation : official journal of the American Society of Transplantation and the American Society of Transplant Surgeons* **13**, 1541-1548, doi:10.1111/ajt.12212 (2013)

50 O'Leary, J. G. *et al.* The Influence of Immunosuppressive Agents on the Risk of De Novo Donor-Specific HLA Antibody Production in Solid Organ Transplant Recipients. *Transplantation* **100**, 39-53, doi:10.1097/tp.0000000000000869 (2016).

51 Taner, T. *et al.* Prevalence, course and impact of HLA donor-specific antibodies in liver transplantation in the first year. *American journal of transplantation : official journal of the American Society of Transplantation and the American Society of Transplant Surgeons* **12**, 1504-1510, doi:10.1111/j.1600-6143.2012.03995.x (2012).

52 O'Leary, J. G. *et al.* Preformed class II donor-specific antibodies are associated with an increased risk of early rejection after liver transplantation. *Liver transplantation : official publication of the American Association for the Study of Liver Diseases and the International Liver Transplantation Society* **19**, 973-980, doi:10.1002/lt.23687 (2013).

53 O'Leary, J. G. *et al.* The role of donor-specific HLA alloantibodies in liver transplantation. *American journal of transplantation : official journal of the American Society of Transplantation and the American Society of Transplant Surgeons* **14**, 779-787, doi:10.1111/ajt.12667 (2014).

54 O'Leary, J. G. *et al.* Impact of IgG3 subclass and C1q-fixing donor-specific HLA alloantibodies on rejection and survival in liver transplantation. *American journal of transplantation : official journal of the American Society of Transplantation and the American Society of Transplant Surgeons* **15**, 1003-1013, doi:10.1111/ajt.13153 (2015).

55 O'Leary, J. G. *et al.* High mean fluorescence intensity donor-specific anti-HLA antibodies associated with chronic rejection Postliver transplant. *American journal of transplantation : official journal of the American Society of Transplantation and the American Society of Transplant Surgeons* **11**, 1868-1876, doi:10.1111/j.1600-6143.2011.03593.x (2011).

56 O'Leary, J. G. *et al.* Proposed Diagnostic Criteria for Chronic Antibody-Mediated Rejection in Liver Allografts. *American journal of transplantation : official journal of the American Society of Transplantation and the American Society of Transplant Surgeons* **16**, 603-614, doi:10.1111/ajt.13476 (2016).

57 Iacob, S. *et al.* Donor-Specific Anti-HLA Antibodies and Endothelial C4d Deposition-Association With Chronic Liver Allograft Failure. *Transplantation* **99**, 1869-1875, doi:10.1097/tp.0000000000000613 (2015).

58 O'Leary, J. G. *et al.* Donor-specific alloantibodies are associated with fibrosis progression after liver transplantation in hepatitis C virus-infected patients. *Liver transplantation : official publication of the American Association for the Study of Liver Diseases and the International Liver Transplantation Society* **20**, 655-663, doi:10.1002/lt.23854 (2014).

59 Song, G. W. *et al.* ABO-Incompatible Adult Living Donor Liver Transplantation Under the Desensitization Protocol With Rituximab. *American journal of transplantation : official journal of the American Society of Transplantation and the American Society of Transplant Surgeons*, doi:10.1111/ajt.13444 (2015).

60 L, D. *Blood Groups and Red Cell Antigens [Internet]. Bethesda (MD): National Center for Biotechnology Information (US); 2005. Available from: https://www.ncbi.nlm.nih.gov/books/NBK2261/*.

61 Morath, C., Zeier, M., Dohler, B., Opelz, G. & Susal, C. ABO-Incompatible Kidney Transplantation. *Frontiers in immunology* **8**, 234, doi:10.3389/fimmu.2017.00234 (2017).

62 Song, G. W. *et al.* Biliary stricture is the only concern in ABO-incompatible adult living donor liver transplantation in the rituximab era. *Journal of hepatology* **61**, 575-582, doi:10.1016/j.jhep.2014.04.039 (2014).

63 Rummler, S. *et al.* ABO-Incompatible Living Donor Liver Transplantation in Focus of Antibody Rebound. *Transfusion medicine and hemotherapy : offizielles Organ der Deutschen Gesellschaft fur Transfusionsmedizin und Immunhamatologie* **44**, 46-51, doi:10.1159/000450792 (2017).

64 Moini, M., Schilsky, M. L. & Tichy, E. M. Review on immunosuppression in liver transplantation. *World journal of hepatology* **7**, 1355-1368, doi:10.4254/wjh.v7.i10.1355 (2015).

65 Mukherjee, S. & Mukherjee, U. A comprehensive review of immunosuppression used for liver transplantation. *Journal of transplantation* **2009**, 701464, doi:10.1155/2009/701464 (2009).

66 Clavien, P. A., Trotter, J.F. *Medical Care of The Liver Transplant Patient 4. ed*. (Wiley-Blackwell, 2013).

67 O'Grady, J. G., Hardy, P., Burroughs, A. K. & Elbourne, D. Randomized controlled trial of tacrolimus versus microemulsified cyclosporin (TMC) in liver transplantation: poststudy surveillance to 3 years. *American journal of transplantation : official journal of the American Society of Transplantation and the American Society of Transplant Surgeons* **7**, 137-141, doi:10.1111/j.1600-6143.2006.01576.x (2007).

68 Villarroel, M. C., Hidalgo, M. & Jimeno, A. Mycophenolate mofetil: An update. *Drugs of today (Barcelona, Spain : 1998)* **45**, 521-532, doi:10.1358/dot.2009.45.7.1384878 (2009).

69 Fung, J., Kelly, D., Kadry, Z., Patel-Tom, K. & Eghtesad, B. Immunosuppression in liver transplantation: beyond calcineurin inhibitors. *Liver transplantation : official publication of the American Association for the Study of Liver Diseases and the International Liver Transplantation Society* **11**, 267-280, doi:10.1002/lt.20373 (2005).

70 Segev, D. L. *et al.* Steroid avoidance in liver transplantation: meta-analysis and meta-regression of randomized trials. *Liver transplantation : official publication of the American Association for the Study of Liver Diseases and the International Liver Transplantation Society* **14**, 512-525, doi:10.1002/lt.21396 (2008).

71 Trouillot, T. E., Shrestha, R., Kam, I., Wachs, M. & Everson, G. T. Successful withdrawal of prednisone after adult liver transplantation for autoimmune hepatitis. *Liver transplantation and surgery : official publication of the American Association for the Study of Liver Diseases and the International Liver Transplantation Society* **5**, 375-380, doi:10.1002/lt.500050514 (1999).

72 Guttmann, R. D., Caudrelier, P., Alberici, G. & Touraine, J. L. Pharmacokinetics, foreign protein immune response, cytokine release, and lymphocyte subsets in patients receiving thymoglobuline and immunosuppression. *Transplantation proceedings* **29**, 24s-26s (1997).

73 Thomson, A. W., Turnquist, H. R. & Raimondi, G. Immunoregulatory functions of mTOR inhibition. *Nature reviews. Immunology* **9**, 324-337, doi:10.1038/nri2546 (2009).

74 Kaplan, B., Qazi, Y. & Wellen, J. R. Strategies for the management of adverse events associated with mTOR inhibitors. *Transplantation reviews (Orlando, Fla.)* **28**, 126-133, doi:10.1016/j.trre.2014.03.002 (2014).

75 Hagness, M. Liver transplantation in treatment of colorectal liver metastases. *Hepatic oncology* **2**, 181-190, doi:10.2217/hep.15.3 (2015).

76 Trotter, J. F. Sirolimus in liver transplantation. *Transplantation proceedings* **35**, 193s-200s (2003).

77 Staudacher, V. [Transplantation of an organ with vascular anastomoses]. *La Riforma medica* **66**, 1060 (1952).

78 Busuttil, R. W. *et al.* The first report of orthotopic liver transplantation in the Western world. *American journal of transplantation : official journal of the American Society of Transplantation and the American Society of Transplant Surgeons* **12**, 1385-1387, doi:10.1111/j.1600-6143.2012.04026.x (2012).

79 Starlz, T. E., Kaupp, H. A., Jr., Brock, D. R., Lazarus, R. E. & Johnson, R. V. Reconstructive problems in canine liver homotransplantation with special reference to the postoperative role of hepatic venous flow. *Surgery, gynecology & obstetrics* **111**, 733-743 (1960).

80 Starzl, T. E. *et al.* HOMOTRANSPLANTATION OF THE LIVER IN HUMANS. *Surgery, gynecology & obstetrics* **117**, 659-676 (1963).

81 Moore, F. D. *et al.* IMMUNOSUPPRESSION AND VASCULAR INSUFFICIENCY IN LIVER TRANSPLANTATION. *Annals of the New York Academy of Sciences* **120**, 729-738 (1964).

82 Kuss R, B., P. *An Illustrated History of Organ Transplantation: The Great Adventure of the Century*. (Sandoz, 1992).

83 **C, D.** *Contemporary Liver Transplantation*. (Springer International Publishing, 2017).

84 Starzl, T. E. *et al.* Orthotopic homotransplantation of the human liver. *Annals of surgery* **168**, 392-415 (1968).

85 Calne, R. Y. & Williams, R. Liver transplantation in man. I. Observations on technique and organization in five cases. *British medical journal* **4**, 535-540 (1968).

86 Starlz TE, P. C. *Experience in Hepatic Transplantation*. (WB Saunders, 1969).

87 Starzl, T. E., Klintmalm, G. B., Porter, K. A., Iwatsuki, S. & Schroter, G. P. Liver transplantation with use of cyclosporin a and prednisone. *The New England journal of medicine* **305**, 266-269, doi:10.1056/nejm198107303050507 (1981).

88 Todo, S. *et al.* Liver, kidney, and thoracic organ transplantation under FK 506. *Annals of surgery* **212**, 295-305; discussion 306-297 (1990).

89 A comparison of tacrolimus (FK 506) and cyclosporine for immunosuppression in liver transplantation. *The New England journal of medicine* **331**, 1110-1115, doi:10.1056/nejm199410273311702 (1994).

90 Randomised trial comparing tacrolimus (FK506) and cyclosporin in prevention of liver allograft rejection. European FK506 Multicentre Liver Study Group. *Lancet* **344**, 423-428 (1994).

91 Pappas, G. *et al.* Hemodynamic changes in clinical orthotopic liver transplantation. *Surgical forum* **22**, 335-336 (1971).

92 Shaw, B. W., Jr. *et al.* Venous bypass in clinical liver transplantation. *Annals of surgery* **200**, 524-534 (1984).

93 Chari, R. S. *et al.* Venovenous bypass in adult orthotopic liver transplantation: routine or selective use? *Journal of the American College of Surgeons* **186**, 683-690 (1998).

94 Wall, W. J. *et al.* Liver transplantation without venous bypass. *Transplantation* **43**, 56-61 (1987).

95 Stock, P. G. *et al.* Rapid infusion technique as a safe alternative to veno-venous bypass in orthotopic liver transplant (TX). *Transplantation proceedings* **21**, 2322-2325 (1989).

96 Levi, D. M. *et al.* Liver transplantation with preservation of the inferior vena cava: lessons learned through 2,000 cases. *Journal of the American College of Surgeons* **214**, 691-698; discussion 698-699, doi:10.1016/j.jamcollsurg.2011.12.039 (2012).

97 Tzakis, A., Todo, S. & Starzl, T. E. Orthotopic liver transplantation with preservation of the inferior vena cava. *Annals of surgery* **210**, 649-652 (1989).

98 Pratschke, S. *et al.* Temporary Intraoperative Porto-Caval Shunts in Piggy-Back Liver Transplantation Reduce Intraoperative Blood Loss and Improve Postoperative Transaminases and Renal Function: A Meta-Analysis. *World journal of surgery* **40**, 2988-2998, doi:10.1007/s00268-016-3656-1 (2016).

99 Figueras, J. *et al.* Temporary portocaval shunt during liver transplantation with vena cava preservation. Results of a prospective randomized study. *Liver transplantation : official publication of the American Association for the Study of Liver Diseases and the International Liver Transplantation Society* **7**, 904-911, doi:10.1053/jlts.2001.27870 (2001).

100 Chen, X. B. & Xu, M. Q. Primary graft dysfunction after liver transplantation. *Hepatobiliary & pancreatic diseases international : HBPD INT* **13**, 125-137 (2014).

101 Bekker, J., Ploem, S. & de Jong, K. P. Early hepatic artery thrombosis after liver transplantation: a systematic review of the incidence, outcome and risk factors. *American journal of transplantation : official journal of the American Society of Transplantation and the American Society of Transplant Surgeons* **9**, 746-757, doi:10.1111/j.1600-6143.2008.02541.x (2009).

102 Vaidya, S., Dighe, M., Kolokythas, O. & Dubinsky, T. Liver transplantation: vascular complications. *Ultrasound quarterly* **23**, 239-253, doi:10.1097/ruq.0b013e31815d6e1d (2007).

103 Settmacher, U. *et al.* Venous complications after orthotopic liver transplantation. *Clinical transplantation* **14**, 235-241 (2000).

104 Welling, T. H. *et al.* Biliary complications following liver transplantation in the model for end-stage liver disease era: effect of donor, recipient, and technical factors. *Liver transplantation : official publication of the American Association for the Study of Liver Diseases and the International Liver Transplantation Society* **14**, 73-80, doi:10.1002/lt.21354 (2008).

105 Qian, Y. B., Liu, C. L., Lo, C. M. & Fan, S. T. Risk factors for biliary complications after liver transplantation. *Archives of surgery (Chicago, Ill. : 1960)* **139**, 1101-1105, doi:10.1001/archsurg.139.10.1101 (2004).

106 Rerknimitr, R. *et al.* Biliary tract complications after orthotopic liver transplantation with choledochocholedochostomy anastomosis: endoscopic findings and results of therapy. *Gastrointestinal endoscopy* **55**, 224-231, doi:10.1067/mge.2002.120813 (2002).

107 Neumann, U. *et al.* A Randomized Multicenter Study Comparing a Tacrolimus-Based Protocol with and without Steroids in HCV-Positive Liver Allograft Recipients. *Journal of transplantation* **2012**, 894215, doi:10.1155/2012/894215 (2012).

108 Fosby, B., Karlsen, T. H. & Melum, E. Recurrence and rejection in liver transplantation for primary sclerosing cholangitis. *World journal of gastroenterology : WJG* **18**, 1-15, doi:10.3748/wjg.v18.i1.1 (2012).

109 Klintmalm, G. B. *et al.* A randomized, multicenter study comparing steroid-free immunosuppression and standard immunosuppression for liver transplant recipients with chronic hepatitis C. *Liver transplantation : official publication of the American Association for the Study of Liver Diseases and the International Liver Transplantation Society* **17**, 1394-1403, doi:10.1002/lt.22417 (2011).

110 Wiesner, R. H. *et al.* Acute hepatic allograft rejection: incidence, risk factors, and impact on outcome. *Hepatology (Baltimore, Md.)* **28**, 638-645, doi:10.1002/hep.510280306 (1998).

111 Charlton, M. R. How important is acute cellular rejection? *Liver transplantation : official publication of the American Association for the Study of Liver Diseases and the International Liver Transplantation Society* **19 Suppl 2**, S9-s13, doi:10.1002/lt.23743 (2013).

112 Marchesini, G., Marzocchi, R., Agostini, F. & Bugianesi, E. Nonalcoholic fatty liver disease and the metabolic syndrome. *Current opinion in lipidology* **16**, 421-427 (2005).

113 Cholankeril, G. & Ahmed, A. Alcoholic Liver Disease Replaces Hepatitis C Virus Infection as the Leading Indication for Liver Transplantation in the United States. *Clinical gastroenterology and hepatology : the official clinical practice journal of the American Gastroenterological Association* **16**, 1356-1358, doi:10.1016/j.cgh.2017.11.045 (2018).

114 Adam, R. *et al.* Evolution of indications and results of liver transplantation in Europe. A report from the European Liver Transplant Registry (ELTR). *Journal of hepatology* **57**, 675-688, doi:10.1016/j.jhep.2012.04.015 (2012).

115 Belli, L. S. *et al.* Impact of DAAs on liver transplantation: Major effects on the evolution of indications and results. An ELITA study based on the ELTR registry. *Journal of hepatology* **69**, 810-817, doi:10.1016/j.jhep.2018.06.010 (2018).

116 Agopian, V. G. *et al.* Liver transplantation for nonalcoholic steatohepatitis: the new epidemic. *Annals of surgery* **256**, 624-633, doi:10.1097/SLA.0b013e31826b4b7e (2012).

117 Mak, L. Y. *et al.* Global Epidemiology, Prevention, and Management of Hepatocellular Carcinoma. *American Society of Clinical Oncology educational book. American Society of Clinical Oncology. Annual Meeting*, 262-279, doi:10.1200/edbk_200939 (2018).

118 Chen, C. L., Kabiling, C. S. & Concejero, A. M. Why does living donor liver transplantation flourish in Asia? *Nature reviews. Gastroenterology & hepatology* **10**, 746-751, doi:10.1038/nrgastro.2013.194 (2013).

119 NLTR, M. E. The Nordic Liver Transplant Registry (NLTR) - Annual report 2017. (2018).

120 O'Leary, J. G., Lepe, R. & Davis, G. L. Indications for liver transplantation. *Gastroenterology* **134**, 1764-1776, doi:10.1053/j.gastro.2008.02.028 (2008).

121 Pievsky, D., Rustgi, N. & Pyrsopoulos, N. T. Classification and Epidemiologic Aspects of Acute Liver Failure. *Clinics in liver disease* **22**, 229-241, doi:10.1016/j.cld.2018.01.001 (2018).

122 Fontana, R. J. *et al.* Two-year outcomes in initial survivors with acute liver failure: results from a prospective, multicentre study. *Liver international : official journal of the International Association for the Study of the Liver* **35**, 370-380, doi:10.1111/liv.12632 (2015).

123 Larson, A. M. Diagnosis and management of acute liver failure. *Current opinion in gastroenterology* **26**, 214-221, doi:10.1097/MOG.0b013e32833847c5 (2010).

124 Durand, F. How to improve long-term outcome after liver transplantation? *Liver international : official journal of the International Association for the Study of the Liver* **38 Suppl 1**, 134-138, doi:10.1111/liv.13651 (2018).

125 Gautam, M., Cheruvattath, R. & Balan, V. Recurrence of autoimmune liver disease after liver transplantation: a systematic review. *Liver transplantation : official publication of the American Association for the Study of Liver Diseases and the International Liver Transplantation Society* **12**, 1813-1824, doi:10.1002/lt.20910 (2006).

126 Folseraas, T. & Boberg, K. M. Cancer Risk and Surveillance in Primary Sclerosing Cholangitis. *Clinics in liver disease* **20**, 79-98, doi:10.1016/j.cld.2015.08.014 (2016).

127 Pfitzmann, R. *et al.* Long-term survival and predictors of relapse after orthotopic liver transplantation for alcoholic liver disease. *Liver transplantation : official publication of the American Association for the Study of Liver Diseases and the International Liver Transplantation Society* **13**, 197-205, doi:10.1002/lt.20934 (2007).

128 Mathurin, P. *et al.* Survival and prognostic factors in patients with severe alcoholic hepatitis treated with prednisolone. *Gastroenterology* **110**, 1847-1853 (1996).

129 Louvet, A. *et al.* Corticosteroids Reduce Risk of Death Within 28 Days for Patients With Severe Alcoholic Hepatitis, Compared With Pentoxifylline or Placebo-a Meta-analysis of

Individual Data From Controlled Trials. *Gastroenterology* **155**, 458-468.e458, doi:10.1053/j.gastro.2018.05.011 (2018).

130 Lim, J., Curry, M. P. & Sundaram, V. Risk factors and outcomes associated with alcohol relapse after liver transplantation. *World journal of hepatology* **9**, 771-780, doi:10.4254/wjh.v9.i17.771 (2017).

131 Lee, B. P. *et al.* Outcomes of Early Liver Transplantation for Patients With Severe Alcoholic Hepatitis. *Gastroenterology* **155**, 422-430.e421, doi:10.1053/j.gastro.2018.04.009 (2018).

132 Nair, S. *et al.* Postoperative morbidity, mortality, costs, and long-term survival in severely obese patients undergoing orthotopic liver transplantation. *The American journal of gastroenterology* **96**, 842-845, doi:10.1111/j.1572-0241.2001.03629.x (2001).

133 Leonard, J., Heimbach, J. K., Malinchoc, M., Watt, K. & Charlton, M. The impact of obesity on long-term outcomes in liver transplant recipients-results of the NIDDK liver transplant database. *American journal of transplantation : official journal of the American Society of Transplantation and the American Society of Transplant Surgeons* **8**, 667-672, doi:10.1111/j.1600-6143.2007.02100.x (2008).

134 Dare, A. J. *et al.* Additive effect of pretransplant obesity, diabetes, and cardiovascular risk factors on outcomes after liver transplantation. *Liver transplantation : official publication of the American Association for the Study of Liver Diseases and the International Liver Transplantation Society* **20**, 281-290, doi:10.1002/lt.23818 (2014).

135 Hakeem, A. R. *et al.* Increased morbidity in overweight and obese liver transplant recipients: a single-center experience of 1325 patients from the United Kingdom. *Liver transplantation : official publication of the American Association for the Study of Liver Diseases and the International Liver Transplantation Society* **19**, 551-562, doi:10.1002/lt.23618 (2013).

136 Bruix, J. & Sherman, M. Management of hepatocellular carcinoma. *Hepatology (Baltimore, Md.)* **42**, 1208-1236, doi:10.1002/hep.20933 (2005).

137 Bruix, J., Reig, M. & Sherman, M. Evidence-Based Diagnosis, Staging, and Treatment of Patients With Hepatocellular Carcinoma. *Gastroenterology* **150**, 835-853, doi:10.1053/j.gastro.2015.12.041 (2016).

138 Shiina, S. *et al.* Percutaneous Ablation for Hepatocellular Carcinoma: Comparison of Various Ablation Techniques and Surgery. *Canadian journal of gastroenterology & hepatology* **2018**, 4756147, doi:10.1155/2018/4756147 (2018).

139 Mazzaferro, V. *et al.* Liver transplantation for the treatment of small hepatocellular carcinomas in patients with cirrhosis. *The New England journal of medicine* **334**, 693-699, doi:10.1056/nejm199603143341104 (1996).

140 Pavel, M. C. & Fuster, J. Expansion of the hepatocellular carcinoma Milan criteria in liver transplantation: Future directions. *World journal of gastroenterology : WJG* **24**, 3626-3636, doi:10.3748/wjg.v24.i32.3626 (2018).

141 Costentin, C. E., Bababekov, Y. J., Zhu, A. X. & Yeh, H. Is it time to reconsider the Milan Criteria for selecting patients with hepatocellular carcinoma for deceased-donor liver transplantation? *Hepatology (Baltimore, Md.)*, doi:10.1002/hep.30278 (2018).

142 Goldaracena, N., Gorgen, A. & Sapisochin, G. Current status of liver transplantation for cholangiocarcinoma. *Liver transplantation : official publication of the American Association for the Study of Liver Diseases and the International Liver Transplantation Society* **24**, 294-303, doi:10.1002/lt.24955 (2018).

143 Iwatsuki, S. *et al.* Treatment of hilar cholangiocarcinoma (Klatskin tumors) with hepatic resection or transplantation. *Journal of the American College of Surgeons* **187**, 358-364 (1998).

144 Jonas, S. *et al.* Orthotopic liver transplantation after extended bile duct resection as treatment of hilar cholangiocarcinoma. First long-terms results. *Transplant international : official journal of the European Society for Organ Transplantation* **11 Suppl 1**, S206-208 (1998).

145 Heimbach, J. K. *et al.* Liver transplantation for unresectable perihilar cholangiocarcinoma. *Seminars in liver disease* **24**, 201-207, doi:10.1055/s-2004-828896 (2004).

146 Zamora-Valdes, D. & Heimbach, J. K. Liver Transplant for Cholangiocarcinoma. *Gastroenterology clinics of North America* **47**, 267-280, doi:10.1016/j.gtc.2018.01.002 (2018).

147 Modlin, I. M. *et al.* Gastroenteropancreatic neuroendocrine tumours. *The Lancet. Oncology* **9**, 61-72, doi:10.1016/s1470-2045(07)70410-2 (2008).

148 Fan, S. T. *et al.* Liver transplantation for neuroendocrine tumour liver metastases. *HPB : the official journal of the International Hepato Pancreato Biliary Association* **17**, 23-28, doi:10.1111/hpb.12308 (2015).

149 Mazzaferro, V. *et al.* The Long-Term Benefit of Liver Transplantation for Hepatic Metastases From Neuroendocrine Tumors. *American journal of transplantation : official journal of the American Society of Transplantation and the American Society of Transplant Surgeons* **16**, 2892-2902, doi:10.1111/ajt.13831 (2016).

150 Le Treut, Y. P. *et al.* Predictors of long-term survival after liver transplantation for metastatic endocrine tumors: an 85-case French multicentric report. *American journal of transplantation : official journal of the American Society of Transplantation and the American Society of Transplant Surgeons* **8**, 1205-1213, doi:10.1111/j.1600-6143.2008.02233.x (2008).

151 Society, A. C. (USA, 2011).

152 Kanas, G. P. *et al.* Survival after liver resection in metastatic colorectal cancer: review and meta-analysis of prognostic factors. *Clinical epidemiology* **4**, 283-301, doi:10.2147/clep.S34285 (2012).

153 Line, P. D., Hagness, M. & Dueland, S. The Potential Role of Liver Transplantation as a Treatment Option in Colorectal Liver Metastases. *Canadian journal of gastroenterology & hepatology* **2018**, 8547940, doi:10.1155/2018/8547940 (2018).

154 Moris, D. *et al.* Liver transplantation for unresectable colorectal liver metastases: A systematic review. *Journal of surgical oncology* **116**, 288-297, doi:10.1002/jso.24671 (2017).

155 Muhlbacher, F. *et al.* Is orthotopic liver transplantation a feasible treatment for secondary cancer of the liver? *Transplantation proceedings* **23**, 1567-1568 (1991).

156 Dueland, S. *et al.* Is Liver Transplantation an Option in Colorectal Cancer Patients with Nonresectable Liver Metastases and Progression on All Lines of Standard Chemotherapy? *Annals of surgical oncology* **22**, 2195-2200, doi:10.1245/s10434-014-4137-0 (2015).

157 Toso, C. *et al.* Liver transplantation for colorectal liver metastasis: Survival without recurrence can be achieved. *Liver transplantation : official publication of the American Association for the Study of Liver Diseases and the International Liver Transplantation Society* **23**, 1073-1076, doi:10.1002/lt.24791 (2017).

158 Kim, W. R. *et al.* OPTN/SRTR 2016 Annual Data Report: Liver. *American journal of transplantation : official journal of the American Society of Transplantation and the American Society of Transplant Surgeons* **18 Suppl 1**, 172-253, doi:10.1111/ajt.14559 (2018).

159 Adam, R. *et al.* 2018 annual report of the European Liver Transplant Registry (ELTR) - 50-year evolution of liver transplantation. *Transplant international : official journal of the European Society for Organ Transplantation*, doi:10.1111/tri.13358 (2018).

160 Eghtesad, B., Hashimoto, K. and Fung, J. in *Schiff's Diseases of the Liver12th ed* (ed W. C. Maddrey and K. R. Reddy E. R. Schiff) (Wiley-Blackwell, 2017).

161 Kim, W. R. *et al.* Liver. *American journal of transplantation : official journal of the American Society of Transplantation and the American Society of Transplant Surgeons* **16 Suppl 2**, 69-98, doi:10.1111/ajt.13668 (2016).

162 Nemes, B. *et al.* Extended criteria donors in liver transplantation Part I: reviewing the impact of determining factors. *Expert review of gastroenterology & hepatology* **10**, 827-839, doi:10.1586/17474124.2016.1149061 (2016).

163 Nemes, B. *et al.* Extended-criteria donors in liver transplantation Part II: reviewing the impact of extended-criteria donors on the complications and outcomes of liver transplantation.

*Expert review of gastroenterology & hepatology* **10**, 841-859, doi:10.1586/17474124.2016.1149062 (2016).

164 Burroughs, A. K. *et al.* 3-month and 12-month mortality after first liver transplant in adults in Europe: predictive models for outcome. *Lancet* **367**, 225-232, doi:10.1016/s0140-6736(06)68033-1 (2006).

165 Rubin, A., Aguilera, V. & Berenguer, M. Liver transplantation and hepatitis C. *Clinics and research in hepatology and gastroenterology* **35**, 805-812, doi:10.1016/j.clinre.2011.04.009 (2011).

166 Lake, J. R. *et al.* Differential effects of donor age in liver transplant recipients infected with hepatitis B, hepatitis C and without viral hepatitis. *American journal of transplantation : official journal of the American Society of Transplantation and the American Society of Transplant Surgeons* **5**, 549-557, doi:10.1111/j.1600-6143.2005.00741.x (2005).

167 Berenguer, M. Risk of extended criteria donors in hepatitis C virus-positive recipients. *Liver transplantation : official publication of the American Association for the Study of Liver Diseases and the International Liver Transplantation Society* **14 Suppl 2**, S45-50, doi:10.1002/lt.21617 (2008).

168 Tao, R. *et al.* Hepatitis C recurrence is not adversely affected by the use of donation after cardiac death liver allografts. *Liver transplantation : official publication of the American Association for the Study of Liver Diseases and the International Liver Transplantation Society* **16**, 1288-1295, doi:10.1002/lt.22168 (2010).

169 Capri, M. *et al.* Identification of miR-31-5p, miR-141-3p, miR-200c-3p, and GLT1 as human liver aging markers sensitive to donor-recipient age-mismatch in transplants. *Aging cell* **16**, 262-272, doi:10.1111/acel.12549 (2017).

170 Zeeh, J. & Platt, D. The aging liver: structural and functional changes and their consequences for drug treatment in old age. *Gerontology* **48**, 121-127, doi:10.1159/000052829 (2002).

171 Schmucker, D. L. Age-related changes in liver structure and function: Implications for disease ? *Experimental gerontology* **40**, 650-659, doi:10.1016/j.exger.2005.06.009 (2005).

172 Wakabayashi, H., Nishiyama, Y., Ushiyama, T., Maeba, T. & Maeta, H. Evaluation of the effect of age on functioning hepatocyte mass and liver blood flow using liver scintigraphy in preoperative estimations for surgical patients: comparison with CT volumetry. *The Journal of surgical research* **106**, 246-253 (2002).

173 Wynne, H. A. *et al.* The effect of age upon liver volume and apparent liver blood flow in healthy man. *Hepatology (Baltimore, Md.)* **9**, 297-301 (1989).

174 Dasari, B. V. M., Schlegel, A., Mergental, H. & Perera, M. The use of old donors in liver transplantation. *Best practice & research. Clinical gastroenterology* **31**, 211-217, doi:10.1016/j.bpg.2017.03.002 (2017).

175 Stewart, Z. A. *et al.* Increased risk of graft loss from hepatic artery thrombosis after liver transplantation with older donors. *Liver transplantation : official publication of the American Association for the Study of Liver Diseases and the International Liver Transplantation Society* **15**, 1688-1695, doi:10.1002/lt.21946 (2009).

176 Grazi, G. L. *et al.* A revised consideration on the use of very aged donors for liver transplantation. *American journal of transplantation : official journal of the American Society of Transplantation and the American Society of Transplant Surgeons* **1**, 61-68 (2001).

177 Serrano, M. T. *et al.* Outcome of liver transplantation using donors older than 60 years of age. *Clinical transplantation* **24**, 543-549, doi:10.1111/j.1399-0012.2009.01135.x (2010).

178 Ghinolfi, D. *et al.* Risk analysis of ischemic-type biliary lesions after liver transplant using octogenarian donors. *Liver transplantation : official publication of the American Association for the Study of Liver Diseases and the International Liver Transplantation Society* **22**, 588-598, doi:10.1002/lt.24401 (2016).

179 Chapman, W. C. *et al.* Donor Age-Based Analysis of Liver Transplantation Outcomes: Short- and Long-Term Outcomes Are Similar Regardless of Donor Age. *Journal of the American College of Surgeons* **221**, 59-69, doi:10.1016/j.jamcollsurg.2015.01.061 (2015).

180 Briceno, J., Marchal, T., Padillo, J., Solorzano, G. & Pera, C. Influence of marginal donors on liver preservation injury. *Transplantation* **74**, 522-526, doi:10.1097/01.Tp.0000026307.14572.8d (2002).

181 Jimenez Romero, C. *et al.* Use of octogenarian livers safely expands the donor pool. *Transplantation* **68**, 572-575 (1999).

182 Chela, H. *et al.* Elderly donor graft for liver transplantation: Never too late. *World journal of transplantation* **7**, 324-328, doi:10.5500/wjt.v7.i6.324 (2017).

183 Jimenez-Romero, C. *et al.* Using old liver grafts for liver transplantation: where are the limits? *World journal of gastroenterology : WJG* **20**, 10691-10702, doi:10.3748/wjg.v20.i31.10691 (2014).

184 Emre, S. *et al.* Safe use of hepatic allografts from donors older than 70 years. *Transplantation* **62**, 62-65 (1996).

185 Cescon, M. *et al.* Long-term survival of recipients of liver grafts from donors older than 80 years: is it achievable? *Liver transplantation : official publication of the American Association for the Study of Liver Diseases and the International Liver Transplantation Society* **9**, 1174-1180, doi:10.1053/jlts.2003.50234 (2003).

186 Nardo, B. *et al.* Liver transplantation from donors aged 80 years and over: pushing the limit. *American journal of transplantation : official journal of the American Society of Transplantation and the American Society of Transplant Surgeons* **4**, 1139-1147, doi:10.1111/j.1600-6143.2004.00472.x (2004).

187 Zapletal, C. *et al.* Does the liver ever age? Results of liver transplantation with donors above 80 years of age. *Transplantation proceedings* **37**, 1182-1185, doi:10.1016/j.transproceed.2004.11.056 (2005).

188 Segev, D. L. *et al.* Minimizing risk associated with elderly liver donors by matching to preferred recipients. *Hepatology (Baltimore, Md.)* **46**, 1907-1918, doi:10.1002/hep.21888 (2007).

189 Anderson, C. D. *et al.* Advanced donor age alone does not affect patient or graft survival after liver transplantation. *Journal of the American College of Surgeons* **207**, 847-852, doi:10.1016/j.jamcollsurg.2008.08.009 (2008).

190 Petridis, I. *et al.* Liver transplantation using donors older than 80 years: a single-center experience. *Transplantation proceedings* **40**, 1976-1978, doi:10.1016/j.transproceed.2008.05.063 (2008).

191 Singhal, A. *et al.* Orthotopic liver transplant using allografts from geriatric population in the United States: is there any age limit? *Experimental and clinical transplantation : official journal of the Middle East Society for Organ Transplantation* **8**, 196-201 (2010).

192 Rauchfuss, F., Voigt, R., Dittmar, Y., Heise, M. & Settmacher, U. Liver transplantation utilizing old donor organs: a German single-center experience. *Transplantation proceedings* **42**, 175-177, doi:10.1016/j.transproceed.2009.11.020 (2010).

193 Cascales Campos, P. A. *et al.* Improving the waiting list by using 75-year-old donors for recipients with hepatocellular carcinoma. *Transplantation proceedings* **42**, 627-630, doi:10.1016/j.transproceed.2010.02.015 (2010).

194 Faber, W. *et al.* Donor age does not influence 12-month outcome after orthotopic liver transplantation. *Transplantation proceedings* **43**, 3789-3795, doi:10.1016/j.transproceed.2011.10.048 (2011).

195 Cuende, N., Grande, L., Sanjuan, F. & Cuervas-Mons, V. Liver transplant with organs from elderly donors: Spanish experience with more than 300 liver donors over 70 years of age. *Transplantation* **73**, 1360 (2002).

196 Jimenez-Romero, C. *et al.* Long-term results using old liver grafts for transplantation: sexagenerian versus liver donors older than 70 years. *World journal of surgery* **37**, 2211-2221, doi:10.1007/s00268-013-2085-7 (2013).

197 Lai, Q. *et al.* Use of elderly donors for liver transplantation: has the limit been reached? *Journal of gastrointestinal and liver diseases : JGLD* **20**, 383-387 (2011).

198 Borchert, D. H., Glanemann, M., Mogl, M., Langrehr, J. & Neuhaus, P. Adult liver transplantation using liver grafts from donors over 70 years of age. *Transplantation proceedings* **37**, 1186-1187, doi:10.1016/j.transproceed.2004.12.261 (2005).

199 Darius, T. *et al.* Septuagenarian and octogenarian donors provide excellent liver grafts for transplantation. *Transplantation proceedings* **44**, 2861-2867, doi:10.1016/j.transproceed.2012.09.076 (2012).

200 Cescon, M. *et al.* Improving the outcome of liver transplantation with very old donors with updated selection and management criteria. *Liver transplantation : official publication of the American Association for the Study of Liver Diseases and the International Liver Transplantation Society* **14**, 672-679, doi:10.1002/lt.21433 (2008).

201 Ghinolfi, D. *et al.* Use of octogenarian donors for liver transplantation: a survival analysis. *American journal of transplantation : official journal of the American Society of Transplantation and the American Society of Transplant Surgeons* **14**, 2062-2071, doi:10.1111/ajt.12843 (2014).

202 Ghinolfi, D., De Simone, P., Tincani, G., Pezzati, D. & Filipponi, F. Beyond the Limit: Approaching Systematic Use of Nonagenarian Donors in Liver Transplantation. *Transplantation* **100**, e37-38, doi:10.1097/tp.0000000000001207 (2016).

203 Barbier, L. *et al.* Liver Transplantation With Older Donors: A Comparison With Younger Donors in a Context of Organ Shortage. *Transplantation* **100**, 2410-2415, doi:10.1097/tp.0000000000001401 (2016).

204 Paterno, F. *et al.* Use of Elderly Allografts in Liver Transplantation. *Transplantation* **100**, 153-158, doi:10.1097/tp.0000000000000806 (2016).

205 Bertuzzo, V. R. *et al.* Actual Risk of Using Very Aged Donors for Unselected Liver Transplant Candidates: A European Single-center Experience in the MELD Era. *Annals of surgery* **265**, 388-396, doi:10.1097/sla.0000000000001681 (2017).

206 Jimenez-Romero, C. *et al.* Octogenarian liver grafts: Is their use for transplant currently justified? *World journal of gastroenterology : WJG* **23**, 3099-3110, doi:10.3748/wjg.v23.i17.3099 (2017).

207 Halazun, K. J. *et al.* No country for old livers? Examining and optimizing the utilization of elderly liver grafts. *American journal of transplantation : official journal of the American Society of Transplantation and the American Society of Transplant Surgeons* **18**, 669-678, doi:10.1111/ajt.14518 (2018).

208 Haugen, C. E., Thomas, A. G., Garonzik-Wang, J., Massie, A. B. & Segev, D. L. Minimizing Risk Associated with Older Liver Donors by Matching to Preferred Recipients: A National Registry and Validation Study. *Transplantation*, doi:10.1097/tp.0000000000002190 (2018).

209 Sampedro, B., Cabezas, J., Fabrega, E., Casafont, F. & Pons-Romero, F. Liver transplantation with donors older than 75 years. *Transplantation proceedings* **43**, 679-682, doi:10.1016/j.transproceed.2011.01.084 (2011).

210 Zhao, Y., Lo, C. M., Liu, C. L. & Fan, S. T. Use of elderly donors (> 60 years) for liver transplantation. *Asian journal of surgery* **27**, 114-119, doi:10.1016/s1015-9584(09)60323-7 (2004).

211 Gonzalez-Sanchez, M. R. *et al.* Donors Older Than 75 Years Do Not Influence the Appearance of Biliary Complications After Liver Transplantation. *Transplantation proceedings* **50**, 640-643, doi:10.1016/j.transproceed.2017.11.048 (2018).

212 Rydberg, L., Breimer, M. E., Brynger, H. & Samuelsson, B. E. ABO-incompatible kidney transplantation (A2 to O). Qualitative and semiquantitative studies of the humoral immune response against different blood group A antigens. *Transplantation* **49**, 954-960 (1990).

213 Rydberg, L. *et al.* Characterisation of the anti-A antibody response following an ABO incompatible (A2 to O) kidney transplantation. *Molecular immunology* **29**, 547-560 (1992).

214 Ying, Y. *et al.* Serological characteristic and molecular basis of A2 subgroup in the Chinese population. *Transfusion and apheresis science : official journal of the World Apheresis Association : official journal of the European Society for Haemapheresis* **48**, 67-74, doi:10.1016/j.transci.2012.08.002 (2013).

215 Egawa, H. *et al.* Present status of ABO-incompatible living donor liver transplantation in Japan. *Hepatology (Baltimore, Md.)* **47**, 143-152, doi:10.1002/hep.21928 (2008).

216 Stayboldt, C., Rearden, A. & Lane, T. A. B antigen acquired by normal A1 red cells exposed to a patient's serum. *Transfusion* **27**, 41-44 (1987).

217 Reid, M. E. & Bird, G. W. Associations between human red cell blood group antigens and disease. *Transfusion medicine reviews* **4**, 47-55 (1990).

218 Nydegger, U. E. *et al.* Histo-blood group antigens as allo- and autoantigens. *Annals of the New York Academy of Sciences* **1050**, 40-51, doi:10.1196/annals.1313.006 (2005).

219 Starzl, T. E. *et al.* Fifteen years of clinical liver transplantation. *Gastroenterology* **77**, 375-388 (1979).

220 Gordon, R. D. *et al.* Liver transplantation across ABO blood groups. *Surgery* **100**, 342-348 (1986).

221 Raut, V. & Uemoto, S. Management of ABO-incompatible living-donor liver transplantation: past and present trends. *Surgery today* **41**, 317-322, doi:10.1007/s00595-010-4437-3 (2011).

222 Zarrinpar, A. & Busuttil, R. W. Liver transplantation: Evading antigens-ABO-incompatible liver transplantation. *Nature reviews. Gastroenterology & hepatology* **12**, 676-678, doi:10.1038/nrgastro.2015.193 (2015).

223 Rego, J. *et al.* Hyperacute rejection after ABO-incompatible orthotopic liver transplantation. *Transplantation proceedings* **19**, 4589-4590 (1987).

224 Gugenheim, J. *et al.* Rejection of ABO incompatible liver allografts in man. *Transplantation proceedings* **21**, 2223-2224 (1989).

225 Gugenheim, J., Samuel, D., Reynes, M. & Bismuth, H. Liver transplantation across ABO blood group barriers. *Lancet* **336**, 519-523 (1990).

226 Sanchez-Urdazpal, L. *et al.* Increased bile duct complications in liver transplantation across the ABO barrier. *Annals of surgery* **218**, 152-158 (1993).

227 Sanchez-Urdazpal, L. *et al.* Increased bile duct complications in ABO incompatible liver transplant recipients. *Transplantation proceedings* **23**, 1440-1441 (1991).

228 Tanabe, M. *et al.* Current progress in ABO-incompatible liver transplantation. *European journal of clinical investigation* **40**, 943-949, doi:10.1111/j.1365-2362.2010.02339.x (2010).

229 Egawa, H. *et al.* Impact of recipient age on outcome of ABO-incompatible living-donor liver transplantation. *Transplantation* **77**, 403-411, doi:10.1097/01.Tp.0000110295.88926.5c (2004).

230 Lee, S. D. *et al.* ABO-incompatible living donor liver transplantation without graft local infusion and splenectomy. *HPB : the official journal of the International Hepato Pancreato Biliary Association* **16**, 807-813, doi:10.1111/hpb.12215 (2014).

231 Egawa, H. *et al.* Impact of rituximab desensitization on blood-type-incompatible adult living donor liver transplantation: a Japanese multicenter study. *American journal of transplantation : official journal of the American Society of Transplantation and the American Society of Transplant Surgeons* **14**, 102-114, doi:10.1111/ajt.12520 (2014).

232 Kim, S. H., Lee, E. C., Shim, J. R. & Park, S. J. A simplified protocol using rituximab and immunoglobulin for ABO-incompatible low-titre living donor liver transplantation. *Liver international : official journal of the International Association for the Study of the Liver*, doi:10.1111/liv.13614 (2017).

233 Lee, E. C., Kim, S. H., Shim, J. R. & Park, S. J. A comparison of desensitization methods: Rituximab with/without plasmapheresis in ABO-incompatible living donor liver transplantation. *Hepatobiliary & pancreatic diseases international : HBPD INT*, doi:10.1016/j.hbpd.2018.02.005 (2018).

234 Kim, J. M. *et al.* ABO-incompatible Living Donor Liver Transplantation with Rituximab and Total Plasma Exchange Does Not Increase Hepatocellular Carcinoma Recurrence. *Transplantation*, doi:10.1097/tp.0000000000002154 (2018).

235 Szczeklik, W. *et al.* Complications in patients treated with plasmapheresis in the intensive care unit. *Anaesthesiology intensive therapy* **45**, 7-13, doi:10.5603/ait.2013.0002 (2013).

236 Genberg, H., Hansson, A., Wernerson, A., Wennberg, L. & Tyden, G. Pharmacodynamics of rituximab in kidney allotransplantation. *American journal of transplantation : official journal of the American Society of Transplantation and the American Society of Transplant Surgeons* **6**, 2418-2428, doi:10.1111/j.1600-6143.2006.01497.x (2006).

237 Kim, S. H. *et al.* Feasibility of ABO-incompatible adult living donor liver transplantation for acute-on-chronic liver failure. *Hepatobiliary & pancreatic diseases international : HBPD INT* **16**, 662-664, doi:10.1016/s1499-3872(17)60060-2 (2017).

238 Shen, T. *et al.* A modified protocol with rituximab and intravenous immunoglobulin in emergent ABO-incompatible liver transplantation for acute liver failure. *Hepatobiliary & pancreatic diseases international : HBPD INT* **13**, 395-401 (2014).

239 Wu, J. *et al.* Recipient outcomes after ABO-incompatible liver transplantation: a systematic review and meta-analysis. *PloS one* **6**, e16521, doi:10.1371/journal.pone.0016521 (2011).

240 Zhou, J. *et al.* ABO-incompatible liver transplantation for severe hepatitis B patients. *Transplant international : official journal of the European Society for Organ Transplantation* **28**, 793-799, doi:10.1111/tri.12531 (2015).

241 Mendes, M. *et al.* ABO-incompatible liver transplantation in acute liver failure: a single Portuguese center study. *Transplantation proceedings* **45**, 1110-1115, doi:10.1016/j.transproceed.2013.02.012 (2013).

242 Lee, E. C., Kim, S. H. & Park, S. J. Outcomes after liver transplantation in accordance with ABO compatibility: A systematic review and meta-analysis. *World journal of gastroenterology : WJG* **23**, 6516-6533, doi:10.3748/wjg.v23.i35.6516 (2017).

243 Skogsberg, U. *et al.* Successful ABO-incompatible liver transplantation using A2 donors. *Transplantation proceedings* **38**, 2667-2670, doi:10.1016/j.transproceed.2006.07.025 (2006).

244 Skogsberg, U. *et al.* Adult ABO-incompatible liver transplantation, using A and B donors. *Xenotransplantation* **13**, 154-159, doi:10.1111/j.1399-3089.2006.00286.x (2006).

245 Kluger, M. D. *et al.* Safety of blood group A2-to-O liver transplantation: an analysis of the United Network of Organ Sharing database. *Transplantation* **94**, 526-531, doi:10.1097/TP.0b013e31825c591e (2012).

246 Fishbein, T. M. *et al.* Safe transplantation of blood type A2 livers to blood type O recipients. *Transplantation* **67**, 1071-1073 (1999).

247 Merion, R. M. *et al.* The survival benefit of liver transplantation. *American journal of transplantation : official journal of the American Society of Transplantation and the American Society of Transplant Surgeons* **5**, 307-313, doi:10.1111/j.1600-6143.2004.00703.x (2005).

248 Cholongitas, E. *et al.* Systematic review: The model for end-stage liver disease--should it replace Child-Pugh's classification for assessing prognosis in cirrhosis? *Alimentary pharmacology & therapeutics* **22**, 1079-1089, doi:10.1111/j.1365-2036.2005.02691.x (2005).

249 Kamath, P. S. *et al.* A model to predict survival in patients with end-stage liver disease. *Hepatology (Baltimore, Md.)* **33**, 464-470, doi:10.1053/jhep.2001.22172 (2001).

250 Schaubel, D. E. *et al.* Survival benefit-based deceased-donor liver allocation. *American journal of transplantation : official journal of the American Society of Transplantation and the*

*American Society of Transplant Surgeons* **9**, 970-981, doi:10.1111/j.1600-6143.2009.02571.x (2009).

251 Schaubel, D. E., Sima, C. S., Goodrich, N. P., Feng, S. & Merion, R. M. The survival benefit of deceased donor liver transplantation as a function of candidate disease severity and donor quality. *American journal of transplantation : official journal of the American Society of Transplantation and the American Society of Transplant Surgeons* **8**, 419-425, doi:10.1111/j.1600-6143.2007.02086.x (2008).

252 Machicao, V. I. Model for End-Stage Liver Disease-Sodium Score: The Evolution in the Prioritization of Liver Transplantation. *Clinics in liver disease* **21**, 275-287, doi:10.1016/j.cld.2016.12.014 (2017).

253 Bernardi, M., Gitto, S. & Biselli, M. The MELD score in patients awaiting liver transplant: strengths and weaknesses. *Journal of hepatology* **54**, 1297-1306, doi:10.1016/j.jhep.2010.11.008 (2011).

254 Wang, Y. W. *et al.* Correlation and comparison of the model for end-stage liver disease, portal pressure, and serum sodium for outcome prediction in patients with liver cirrhosis. *Journal of clinical gastroenterology* **41**, 706-712, doi:10.1097/MCG.0b013e31802dabb3 (2007).

255 Arvaniti, V. *et al.* Infections in patients with cirrhosis increase mortality four-fold and should be used in determining prognosis. *Gastroenterology* **139**, 1246-1256, 1256.e1241-1245, doi:10.1053/j.gastro.2010.06.019 (2010).

256 Mutimer, D. J. *et al.* Impact of donor age and year of transplantation on graft and patient survival following liver transplantation for hepatitis C virus. *Transplantation* **81**, 7-14 (2006).

257 Berenguer, M. *et al.* Contribution of donor age to the recent decrease in patient survival among HCV-infected liver transplant recipients. *Hepatology (Baltimore, Md.)* **36**, 202-210, doi:10.1053/jhep.2002.33993 (2002).

258 Volk, M. L., Lok, A. S., Pelletier, S. J., Ubel, P. A. & Hayward, R. A. Impact of the model for end-stage liver disease allocation policy on the use of high-risk organs for liver transplantation. *Gastroenterology* **135**, 1568-1574 (2008).

259 Rauchfuss, F. *et al.* Waiting time, not donor-risk-index, is a major determinant for beneficial outcome after liver transplantation in high-MELD patients. *Annals of transplantation : quarterly of the Polish Transplantation Society* **18**, 243-247, doi:10.12659/aot.883924 (2013).

260 Halldorson, J. B., Bakthavatsalam, R., Fix, O., Reyes, J. D. & Perkins, J. D. D-MELD, a simple predictor of post liver transplant mortality for optimization of donor/recipient matching. *American journal of transplantation : official journal of the American Society of Transplantation and the American Society of Transplant Surgeons* **9**, 318-326, doi:10.1111/j.1600-6143.2008.02491.x (2009).

261 Briceno, J., Ciria, R. & de la Mata, M. Donor-recipient matching: myths and realities. *Journal of hepatology* **58**, 811-820, doi:10.1016/j.jhep.2012.10.020 (2013).

262 Rana, A. *et al.* Survival outcomes following liver transplantation (SOFT) score: a novel method to predict patient survival following liver transplantation. *American journal of transplantation : official journal of the American Society of Transplantation and the American Society of Transplant Surgeons* **8**, 2537-2546, doi:10.1111/j.1600-6143.2008.02400.x (2008).

263 Dutkowski, P. *et al.* Are there better guidelines for allocation in liver transplantation? A novel score targeting justice and utility in the model for end-stage liver disease era. *Annals of surgery* **254**, 745-753; discussion 753, doi:10.1097/SLA.0b013e3182365081 (2011).

264 Braat, A. E. *et al.* The Eurotransplant donor risk index in liver transplantation: ET-DRI. *American journal of transplantation : official journal of the American Society of Transplantation and the American Society of Transplant Surgeons* **12**, 2789-2796, doi:10.1111/j.1600-6143.2012.04195.x (2012).

265 Winter, A. *et al.* External validation of the Donor Risk Index and the Eurotransplant Donor Risk Index on the French liver transplantation registry. *Liver international : official journal of*

*the International Association for the Study of the Liver* **37**, 1229-1238, doi:10.1111/liv.13378 (2017).

266 Blok, J. J. *et al.* Combined effect of donor and recipient risk on outcome after liver transplantation: Research of the Eurotransplant database. *Liver transplantation : official publication of the American Association for the Study of Liver Diseases and the International Liver Transplantation Society* **21**, 1486-1493, doi:10.1002/lt.24308 (2015).

267 Washburn, K., Harper, A., Baker, T. & Edwards, E. Changes in liver acceptance patterns after implementation of Share 35. *Liver transplantation : official publication of the American Association for the Study of Liver Diseases and the International Liver Transplantation Society* **22**, 171-177, doi:10.1002/lt.24348 (2016).

268 Busuttil, R. W. & Tanaka, K. The utility of marginal donors in liver transplantation. *Liver transplantation : official publication of the American Association for the Study of Liver Diseases and the International Liver Transplantation Society* **9**, 651-663, doi:10.1053/jlts.2003.50105 (2003).

269 Moore, D. E. *et al.* Impact of donor, technical, and recipient risk factors on survival and quality of life after liver transplantation. *Archives of surgery (Chicago, Ill. : 1960)* **140**, 273-277, doi:10.1001/archsurg.140.3.273 (2005).

270 Kim, D. Y. *et al.* Routine use of livers from deceased donors older than 70: is it justified? *Transplant international : official journal of the European Society for Organ Transplantation* **18**, 73-77, doi:10.1111/j.1432-2277.2004.00017.x (2005).

271 Gastaca, M. *et al.* Donors older than 70 years in liver transplantation. *Transplantation proceedings* **37**, 3851-3854, doi:10.1016/j.transproceed.2005.10.040 (2005).

272 Chedid, M. F., Rosen, C. B., Nyberg, S. L. & Heimbach, J. K. Excellent long-term patient and graft survival are possible with appropriate use of livers from deceased septuagenarian and octogenarian donors. *HPB : the official journal of the International Hepato Pancreato Biliary Association* **16**, 852-858, doi:10.1111/hpb.12221 (2014).

273 Dasari, B. V. *et al.* Systematic review and meta-analysis of liver transplantation using grafts from deceased donors aged over 70 years. *Clinical transplantation* **31**, doi:10.1111/ctr.13139 (2017).

274 Ghinolfi, D. *et al.* Use of Elderly Donors in Liver Transplantation: A Paired-match Analysis at a Single Center. *Annals of surgery*, doi:10.1097/sla.0000000000002305 (2017).

275 Foley, D. P. *et al.* Biliary complications after liver transplantation from donation after cardiac death donors: an analysis of risk factors and long-term outcomes from a single center. *Annals of surgery* **253**, 817-825, doi:10.1097/SLA.0b013e3182104784 (2011).

276 Roos, F. J. M., Poley, J. W., Polak, W. G. & Metselaar, H. J. Biliary complications after liver transplantation; recent developments in etiology, diagnosis and endoscopic treatment. *Best practice & research. Clinical gastroenterology* **31**, 227-235, doi:10.1016/j.bpg.2017.04.002 (2017).

277 Nemes, B., Gaman, G. & Doros, A. Biliary complications after liver transplantation. *Expert review of gastroenterology & hepatology* **9**, 447-466, doi:10.1586/17474124.2015.967761 (2015).

278 Seehofer, D., Eurich, D., Veltzke-Schlieker, W. & Neuhaus, P. Biliary complications after liver transplantation: old problems and new challenges. *American journal of transplantation : official journal of the American Society of Transplantation and the American Society of Transplant Surgeons* **13**, 253-265, doi:10.1111/ajt.12034 (2013).

279 Heidenhain, C. *et al.* Incidence of and risk factors for ischemic-type biliary lesions following orthotopic liver transplantation. *Transplant international : official journal of the European Society for Organ Transplantation* **23**, 14-22, doi:10.1111/j.1432-2277.2009.00947.x (2010).

280 Cursio, R. & Gugenheim, J. Ischemia-Reperfusion Injury and Ischemic-Type Biliary Lesions following Liver Transplantation. *Journal of transplantation* **2012**, 164329, doi:10.1155/2012/164329 (2012).

281 Zhang, Y. C. *et al.* New diagnosis and therapy model for ischemic-type biliary lesions following liver transplantation--a retrospective cohort study. *PloS one* **9**, e105795, doi:10.1371/journal.pone.0105795 (2014).

282 Diaz Jaime, F. & Berenguer, M. Pushing the donor limits: Deceased donor liver transplantation using organs from octogenarian donors. *Liver transplantation : official publication of the American Association for the Study of Liver Diseases and the International Liver Transplantation Society* **23**, S22-s26, doi:10.1002/lt.24841 (2017).

283 Berstad, A. E., Brabrand, K. & Foss, A. Clinical utility of microbubble contrast-enhanced ultrasound in the diagnosis of hepatic artery occlusion after liver transplantation. *Transplant international : official journal of the European Society for Organ Transplantation* **22**, 954-960, doi:10.1111/j.1432-2277.2009.00898.x (2009).

284 Ploeg, R. J. *et al.* Risk factors for primary dysfunction after liver transplantation--a multivariate analysis. *Transplantation* **55**, 807-813 (1993).

285 Cameron, A. M. *et al.* Optimal utilization of donor grafts with extended criteria: a single-center experience in over 1000 liver transplants. *Annals of surgery* **243**, 748-753; discussion 753-745, doi:10.1097/01.sla.0000219669.84192.b3 (2006).

286 Guo, H. *et al.* [Histopathological study of 268 hepatic allograft biopsies]. *Zhonghua yi xue za zhi* **91**, 3401-3404 (2011).

287 Kim, D. Y. *et al.* Liver transplantation using elderly donors: a risk factor analysis. *Clinical transplantation* **25**, 270-276, doi:10.1111/j.1399-0012.2010.01222.x (2011).

288 Franco, C. C. *et al.* Results of liver transplants from donors aged 70 plus: analysis of Andalusian transplant register. *Transplantation proceedings* **45**, 3647-3649, doi:10.1016/j.transproceed.2013.10.024 (2013).

289 Alkofer, B. *et al.* Extended-donor criteria liver allografts. *Seminars in liver disease* **26**, 221-233, doi:10.1055/s-2006-947292 (2006).

290 Adam, R. *et al.* Normalised intrinsic mortality risk in liver transplantation: European Liver Transplant Registry study. *Lancet* **356**, 621-627 (2000).

291 Cassuto, J. R., Patel, S. A., Tsoulfas, G., Orloff, M. S. & Abt, P. L. The cumulative effects of cold ischemic time and older donor age on liver graft survival. *The Journal of surgical research* **148**, 38-44, doi:10.1016/j.jss.2008.03.018 (2008).

292 Uygun, B. E., Yarmush, M. L. & Uygun, K. Application of whole-organ tissue engineering in hepatology. *Nature reviews. Gastroenterology & hepatology* **9**, 738-744, doi:10.1038/nrgastro.2012.140 (2012).

293 Vagefi, P. A., Dodge, J. L., Yao, F. Y. & Roberts, J. P. Potential role of the donor in hepatocellular carcinoma recurrence after liver transplantation. *Liver transplantation : official publication of the American Association for the Study of Liver Diseases and the International Liver Transplantation Society* **21**, 187-194, doi:10.1002/lt.24042 (2015).

294 Gordon Burroughs, S. & Busuttil, R. W. Optimal utilization of extended hepatic grafts. *Surgery today* **39**, 746-751, doi:10.1007/s00595-008-4022-1 (2009).

295 Busquets, J. *et al.* The impact of donor age on liver transplantation: influence of donor age on early liver function and on subsequent patient and graft survival. *Transplantation* **71**, 1765-1771 (2001).

296 Fiel, M. I., Deniz, K., Elmali, F. & Schiano, T. D. Increasing hepatic arteriole wall thickness and decreased luminal diameter occur with increasing age in normal livers. *Journal of hepatology* **55**, 582-586, doi:10.1016/j.jhep.2010.12.018 (2011).

297 Detry, O. *et al.* Cadaveric liver transplantation for non-acetaminophen fulminant hepatic failure: a 20-year experience. *World journal of gastroenterology : WJG* **13**, 1427-1430 (2007).

298 Farges, O. *et al.* Long-term results of ABO-incompatible liver transplantation. *Transplantation proceedings* **27**, 1701-1702 (1995).

299 Busquets, J. *et al.* Liver transplantation across Rh blood group barriers increases the risk of biliary complications. *Journal of gastrointestinal surgery : official journal of the Society for Surgery of the Alimentary Tract* **11**, 458-463, doi:10.1007/s11605-007-0116-0 (2007).

300 McCaughan, J. A. *et al.* Preformed donor-specific HLA antibodies are associated with increased risk of early mortality after liver transplantation. *Clinical transplantation* **30**, 1538-1544, doi:10.1111/ctr.12851 (2016).

301 Abu-Elmagd, K. M. *et al.* Preformed and de novo donor specific antibodies in visceral transplantation: long-term outcome with special reference to the liver. *American journal of transplantation : official journal of the American Society of Transplantation and the American Society of Transplant Surgeons* **12**, 3047-3060, doi:10.1111/j.1600-6143.2012.04237.x (2012).

302 Salah, A. *et al.* Application of complement component 4d immunohistochemistry to ABO-compatible and ABO-incompatible liver transplantation. *Liver transplantation : official publication of the American Association for the Study of Liver Diseases and the International Liver Transplantation Society* **20**, 200-209, doi:10.1002/lt.23789 (2014).

303 Olausson, M. *et al.* Successful combined partial auxiliary liver and kidney transplantation in highly sensitized cross-match positive recipients. *American journal of transplantation : official journal of the American Society of Transplantation and the American Society of Transplant Surgeons* **7**, 130-136, doi:10.1111/j.1600-6143.2006.01592.x (2007).

304 Taner, T. *et al.* Decreased chronic cellular and antibody-mediated injury in the kidney following simultaneous liver-kidney transplantation. *Kidney international* **89**, 909-917, doi:10.1016/j.kint.2015.10.016 (2016).

305 Haga, H. *et al.* Periportal edema and necrosis as diagnostic histological features of early humoral rejection in ABO-incompatible liver transplantation. *Liver transplantation : official publication of the American Association for the Study of Liver Diseases and the International Liver Transplantation Society* **10**, 16-27, doi:10.1002/lt.20002 (2004).

306 Rydberg, L., Skogsberg, U. & Molne, J. ABO antigen expression in graft tissue: is titration against donor erythrocytes relevant? *Transplantation* **84**, S10-12, doi:10.1097/01.tp.0000296017.44394.e2 (2007).

307 Detry, O. Should ABO-incompatible deceased liver transplantation be reconsidered? *Transplant international : official journal of the European Society for Organ Transplantation* **28**, 788-789, doi:10.1111/tri.12573 (2015).

308 Watts, R. P., Thom, O. & Fraser, J. F. Inflammatory signalling associated with brain dead organ donation: from brain injury to brain stem death and posttransplant ischaemia reperfusion injury. *Journal of transplantation* **2013**, 521369, doi:10.1155/2013/521369 (2013).

309 The Japanese Liver Transplant Society. Liver Transplantation in Japan — Registry by the Japanese Liver Transplantation Society. *Japan Soc Transplant Ishoku (Japan J Transplant)* **47**, 416-428 (2012).

310 Bruggenwirth, I. M. A., Dolgin, N. H., Porte, R. J., Bozorgzadeh, A. & Martins, P. N. A. Donor Diabetes and Prolonged Cold Ischemia Time Synergistically Increase the Risk of Graft Failure After Liver Transplantation. *Transplantation direct* **3**, e173, doi:10.1097/txd.0000000000000692 (2017).

311 Gilbo, N. *et al.* Reducing Non-Anastomotic Biliary Strictures in Donation After Circulatory Death Liver Transplantation: Cold Ischemia Time Matters! *Annals of surgery* **266**, e118-e119, doi:10.1097/sla.0000000000001949 (2017).

312 Segev, D. L. *et al.* Effect modification in liver allografts with prolonged cold ischemic time. *American journal of transplantation : official journal of the American Society of Transplantation and the American Society of Transplant Surgeons* **8**, 658-666, doi:10.1111/j.1600-6143.2007.02108.x (2008).

313 Nordin, A. *et al.* Iatrogenic lesion at cholecystectomy resulting in liver transplantation. *Transplantation proceedings* **33**, 2499-2500 (2001).

314 Buell, J. F. *et al.* Devastating and fatal complications associated with combined vascular and bile duct injuries during cholecystectomy. *Archives of surgery (Chicago, Ill. : 1960)* **137**, 703-708; discussion 708-710 (2002).

315 Fernandez, J. A. *et al.* Laparoscopic iatrogeny of the hepatic hilum as an indication for liver transplantation. *Liver transplantation : official publication of the American Association for the Study of Liver Diseases and the International Liver Transplantation Society* **10**, 147-152, doi:10.1002/lt.20021 (2004).

316 Zaydfudim, V., Wright, J. K. & Pinson, C. W. Liver transplantation for iatrogenic porta hepatis transection. *The American surgeon* **75**, 313-316 (2009).

317 Ardiles, V. *et al.* Experience using liver transplantation for the treatment of severe bile duct injuries over 20 years in Argentina: results from a National Survey. *HPB : the official journal of the International Hepato Pancreato Biliary Association* **13**, 544-550, doi:10.1111/j.1477-2574.2011.00322.x (2011).

318 Wang, Z. *et al.* Therapeutic strategies of iatrogenic portal vein injury after cholecystectomy. *The Journal of surgical research* **185**, 934-939, doi:10.1016/j.jss.2013.06.032 (2013).

319 Parrilla, P. *et al.* Liver transplantation for bile duct injury after open and laparoscopic cholecystectomy. *The British journal of surgery* **101**, 63-68, doi:10.1002/bjs.9349 (2014).

320 Leale, I. *et al.* Role of Liver Transplantation in Bilio-Vascular Liver Injury After Cholecystectomy. *Transplantation proceedings* **48**, 370-376, doi:10.1016/j.transproceed.2015.12.035 (2016).

321 Huerta, S., Li, Z. & Livingston, E. H. Outcome of portal injuries following bariatric operations. *Obesity surgery* **16**, 105-109, doi:10.1381/096089206775222203 (2006).

322 Lauterio, A. *et al.* Liver transplantation in the treatment of severe iatrogenic liver injuries. *World journal of hepatology* **9**, 1022-1029, doi:10.4254/wjh.v9.i24.1022 (2017).

323 Di Benedetto, F. *et al.* Liver transplantation due to iatrogenic injuries: two case reports. *Transplantation proceedings* **42**, 1375-1377, doi:10.1016/j.transproceed.2010.03.077 (2010).

324 Tessier, D. J. *et al.* Previously unreported high-grade complications of adrenalectomy. *Surgical endoscopy* **23**, 97-102, doi:10.1007/s00464-008-9947-3 (2009).

325 Aseni, P. *et al.* Life-saving super-urgent liver transplantation with replacement of retrohepatic vena cava by dacron graft. *HPB surgery : a world journal of hepatic, pancreatic and biliary surgery* **2010**, doi:10.1155/2010/828326 (2010).

326 Lauterio, A. *et al.* Current status and perspectives in split liver transplantation. *World journal of gastroenterology : WJG* **21**, 11003-11015, doi:10.3748/wjg.v21.i39.11003 (2015).

327 Ravikumar, R. *et al.* Liver Transplantation After Ex Vivo Normothermic Machine Preservation: A Phase 1 (First-in-Man) Clinical Trial. *American journal of transplantation : official journal of the American Society of Transplantation and the American Society of Transplant Surgeons* **16**, 1779-1787, doi:10.1111/ajt.13708 (2016).

328 Pezzati, D. *et al.* Salvage of an Octogenarian Liver Graft Using Normothermic Perfusion: A Case Report. *Transplantation proceedings* **49**, 726-728, doi:10.1016/j.transproceed.2017.02.014 (2017).

329 Rayar, M. *et al.* A Preliminary Clinical Experience Using Hypothermic Oxygenated Machine Perfusion for Rapid Recovery of Octogenarian Liver Grafts. *Progress in transplantation (Aliso Viejo, Calif.)*, 1526924818817072, doi:10.1177/1526924818817072 (2018).

330 Ghinolfi, D. *et al.* Pilot, open, randomized, prospective trial for normothermic machine perfusion evaluation in liver transplantation from older donors. *Liver transplantation : official publication of the American Association for the Study of Liver Diseases and the International Liver Transplantation Society*, doi:10.1002/lt.25362 (2018).

331 Jia, J. J. *et al.* Machine perfusion for liver transplantation: A concise review of clinical trials. *Hepatobiliary & pancreatic diseases international : HBPD INT* **17**, 387-391, doi:10.1016/j.hbpd.2018.06.003 (2018).

332 Perera, T. *et al.* First human liver transplantation using a marginal allograft resuscitated by normothermic machine perfusion. *Liver transplantation : official publication of the American*

*Association for the Study of Liver Diseases and the International Liver Transplantation Society* **22**, 120-124, doi:10.1002/lt.24369 (2016).

333 Graham, J. A. & Guarrera, J. V. "Resuscitation" of marginal liver allografts for transplantation with machine perfusion technology. *Journal of hepatology* **61**, 418-431, doi:10.1016/j.jhep.2014.04.019 (2014).

334 Guarrera, J. V. Assist devices: machine preservation of extended criteria donors. *Liver transplantation : official publication of the American Association for the Study of Liver Diseases and the International Liver Transplantation Society* **18 Suppl 2**, S31-33, doi:10.1002/lt.23505 (2012).

335 Angelico, R. *et al.* Normothermic Machine Perfusion of Deceased Donor Liver Grafts Is Associated With Improved Postreperfusion Hemodynamics. *Transplantation direct* **2**, e97, doi:10.1097/txd.0000000000000611 (2016).

336 Faitot, F. *et al.* Impact of real-time metabolomics in liver transplantation: Graft evaluation and donor-recipient matching. *Journal of hepatology*, doi:10.1016/j.jhep.2017.11.022 (2017).

III

# Liver transplantation as a lifesaving procedure for posthepatectomy liver failure and iatrogenic liver injuries

**Trygve Thorsen, MD [a,b], Jon Magnus Solheim, MD [a,b], Knut Jørgen Labori MD, PhD [c], Pål-Dag Line, MD, PhD [a,b], and Einar Martin Aandahl, MD, PhD [a,d]**

[a] Section for Transplant Surgery, Oslo University Hospital Rikshospitalet, Oslo, Norway

[b] Institute for Clinical Medicine, University of Oslo, Oslo, Norway

[c] Department of Hepato-Pancreato-Biliary Surgery, Oslo University Hospital Rikshospitalet, Oslo, Norway

[d]Institute for Cancer Research, Oslo University Hospital, Oslo, Norway

**Corresponding author**: Trygve Thorsen, Oslo University Hospital Rikshospitalet, Sognsvannsveien 20, 0372 Oslo, Norway

Primary email: thorsen_t@yahoo.no

Secondary email: tthorsen@ous-hf.no

Phone: +47 93433693

**Manuscript category**: original article – retrospective study

**Keywords:** Liver Transplantation – Rescue – Iatrogenic injuries – Posthepatectomy liver failure

**ABSTRACT**

**Background**: Iatrogenic injuries to vital structures of the liver and posthepatectomy liver failure are associated with high mortality. The current donor situation in Norway allows liver transplantation of patients beyond conventional criteria.

**Methods**: From 1984 to 2017 a total of 1510 liver transplantations were performed. In this retrospective study, we report the results of 13 patients undergoing liver transplantation due to iatrogenic injuries to the liver vasculature or posthepatectomy liver failure.

**Results:** Twelve men and one woman with a median age of 55 years (range 22 - 69) were included. Seven patients underwent radical surgery for cancer prior to transplantation. The median follow-up time was 70.5 months (range 2.2 - 177). Three of the patients with malignant disease did not experience disease recurrence, whereas four patients had cancer recurrence and died 7, 24, 45 and 78 months after transplantation. Five of six patients with non-malignant disease fully recovered, but one patient died after 9 months due to infectious complications.

**Conclusions:** Liver transplantation for liver failure due to portal vein and hepatic artery injury in patients with non-malignant disease seems justified. However, it may be questioned whether patients with malignant disease beyond established criteria should be offered liver transplantation.

**Keywords:** Liver Transplantation – Rescue – Iatrogenic injuries – Posthepatectomy liver failure

**Authors contributions**

T.T: Study conception and design, acqusition of data, analysis and interpretetation of data and drafting of manuscript.

J.M.S: Acqusition of data and drafting of manuscript.

K.J.L: Study conception and design, and critical revision of manuscript.

P-D.L: Study conception and design, and critical revision of manuscript.

E.M.AA: : Study conception and design, analysis and interpretetation of data and drafting of manuscript.

All authors have approved the final article.

## INTRODUCTION

Liver transplantation (LT) is the only curative treatment for a wide range of diseases resulting in end-stage liver disease. LT is also offered to patients with hepatocellular carcinoma (HCC) within established criteria for disease stage. However, few studies have evaluated urgent rescue LT as a lifesaving treatment for patients suffering from acute liver failure (ALF) after liver resection or severe injuries to vital structures of the liver. Previous studies have mainly examined LT after fatal bile duct injuries acquired during cholecystectomy [1-8], while other indications such as iatrogenic injuries to the portal vein during bariatric surgery [9], bleeding-complications related to insertion of transjugular intrahepatic portosystemic shunt [10] and ALF after coiling of the hepatic artery [11] have been reported in smaller case series or single cases. Emergent LT as treatment after fatal injuries to the portal vein and hepatic artery during open adrenalectomy [12,4], and LT as a bailout solution after uncontrollable bleeding in relation to resection surgery [13,11] have also been described.

The donation rates in Norway have varied between 20.4 and 21.8 per million of the population during the last five years and the wait list mortality has been below 3%. Our hospital is the only transplant center in Norway, and performs around 100 LTs per year. The current situation with short waiting list and good access to organs has allowed us to explore expanded indications for LT, giving rise to the SECA-study [14-16], the RAPID concept [17] and extended criteria for LT in patients with HCC beyond those established internationally [18]. The aim of the study was to report our institutional experience with rescue LT in patients suffering from ALF due to injuries to the portal vein and hepatic artery or following liver resection.

## METHODS

Between 1984 and 2017, 1510 LTs were performed in our institution. Thirteen LTs were done as urgent procedures due to iatrogenic injuries of vital hepatic vasculature or ALF after prior liver resections. Complete medical files of all patients were available for data-extraction. Data on the corresponding donors were collected from our local registry. The study was approved by the institutional review board according to the general guidelines provided by the regional ethics committee (2015/1442).

All patients received standard triple immunosuppression (IS) with tacrolimus in combination with steroids and mycophenolate mofetil, and except patient no. 12 all received induction therapy with anti-IL-2 receptor antibody (basiliximab) with delayed introduction of tacrolimus due to kidney dysfunction at the time of transplantation. Due to ABO-incompatibility patient no. 10 received 45 g intravenous immunoglobulins for 4 days and a single dose of

850 mg anti-CD20 antibody (rituximab) in addition to standard IS. Due to changes in our IS-protocol during the study period, patient no. 4 and 11 did not receive IVIG/rituximab despite ABO-incompatible LT. All operations were performed using piggy-back technique without use of veno-venous bypass, and only full liver grafts were used. The severity of liver failure was categorized according to MELD score [19].

**RESULTS**

Twelve men and one woman were included in the study. The median age was 55 years (range 22-69). Patient characteristics, indications for liver transplantation and main outcome are presented in Table 1. Six patients (cases 1-6, Table 1) were transplanted due to complications after surgical procedures for non-malignant tumours or conditions.

Seven patients (cases 7-13, Table 1) had undergone radical surgery for various malignancies prior to transplantation.

Perioperative data are presented in Table 2. Median time from the primary procedure until LT was 17 days (range 0-37), and the median waiting-time after the patient was listed for LT was one day (range 0-7). Median MELD score was 33.5 (range 22-40). The median donor age was 56 years (range 18-83). Median cold ischemia time (CIT) was 423 minutes (range 210-642). The median intraoperative blood transfusion was 3000 ml (range 250-27250).

There were three incidents of primary non-function (PNF) or dysfunction requiring early retransplantation after the rescue procedure. All these were related to marginal liver graft quality with no signs of technical issues as the causeof PNF. Two of the primary LTs (A→0) and one retransplantation (AB→A) were ABO-incompatible transplantations. Median ICU-stay after LT was 10 days (range 1-97). Two patients were treated for biopsy proven rejection. Two patients were diagnosed with vascular complications (one with hepatic artery stenosis and one with portal vein thrombosis), and four patients had biliary complications (two patients with leakage from the cystic duct and two with leakage from the hepatico-jejunostomy). Eleven patients were in need of temporary renal replacement therapy after transplantation. Median follow-up time was 70.5 months (range 2.2-171).

Survival time was calculated from the day of LT until December 31, 2017, or to patient death. For the patients that were re-transplanted due to PNF, the calculation is based on the last liver graft. Five and ten-year Kaplan-Meier (KM) estimateted survival for the whole cohort was 64.8% and 51.9%, respectively. The five and ten-year KM estimated survival for the patients with benign disease were both 80.0%, whereas the corresponding results for the patients with malignant disease was 57.1% and 38.1%, respectively (Figure 1 a and b). Table 2 provides further

details on MELD score, perioperative morbidity, waiting time, donor-data and main postoperative complications. Table 3 presents details on the four patients transplanted due to CRLM.

## DISCUSSION

In this report, we present our experience with salvage LT for iatrogenic vascular injuries and for complications to cancer surgery beyond conventional criteria. The study shows that LT is a lifesaving procedure for patients with devastating iatrogenic injuries to the portal vein and hepatic artery and for patients experiencing posthepatectomy liver failure. Long-term survival was achieved for the majority of the patients. However, more than half of the patients with malignant disease died of cancer recurrence.

Salvage LT in case of iatrogenic injuries or posthepatectomy liver failure poses not only medical considerations, but also ethical concerns in the face of organ shortage [20-24]. The availability of organs for transplantation is a crucial limitation, which directs the prioritization at each center and complicates the establishment of generally accepted indications, criteria for acceptance and allocation policies. ALF caused by iatrogenic injuries may be a less controversial indication for LT compared to salvage LT for surgical complications after treatment for malignancies not generally accepted for transplantation. Our study demonstrates that it is difficult to predict the outcome in both circumstances. The cohort includes a heterogeneous group of patients in terms of primary diagnosis, operative procedures, mechanism of liver failure and the general clinical status. This is partly in line with the experiences with LT for ALF within conventional criteria such as toxic liver failure, acute viral hepatitis and idiopathic acute and subacute liver failure [25,26]. However, for these conditions, there are well-established recommendations such as the King´s college guidelines [27]. For iatrogenic injuries, the reported experiences are scarce [11]. Patient 1 and 2 in our cohort underwent LT after vascular injuries, and the primary procedure was performed due to a non-malignant medical condition. Both patients recovered and, in our view, represent cases that should be offered transplantation whenever possible. Thus, these groups are now included in the conventional ALF-group considered for LT in our center. Iatrogenic injuries that occur during surgery for cancer or suspected malignant disease are more challenging. Patient 3 and 4 underwent surgery for suspected renal and duodenal cancer, respectively. However, malignancy was excluded by final histology prior to transplantation in patient 4. The liver failure in this patient was caused by thrombosis in a stent in the hepatic artery placed due to a pseudoaneurysm after a pancreatoduodenectomy. This may be considered as an iatrogenic vascular injury finally

indicating LT. Hepatic artery pseudoaneurysm after pancreatoduodenectomy carries a high mortality rate. Radiological intervention with stent placement is the first-line treatment. However, with complex arterial pathologies, as were the case in the two patients in our study, surgical revascularization, or even LT, may be the only lifesaving option to avoid lethal liver failure. To the best of our knowledge, LT has not been reported as a treatment option in this setting. Patient 3 was transplanted at a time when malignant kidney tumor was still suspected, and the benign diagnosis was confirmed after transplantation. This case poses several questions as no definite diagnosis had been made at the time of transplantation. Our decision to offer this patient transplantation was partly based on the dramatic consequences of an iatrogenic injury that occurred during surgery of a potentially curable kidney tumor.

Eight patients in our cohort underwent a liver resection for suspected or verified malignancy and were transplanted due to remnant liver failure. Two of these patients (5 and 6) did not have cancer and one patient (7) had HCC in a cirrhotic liver where the tumor was within established criteria for LT. However, the fourth and the fifth patient (8, 9) developed liver remnant failure after right hepatectomy for CRLM. All these five patients (5-9) developed grade C posthepatectomy liver failure according to the classification suggested by the International Study Group of Liver Surgery [28]. The remaining three patients with posthepatectmy liver failure were also transplanted after primary surgery for a malignancy outside established criteria for LT. If salvage LT is considered after surgery for suspected malignant disease, a detailed histology report should be available to confirm the diagnosis, and to evaluate the tumor stage and prognosis before a final decision for LT is made. Two of the patients had CRLM (10 and 11), and one had liver metastases from a pancreatic neuroendocrine tumor (12). These indications for LT are certainly controversial in a situation with organ shortage, and three of the four patients with CRLM died due to recurrence of cancer. Our institution has explored the potential benefit of transplanting patients with CRLM without extrahepatic disease in the SECA-study with encouraging results [14]. According to the results from the SECA trial, two of these patients could be considered as low risk based on maximal tumor diameter < 5 cm, pre LT CEA level below 80 μg/L and objective response on chemotherapy (14). One of these patients died 78 months after transplantation and the other patient has still not developed recurrence after 72 months follow-up. Since transplantation for CRLM is still experimental and definitive selection criteria has not been established, it is difficult to predict the potential outcome in the setting of post resection failure. This study show that some patients may have acceptable or even exceptional results. Prolonged disease free and overall survival after LT for CRLM have also recently been published in a cohort from some European centers. Importantly they found that

compassionate transplantation as a salvage procedure was associated with poorer outcome with respect to disease free survival than patients were where LT was a planned procedure [29]. To further conclude regarding the outcome of LT for patients with malignancy beyond the conventional criteria, further studies with larger sample size are needed.

Patient survival in some of the previous reports on the use of urgent LT has not been encouraging, with short-term mortality up to 80 % [7,11]. Table 4 summarizes selected earlier reports on acute rescue LT. These results may partly be related to that the patients were in a dismal state at the time of transplantation such as suffering from infections, sepsis or multiorgan failure. These factors are predictors of poor survival in any candidate undergoing LT regardless of the underlying disease

Certain limitations of this study must be acknowledged. First, this was a retrospective analysis of patients treated at a single institution with all the inherent biases associated with this study design. However, the clinical database used was prospectively maintained and provided complete follow-up data. Second, the sample size was small. However, despite the limitations of the small sample size, this is one of the largest series to date evaluating the outcome of salvage LT for posthepatectomy liver failure and iatrogenic injuries to vital structures of the liver. Last, and most important, the patients were highly selected and underwent LT without a study protocol and predefined criteria. As previously discussed a fortunate donor situation and short waiting times has enabled LT to patients beyond conventional criteria. Thus, in case of organ shortage or other systems for allocation of available donor organs the external validity of the study should be considered with caution.

In conclusion, LT can be a lifesaving procedure for patients suffering from iatrogenic liver injuries or posthepatectomy liver failure. LT for liver failure due to portal vein and hepatic artery injuries in patients with non-malignant disease seems justified. However, it is debatable whether patients with known malignant disease beyond accepted LT-criteria should be offered LT in a situation with organ scarcity.

**Disclosure**

The authors of this manuscript have no conflicts of interest to disclose.

**Funding**

This research did not receive any specific grant from funding agencies in the public, commercial, or not-for-profit sectors.

**Ethical standards**

The study was approved by the institutional review board according to the general guidelines provided by the regional ethics committee (2015/1442). For this type of study formal consent is not required (retrospective study).

**Ethical approval**

All procedures performed in studies involving human participants were in accordance with the ethical standards of the institutional and/or national research committee and with the 1964 Helsinki declaration and its later amendments or comparable ethical standards.

## References


1. Nordin A, Makisalo H, Isoniemi H, Halme L, Lindgren L, Hockerstedt K (2001) Iatrogenic lesion at cholecystectomy resulting in liver transplantation. Transplantation proceedings 33 (4):2499-2500

2. Buell JF, Cronin DC, Funaki B, Koffron A, Yoshida A, Lo A, Leef J, Millis JM (2002) Devastating and fatal complications associated with combined vascular and bile duct injuries during cholecystectomy. Archives of surgery (Chicago, Ill : 1960) 137 (6):703-708; discussion 708-710

3. Fernandez JA, Robles R, Marin C, Sanchez-Bueno F, Ramirez P, Parrilla P (2004) Laparoscopic iatrogeny of the hepatic hilum as an indication for liver transplantation. Liver transplantation : official publication of the American Association for the Study of Liver Diseases and the International Liver Transplantation Society 10 (1):147-152. doi:10.1002/lt.20021

4. Zaydfudim V, Wright JK, Pinson CW (2009) Liver transplantation for iatrogenic porta hepatis transection. The American surgeon 75 (4):313-316

5. Ardiles V, McCormack L, Quinonez E, Goldaracena N, Mattera J, Pekolj J, Ciardullo M, de Santibanes E (2011) Experience using liver transplantation for the treatment of severe bile duct injuries over 20 years in Argentina: results from a National Survey. HPB : the official journal of the International Hepato Pancreato Biliary Association 13 (8):544-550. doi:10.1111/j.1477-2574.2011.00322.x

6. Wang Z, Yu L, Wang W, Xia J, Li D, Lu Y, Wang B (2013) Therapeutic strategies of iatrogenic portal vein injury after cholecystectomy. The Journal of surgical research 185 (2):934-939. doi:10.1016/j.jss.2013.06.032

7. Parrilla P, Robles R, Varo E, Jimenez C, Sanchez-Cabus S, Pareja E (2014) Liver transplantation for bile duct injury after open and laparoscopic cholecystectomy. The British journal of surgery 101 (2):63-68. doi:10.1002/bjs.9349

8. Leale I, Moraglia E, Bottino G, Rachef M, Dova L, Cariati A, De Negri A, Diviacco P, Andorno E (2016) Role of Liver Transplantation in Bilio-Vascular Liver Injury After Cholecystectomy. Transplantation proceedings 48 (2):370-376. doi:10.1016/j.transproceed.2015.12.035

9. Huerta S, Li Z, Livingston EH (2006) Outcome of portal injuries following bariatric operations. Obesity surgery 16 (1):105-109. doi:10.1381/096089206775222203

10. Di Benedetto F, Mimmo A, D'Amico G, De Ruvo N, Cautero N, Montalti R, Guerrini GP, Ballarin R, Spaggiari M, Tarantino G, Serra V, Pecchi A, De Santis M, Gerunda GE (2010) Liver transplantation due to iatrogenic injuries: two case reports. Transplantation proceedings 42 (4):1375-1377. doi:10.1016/j.transproceed.2010.03.077

11. Lauterio A, De Carlis R, Di Sandro S, Ferla F, Buscemi V, De Carlis L (2017) Liver transplantation in the treatment of severe iatrogenic liver injuries. World journal of hepatology 9 (24):1022-1029. doi:10.4254/wjh.v9.i24.1022

12. Tessier DJ, Iglesias R, Chapman WC, Kercher K, Matthews BD, Gorden DL, Brunt LM (2009) Previously unreported high-grade complications of adrenalectomy. Surgical endoscopy 23 (1):97-102. doi:10.1007/s00464-008-9947-3

13. Aseni P, Lauterio A, Slim AO, Giacomoni A, Lamperti L, De Carlis L (2010) Life-saving super-urgent liver transplantation with replacement of retrohepatic vena cava by dacron graft. HPB surgery : a world journal of hepatic, pancreatic and biliary surgery 2010. doi:10.1155/2010/828326

14. Hagness M, Foss A, Line PD, Scholz T, Jorgensen PF, Fosby B, Boberg KM, Mathisen O, Gladhaug IP, Egge TS, Solberg S, Hausken J, Dueland S (2013) Liver transplantation for nonresectable liver metastases from colorectal cancer. Annals of surgery 257 (5):800-806. doi:10.1097/SLA.0b013e3182823957

15. Hagness M, Foss A, Egge TS, Dueland S (2014) Patterns of recurrence after liver transplantation for nonresectable liver metastases from colorectal cancer. Annals of surgical oncology 21 (4):1323-1329. doi:10.1245/s10434-013-3449-9

16. Dueland S, Hagness M, Line PD, Guren TK, Tveit KM, Foss A (2015) Is Liver Transplantation an Option in Colorectal Cancer Patients with Nonresectable Liver Metastases and Progression on All Lines of Standard Chemotherapy? Annals of surgical oncology 22 (7):2195-2200. doi:10.1245/s10434-014-4137-0

17. Line PD, Hagness M, Berstad AE, Foss A, Dueland S (2015) A Novel Concept for Partial Liver Transplantation in Nonresectable Colorectal Liver Metastases: The RAPID Concept. Annals of surgery 262 (1):e5-9. doi:10.1097/sla.0000000000001165

18. Dueland S, Foss A, Solheim JM, Hagness M, Line PD (2018) Survival following liver transplantation for liver-only colorectal metastases compared with hepatocellular carcinoma. The British journal of surgery 105 (6):736-742. doi:10.1002/bjs.10769

19. Kamath PS, Wiesner RH, Malinchoc M, Kremers W, Therneau TM, Kosberg CL, D'Amico G, Dickson ER, Kim WR (2001) A model to predict survival in patients with end-stage liver disease. Hepatology (Baltimore, Md) 33 (2):464-470. doi:10.1053/jhep.2001.22172

20. Biggins SW (2012) Futility and rationing in liver retransplantation: when and how can we say no? Journal of hepatology 56 (6):1404-1411. doi:10.1016/j.jhep.2011.11.027

21. Neuberger J, Mulligan D (2015) Liver allocation: can we ever get it right and should we ever get it right? Hepatology (Baltimore, Md) 61 (1):28-31. doi:10.1002/hep.27359

22. Schaefer B, Zoller H, Schneeberger S (2018) Con: Liver transplantation for expanded criteria malignant diseases. Liver transplantation : official publication of the American Association for the Study of Liver Diseases and the International Liver Transplantation Society 24 (1):104-111. doi:10.1002/lt.24975

23. Zarrinpar A, Busuttil RW (2013) Liver transplantation: past, present and future. Nature reviews Gastroenterology & hepatology 10 (7):434-440. doi:10.1038/nrgastro.2013.88

24. Roberts JP (2015) Transplantation: Allocation of liver transplants-a road to consensus. Nature reviews Gastroenterology & hepatology 12 (11):615-616. doi:10.1038/nrgastro.2015.181

25. Stravitz RT, Kramer DJ (2009) Management of acute liver failure. Nature reviews Gastroenterology & hepatology 6 (9):542-553. doi:10.1038/nrgastro.2009.127

26. Bunchorntavakul C, Reddy KR (2017) Acute Liver Failure. Clinics in liver disease 21 (4):769-792. doi:10.1016/j.cld.2017.06.002

27. McPhail MJ, Farne H, Senvar N, Wendon JA, Bernal W (2016) Ability of King's College Criteria and Model for End-Stage Liver Disease Scores to Predict Mortality of Patients With Acute Liver Failure: A Meta-analysis. Clinical gastroenterology and hepatology : the official clinical practice journal of the American Gastroenterological Association 14 (4):516-525.e515; quiz e543-e545. doi:10.1016/j.cgh.2015.10.007

28. Rahbari NN, Garden OJ, Padbury R, Brooke-Smith M, Crawford M, Adam R, Koch M, Makuuchi M, Dematteo RP, Christophi C, Banting S, Usatoff V, Nagino M, Maddern G, Hugh TJ, Vauthey JN, Greig P, Rees M, Yokoyama Y, Fan ST, Nimura Y, Figueras J, Capussotti L, Buchler MW, Weitz J (2011) Posthepatectomy

liver failure: a definition and grading by the International Study Group of Liver Surgery (ISGLS). Surgery 149 (5):713-724. doi:10.1016/j.surg.2010.10.001

29. Toso C, Pinto Marques H, Andres A, Castro Sousa F, Adam R, Kalil A, Clavien PA, Furtado E, Barroso E, Bismuth H (2017) Liver transplantation for colorectal liver metastasis: Survival without recurrence can be achieved. Liver transplantation : official publication of the American Association for the Study of Liver Diseases and the International Liver Transplantation Society 23 (8):1073-1076. doi:10.1002/lt.24791

**Table 1. Patient characteristics, indications for liver transplantation and main outcome**

| Pat | Age/sex | Disease | Primary procedure | Primary procedure performed at OUS | Cancer | Reason for LT | Time from primary procedure to LT (days) | Follow up (months) | Alive | Outcome |
|---|---|---|---|---|---|---|---|---|---|---|
| 1 | 46/M | Gallstones. HCV | Laparoscopic cholecystectectomy | No | No | Iatrogenic injury to PV and HA | 2 | 121 | Yes | Recurrence of HCV, otherwise well |
| 2 | 22/M | Knife stab injury to abdomen | Abdominal packing for hemorrhage | No | No | Liver failure after hypovolemia and abdominal packing | 4 | 71 | Yes | Recovered |
| 3 | 42/F | Suspected renal carcinoma (benign) | Laparoscopic nephrectomy | No | No | Iatrogenic injury to PV and HA | 17 | 97 | Yes | Recovered. Stented HAS. Repeated episodes of cholangitis due to stenotic biliary tracts in segment 5/8, successfully treated with partial PV embolization |
| 4 | 62/M | Suspected duodenal cancer (high grade dysplasia) | Pancreatoduodenectomy | Yes | No | Liver failure after occlusion of stented HA-pseudoaneurysm | 37 | 7 | No | Death due to pneumonia and sepsis |
| 5 | 63/M | Suspected HCC (regeneration nodules) | Right hepatectomy | Yes | No | Remnant liver failure | 27 | 13 | Yes | Recovered |
| 6 | 52/M | Suspected CCA (benign IgG4 inflammation) | Right hepatectomy | Yes | No | Remnant liver failure | 17 | 2 | Yes | Recovered |
| 7 | 49/M | HCC 2,1 cm / Child A cirrhosis | Local liver resection | No | Yes | Remnant liver failure | 22 | 110 | Yes | No signs of recurrence, doing well |
| 8 | 69/M | CRLM | Right hepatectomy | Yes | Yes | Remnant liver failure (PV thrombosis) | 9 | 78 | No | Death due to recurrence of cancer |
| 9 | 69/M | CRLM | Right hepatectomy | No | Yes | Remnant liver failure | 20 | 72 | Yes | Recovered, no signs of recurrence |
| 10 | 67/M | CRLM | Planned right hepatectomy | Yes | Yes | Iatrogenic injury to left branches of PV and HA | 6 | 45 | No | Death due to recurrence of cancer |
| 11 | 63/M | CRLM | Resection and auto-transplantation liver | Yes | Yes | Remnant liver failure due to intraoperatively massive bleeding | 0 | 9 | No | Death due to recurrence of cancer |
| 12 | 42/M | PNET /liver metastasis | Planned distal pancreatic resection | Yes | Yes | Iatrogenic injury to PV and HA | 1 | 177 | Yes | Recovered, no signs of recurrence |
| 13 | 55/M | Duodenal cancer | Pancreatoduodenectomy | Yes | Yes | Liver failure after HA-pseudoaneurysm and HAT | 22 | 24 | No | Death due to recurrence of cancer |

OUS, Oslo University Hospital; LT, liver transplantation; PNET, pancreatic neuroendocrine tumor; PV, portal vein; HA, hepatic artery; HCV, hepatits C virus; HCC, hepatocellular carcinoma; HAS, hepatic artery stenosis; CRLM, colorectal liver metastasis; HAT, hepatic artery thrombosis

**Table 2. Details on patients, procedures and results**

| Pat | MELD score | AKF w/ dialysis at time of listing | WT for LT (days) | Donor age (years) | Blood type donor→ recipient (LT1 / LT2) | CIT (min) | Blood trans-fusion perop (ml) | ICU-stay (days) | ACR | Graft-loss within 30 days | Cause of re-tx | AKF postop | Vasc. compl | Biliary compl | Alive | DWFG |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| 1 | 40 | Yes | 0 | 62 | 0→0 | 642 | 250 | 10 | No | | | Yes | No | No | Yes | |
| 2 | 38 | No | 2 | 83 / 69 | A→A / 0→A | 380/334 | 250 / 2000 | 19 | No | Yes | PNF | Yes | No | No | Yes | |
| 3 | 34 | Yes | 4 | 46 / 58 | 0→0 / 0→0 | 514/202 | 3750 / 500 | 78 | No | Yes | PNF | Yes | Yes | Yes | Yes | |
| 4 | 32 | Yes | 1 | 56 / 62 | A→A / AB→A | 531/651 | 3000 / 750 | 97 | No | Yes | PNF | Yes | Yes | Yes | No | Yes |
| 5 | 40 | Yes | 0 | 26 | 0→A | 492 | 7500 | 9 | No | | | Yes | No | Yes | Yes | |
| 6 | 22 | No | 7 | 54 | 0→A | 350 | 2000 | 3 | Yes | | | No | No | No | Yes | |
| 7 | 27 | No | 1 | 31 | 0→0 | 210 | 5400 | 20 | No | | | No | No | No | Yes | |
| 8 | 39 | Yes | 1 | 55 | A→A | 423 | 3500 | 14 | No | | | Yes | No | No | No | Yes |
| 9 | 40 | Yes | 2 | 62 | 0→0 | 459 | 1250 | 5 | No | | | Yes | No | No | Yes | |
| 10 | 29 | Yes | 2 | 69 | A→0 | 413 | 1000 | 1 | No | | | Yes | No | No | No | Yes |
| 11 | 22 | No | 0 | 70 | A→0 | 380 | 27 250 | 67 | Yes | | | Yes | No | No | No | Yes |
| 12 | N/A | Yes | 1 | 59 | 0→0 | 275 | 3000 | 9 | No | | | Yes | No | Yes | Yes | |
| 13 | 33 | Yes | 2 | 18 | A→A | 703 | 3250 | 8 | No | | | Yes | No | No | No | Yes |

MELD, Model for End-Stage Liver Disease; AKF, acute kidney failure; WT, waiting time; LT, liver transplantation; CIT, cold ischemia time; ICU, intensive care unit; ACR, acute cellular rejection; Re-tx, retransplantation; DWFG, dead with functioning graft; N/A, not applicable; PNF, primary non-function

**Table 3. Details on patients undergoing liver transplantation for colorectal liver metastasis**

| Pat | Diameter of largest tumor (cm) | Number of tumors | Last CEA before LT (µg/l) | Preop. chemotherapy (Y/N) | Response to chemotherapy according to RECIST (Y/N/SD) | Time from primary diagnosis to LT (months) | Survival (months) | Alive (Y/N) |
|---|---|---|---|---|---|---|---|---|
| 8 | 3.0 | 1 | 3.5 | Y | Y | 5.9 | 78 | N |
| 9 | 2.3 | 3 | <1.0 | Y | Y | 11.5 | 72 | Y |
| 10 | 14.0 | 4 | 189 | Y | SD | 7.3 | 45 | N |
| 11 | 5.5 | 2 | 137 | Y | SD | 24.5 | 9 | N |

CEA, carcinoembryonic antigen; LT, liver transplantation

**Table 4. Selected studies on acute rescue liver transplantation**

| Author, year | No. pat. with acute LT* | Reason for performing LT | Outcome |
|---|---|---|---|
| Nordin et al, 2001[1] | 1 | Transection of hilum during OC | Alive after 2 years follow up |
| Fernandez et al, 2004[3] | 1 | Ligation of PV and HA during LC | Death after 35 days |
| Zaydfudim et al, 2009[4] | 2 | Transection of hilum during 1) open right adrenalectomy and 2) LC | Alive after 2 and 6 years follow up |
| Parilla et al, 2013[7] | 5 | Severe injury to hilum with BVI during LC | 4 of 5 patients died within 30 days after LT |
| Leale et al, 2016[8] | 2 | 1) Acute-on-chronic liver failure after OC (Child C)<br>2) Massive liver-bleeding during LC | Alive after 2 and 8 years follow up |
| Huerta et al, 2006[9] | 3 | Severe injury to PV during bariatric surgery | Death after few days, 6 weeks and 8 weeks |
| Benedetto et al, 2010[10] | 2 | Bleeding complications after TIPS | Long term survival |
| Lauterio et al, 2017[11] | 2 | 1) Massive bleeding during liver resection<br>2) ALF after HA embolization due to bleeding | Long term survival |
| Tessier et al, 2009[12] | 1 | Transection of hilum during lap. adrenalectomy | Long term survival |

LT, liver transplantation; * defined as LT within 6 weeks after time of primary surgery; OC, open cholecystectomy

PV, portal vein; HA, hepatic artery; LC, laparoscopic cholecystectomy; BVI, bilio-vascular injury

TIPS, transjugular intrahepatic portosystemic shunt

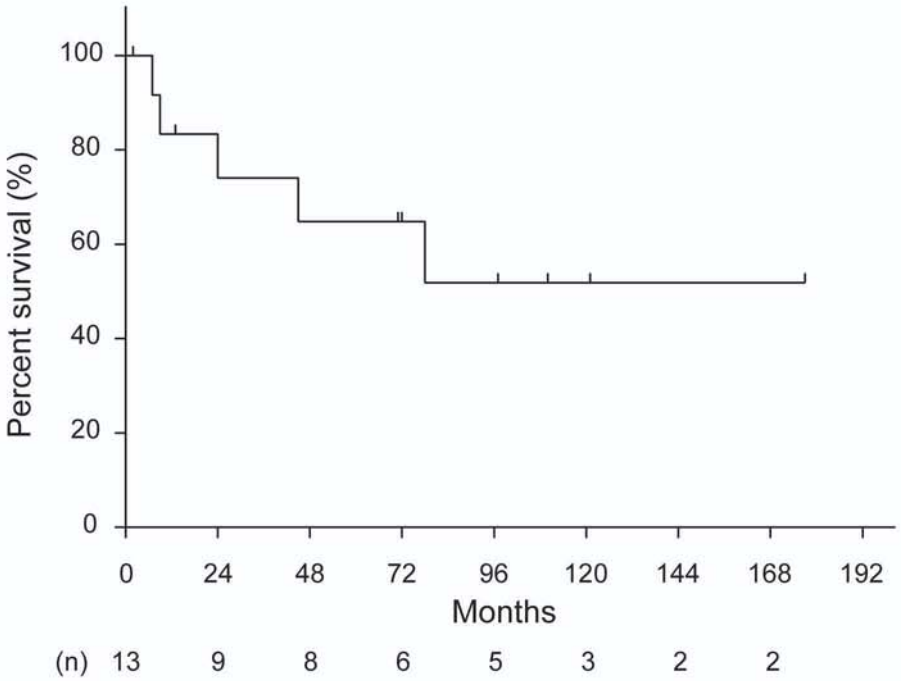
**Figure legends**

**Figure 1a.** Kaplan-Meier plot of patient and graft survival for the whole rescue group. For the three patients that were re-transplanted due to primary non-function, the calculation is based on the last liver graft.

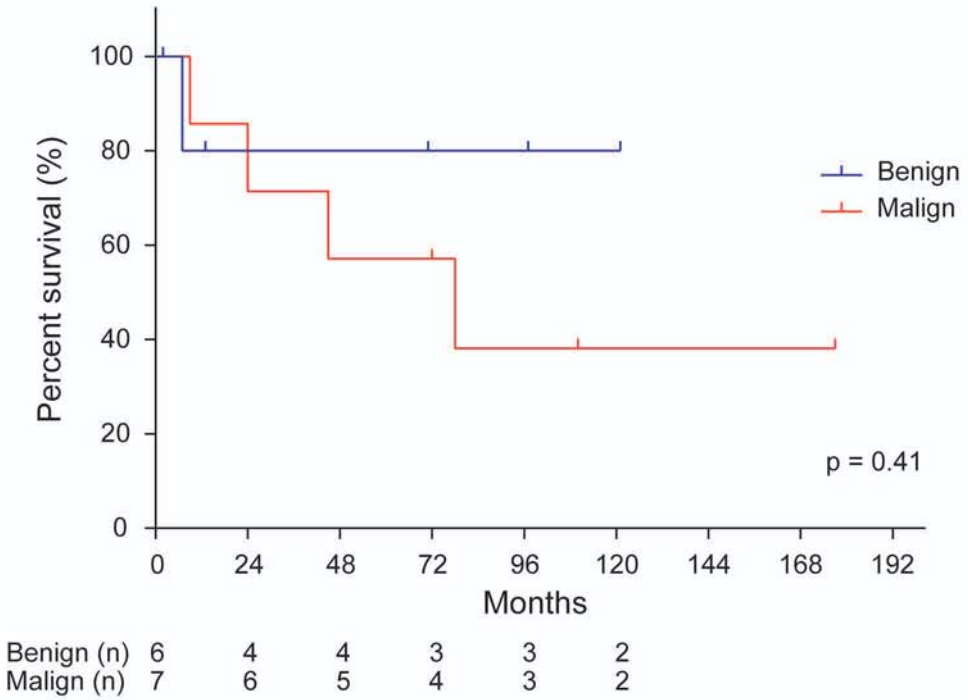
**Figure 1 b.** Kaplan-Meier plots of patient and graft survival in the benign group vs malign group. For the three patients that were re-transplanted due to primary non-function, the calculation is based on the last liver graft. Curves were compared using the log-rank test.

Figure 1.

a)


Percent survival (%)
100
80
60
40
20
0
0
24
48
72
96
120
144
168
192
Months
(n) 13 9 8 6 5 3 2 2

b)


Percent survival (%)
100
80
60
40
20
0
0
24
48
72
96
120
144
168
192
Months
Benign
Malign
p = 0.41
Benign (n) 6 4 4 3 3 2
Malign (n) 7 6 5 4 3 2