ADULT-TO-ADULT LIVING DONOR LIVER TRANSPLANTATION COHORT STUDY

Enclosed please find a synopsis of the A2ALL Cohort study and summaries of the following sub-studies: Restoration of Hepatic Function in Living Liver Donors (DQLFT) and Genomics and Regeneration in the Transplant Setting (GRITS).

For information on the A2ALL sub-study Genes Related to HCC Progression please visit:
http://www.vcucriticalcare.com/transplant/laboratory_research/GR2HCC/gr2hcc.htm

1. Introduction

Adult to adult living donor liver transplantation (LDLT) is a relatively new procedure increasingly used at major transplantation centers. Too few cases are performed at any one center and approaches to the patient and donor are too diverse across centers to provide reliable and generalizable information on donor and recipient outcomes from individual centers. Therefore, the National Institutes of Health has organized a network of nine leading liver transplantation centers and a data coordination center (DCC) to accrue and follow sufficient numbers of patients being considered for and undergoing LDLT to provide generalizable results from adequately powered studies. This network has established the Adult to Adult Living Donor Liver Transplantation Cohort Study (A2ALL) that will conduct both retrospective and prospective studies of LDLT.

2. Background/Significance

Over the last 20 years liver transplantation has become the standard of care and the only cure for end stage liver disease. Its success has led to over 4,000 transplants performed yearly. But there are at least 17,000 patients on the transplantation list awaiting deceased donor (DD) liver donation. As the waiting list has expanded, waiting time has also grown. As a result, patient mortality has increased while awaiting transplantation, and patients are often critically ill by the time of transplantation (*1*). Among possible remedies, living donor transplantation has become widely accepted for pediatric transplantation. Adult-to-adult LDLT is a more challenging procedure and entails potentially greater risk to the donor because of the larger portion of liver that is required (*2*). Right lobe adult-to-adult LDLT is a recently developed procedure, but nearly a thousand have already been performed in the United States. Although still a small number relative to the several thousand adult deceased donor liver transplants (DDLT) performed annually, LDLT has the potential for changing the face of liver transplantation. Not only does LDLT avoid the lengthening waiting period for a deceased donor transplant, it greatly reduces the ischemic period of the transplanted organ, allows more time for evaluation of the donor, and changes the operation from an emergency into a scheduled procedure. The major disadvantage of LDLT is that it is a difficult and potentially fatal operation for the donor. It also provides the recipient with a smaller portion of liver than would have been received with deceased donor transplantation.

The research objectives of the LDLT Cohort Study concern factors that influence the outcomes of adult-to-adult LDLT as well as a study of the biological differences between living donor (LD) and DD grafts in the recipients. Adult patients and potential donors being considered for LDLT will be recruited into this longitudinal cohort study. Recipients and their donors will be followed for sufficient time to determine outcomes related to LDLT. These outcomes will be compared with those of transplant candidates who are evaluated for but do not receive LDLT. The primary objective concerns comparison of morbidity and mortality of patients who receive LDLT with a group or groups of patients with similar illnesses and prognoses. A critical question to answer with this information is how the outcomes of LDLT compare with those of deceased donor transplantation. Transplant physicians need this information on outcomes to advise

prospective recipients and donors. Therefore, sufficient recipient and donor pairs will be recruited to determine whether recipients of LDLT have substantially different survival than non-LDLT recipients. A large number of donors and recipients from several geographically distributed institutions will be necessary to reliably determine if outcomes are different with the two approaches.

The differences between LDLT and DDLT are inherent both in the application and the biology of the procedures. By its nature DDLT includes uncertainty about both the time of transplantation and the condition of the recipient at the time of the eventual transplantation. Because the LDLT is elective, pre-transplant morbidity and mortality are minimized in the LDLT group. This means that pre-transplant morbidity and mortality are major areas in which the potential advantage of LDLT needs to be quantified. In contrast to the expected benefits of enhanced access to transplantation, the recipient of LDLT faces a procedure which is more complex than DDLT and which provides only a partial graft. Thus, the penalty paid by opting for LDLT rather than opting for DDLT also needs to be quantified.

LDLT offers a unique opportunity to study human liver regeneration and its impact on several key clinical biological issues in transplantation: the immune response, the recurrence of hepatitis C (HCV), and the approach to the treatment of hepatocellular carcinoma HCC (a growing indication for liver replacement therapy). In this protocol we plan to systematically collect clinical and biological data in recipients of LDLT and appropriate control recipients of DDLT to compare the impact of the hemigraft on these parameters. Entry into the cohort study will result in a relatively standardized clinical management protocol and the collection of the defined set of data points for all patients entered. A subset of patients may be recruited into ancillary studies that will entail a more extensive examination of focused topics.

The other major mandate in the development of the cohort study is the prospective assessment of the impact of donation on the healthy living donor. There is widespread interest in this subject among the medical community and the public at large, brought on in large measure by the recent, highly publicized death of a living donor in 2002. Concerns about the ethical issues regarding donor safety will be addressed by the organized study of the surgical, biological, and psychosocial effects of donation on donors compared to a control population of potential donors who are not selected for the procedure.

3. Study Objectives/Specific Aims

The primary study objective is to analyze the effect of choosing living donation rather than the wait for a deceased donor liver transplant. The principal hypothesis is that pursuit of a living liver allograft leads to decreased pre-transplant morbidity and mortality and better long term outcomes for patients starting from the point at which listed patients have a potential donor evaluated with at least a history and physical examination. Emerging data suggest that LDLT provides an inferior graft because of small size and technical complexity when compared to a whole liver used for DDLT. The magnitude of

the disadvantage to the LD graft will be assessed by comparing results between LDLT and DDLT from the time of transplant. Finally, the study of the donor is included as a primary objective because of the tremendous importance of this issue to the patient and the public.

Secondary objectives will address selected biological and clinical issues in transplantation structured around the comparison between DDLT and LDLT.

3.1. Primary Aim 1: To quantify the impact of choosing LDLT on the candidate for transplantation.

1. The natural history of choosing to pursue LDLT versus waiting for a DDLT will be characterized. The overall survival comparison is between those receive LDLT versus those with a donor evaluated for LDLT but who do not receive LDLT. Time to transplantation and time to death will be determined.
2. Comparative analysis of pre-transplant morbidity and resource utilization will be determined by comparing the overall cohort from the time of enrollment.

3.2. Primary Aim 2: To characterize the differences between LDLT and DDLT in terms of post-transplant outcomes including patient and graft survival, surgical morbidity, and resource utilization on the recipient of a transplant.

1. Patient and graft survival analysis starting from the time of transplantation
2. Comparison of the incidence of defined medical and surgical complications after transplant between LDLT and DDLT
3. Comparison of resource utilization (hospitalization and emergency room visits) between LDLT and DDLT.

3.3. Primary Aim 3: To determine the short and long term health and quality of life (QOL) impact of donation, including (a) morbidity after liver donation, and (b) long term health-related QOL of donors compared to a control population.

1. To determine the rate of significant morbidity after liver donation.
2. To evaluate long term health-related QOL of donors compared to persons who were evaluated but did not donate.

3.4. Primary Aim 4: To standardize and assess the role of “informed consent” in affecting the decision to donate and satisfaction after living liver donation.

1. To measure the capacity of potential donors to understand information that is presented and to stratify the potential donor’s capacity to understand

information in general and the delivered information regarding the donation in specific.
2. To measure the motivations of the potential donors with standardized instruments and to determine if certain personality characteristics are associated with a more favorable predisposition to proceed to donation.
3. To assess whether disclosed information or life situations are the main influential factors in the potential donor's decision to proceed or withdraw from the donation process.
4. To correlate donor "satisfaction" with measurable outcomes of the donor, recipient, or perceptions of family support.
5. To measure the acceptance of adverse clinical outcomes, as a function of understanding of the disclosed risks versus the presence of life situational pressures.

3.5. Secondary Aim 1: To compare the severity of recurrence of hepatitis C between LDLT and DDLT recipients.

Primary Objective

1. To determine whether HCV disease progression differs in patients receiving LDLT compared to DDLT.

Secondary Objectives

1. To determine if recurrent HCV disease at one year (± 3 months), as observed histologically, is more frequent and severe in patients undergoing LDLT as compared to DDLT transplant.
2. To compare the rate of fibrosis progression (change in Ishak fibrosis score (*3*) per year) in LDLT and DDLT recipients by biopsies at months 3, 12, 24, and 36 after transplantation.
3. To compare time to recurrent disease between LDLT and DDLT recipients as determined by proportion of patients with histological evidence of recurrent HCV at 3 months.
4. To determine if HCV viral level at day 7 and months 1, 3, 12, 24 and 36 months differ in LDLT and DDLT recipients, and whether viral level is predictive of disease severity.
5. To determine if rejection episodes requiring treatment occur at a higher rate in HCV patients who undergo LDLT as compared to DDLT transplant and to correlate this frequency of treatment of rejection to aggressive recurrence of HCV as defined histologically.
6. To compare biochemical markers of disease activity (ALT/AST/total bilirubin) at 3 and 12 months and annually in LDLT and DDLT.
7. To determine if cholestatic hepatitis in transplanted patients with HCV occurs in a higher proportion of LDLT as compared to DDLT recipients.
8. To compare graft loss and patient survival between LDLT recipients and DDLT recipients.

3.6. Secondary Aim 2: Recurrence of HCC for DDLT versus LDLT.

Primary Objectives

1. To determine if LDLT is associated with decreased death on waiting list from progressive tumor growth versus DDLT.
2. Assess comparative HCC recurrence following LDLT or DDLT.
3. Compare long-term survival and disease free survival in patients who undergo LDLT or DDLT.

Secondary Objectives

1. Determine if LDLT recipients require a reduced number of palliative ablative procedures to control HCC when compared to those who wait for DDLT.
2. Compare rates of surgical and post-operative complication in HCC recipients of LDLT and DDLT.

3.7. Secondary Aim 3: To systematically characterize liver regeneration and function in donors and recipients.

Donors and recipients enrolled in the cohort study will be evaluated for evidence of recovery of liver mass and function following the surgical procedures (partial transplantation for recipients of LDLT, and partial hepatectomy for donors).

In the cohort protocol all donors and recipients of LDLT will undergo standardized assessments of liver volume and function to characterize the rate of restoration of the liver. In the recipient, in which the relative size of the graft will vary based on the unique donor/recipient combinations, the large sample provided in the study will permit us to correlate graft function with a number of donor and recipient parameters.

Primary Objective

1. To measure hepatic function and mass in living donors at enrollment, intraoperatively, and following hepatectomy, in order to determine whether return of hepatic function following donation correlates with rate of liver volume regeneration, biochemical impairment, and clinical events, and to see whether return of function is complete by 3 months post-resection.

Secondary Objectives

1. To correlate liver function in donors with long-term health outcomes and the incidence of clinical complications.
2. To correlate success or failure of regeneration with a series of selected clinical and laboratory variables in donors and recipients.
3. To collect liver biopsy and serum samples prospectively from a large series of donors and recipients which may form the basis for subsequent characterization of protein and gene expression of selected inflammatory and growth-related molecules.

3.8. Secondary Aim 4: To evaluate differences in the immune response to LDLT vs. DDLT grafts.

Primary objectives

1. To determine whether LDLT, which is associated with a regenerating liver, meaningfully increases the incidence of clinical rejection. In the cohort, we will compare the incidence of immunologic complications, specifically the incidence and severity of rejection between LDLT and DDLT in a defined set of patients with a sample large enough to detect meaningful differences in the rejection rate.
2. To systematically collect serum and tissue samples that can be used to correlate clinical parameters in donors and recipients with immunologic outcomes, as defined by clinical and histologic endpoints.

3.9. Secondary Aim 5: To establish a robust data and sample repository on liver transplantation that may be used to study clinical and biological questions as new technologies and resources become available.

Primary objectives

1. To facilitate additional studies on samples and data collected in this study, thus enhancing the value of this and future investigations.
2. To ensure that samples are stored under uniform conditions, and to simplify access by other scientists to samples. Similarly, study datasets will be maintained to facilitate new analyses after the study closes.
3. To allow cost effective and high quality processing of genetic samples.

4. Investigational Plan

Potential recipients for transplantation will be evaluated and invited to participate in the study if they are eligible for LDLT using standard criteria for this procedure according to the practice of the transplant center. Recipients will enter the cohort within four weeks of the time a potential donor is scheduled for evaluation at the transplant center with an initial screening history and physical examination (H&P) (see Figure 1). Our preliminary data indicate that, after initial screening of a potential living donor, at least one-half of recipient candidates fail to receive LDLT and go on to wait for DDLT. These latter patients form the recipient control subjects of the study whose fate on the waiting list will be compared to those who undergo LDLT. The potential donors will be enrolled at the time of the initial H&P and will either go on to donate, or may serve as a control population for assessment of the impact of donation on the donors.

We will recruit additional patients (potential and actual recipients, actual donors and donor candidates who have not yet donated, but are early enough in their donation evaluation so that it is unclear whether they will go on to donation) from the A2ALL Retrospective Study (Grant 5 R01 DK62498-02) who are still alive at the start of the cohort study. We will also recruit those patients (recipients, recipient candidates, donors and donor candidates still being evaluated) whose donor evaluation occurred between the

end of the Retrospective Study (2/28/2003) and the start of this prospective study at each site. Another cohort who will be approached for participation are those subjects (recipient candidates) whose date of donor evaluation occurs more than 4 weeks from the time the patient is approached. These subjects will be consented, despite the fact that they will have already passed the entry milestone of the living donor evaluation. Donor candidates who have not yet donated will be utilized as donor controls. Data from time of listing to cohort study enrollment will be collected retrospectively. Subjects will be followed prospectively according to the cohort study schedule of events, starting at the time of their enrollment. This enables a seamless capture of data and analysis of living donor transplantation from its inception into the future.

1 Hypothesis

Quantitative hepatic function and hepatic mass of living liver donors returns to normal (baseline) within 6 months of right hepatic lobe resection.

2 Background and Significance:

A major question for donors of a right lobe for transplant is the pace and completeness of restoration of normal liver function after donor right hepatic lobectomy. A primary concern in adult-to-adult living donor liver transplantation (LDLT) is donor safety and outcome. Stringent selection criteria have been adopted to minimize potential operative mortality and morbidity. Recently reported rates of donor mortality are < 0.3%, important morbid events are noted in upwards of 15% of cases, and the average length of hospital stay is approximately six days (1). Despite these favorable operative and perioperative statistics, little is known about the rate and extent of return of hepatic function and the relationship of hepatic function to long-term clinical outcome. Existing literature suggests that liver size of donors returns to approximately 80% to 100% of baseline by 14 days but biochemical abnormalities may persist beyond two months (2, 3). To answer these questions it is necessary to assess hepatic function in a quantitative fashion in living liver donors prior to and after hepatic resection.

Investigators have used the clearance of aminopyrine, antipyrine, bile acids, caffeine, erythromycin, galactose, indocyanine green, lidocaine, midazolam, omeprazole, dextromethorphan and phenacetin to assess hepatic function at the microsomal, mitochondrial and cytosolic level. Clearances of test compounds are typically defined as dependent upon either hepatic metabolism (aminopyrine, antipyrine, caffeine, erythromycin) or hepatic blood flow (bile acids, indocyanine green [ICG]) (4, 5). The SPECT liver-spleen scan measures perfused hepatic mass, which correlates with ICG clearance and other tests of hepatocyte function (6, 7). Each quantitative test has advantages and disadvantages over other tests and few studies have compared multiple tests within the same cohort of patients.

Two recent studies have examined restoration of volume and hepatic function in donors after right lobectomy for LDLT (8, 9). Humar and colleagues studied CT liver volumes 3 months after liver transplantation in 24 donors of right lobe grafts, 24 recipients of right lobe grafts and 11 recipients each of right or left sided split deceased donor grafts (8). All recipient grafts increased to 100% or more of calculated ideal liver volume. In contrast, donor volume was only 78.6% of calculated ideal liver volume. Drawbacks of this study were the absence of baseline measurement of volume in donors and lack of quantitative testing of liver function. Nadalin and colleagues studied liver volume by MRI and galactose elimination capacity (GEC) in 27 donors (9) at baseline prior to surgery, and at 10 d, 90d, 180d, and 360d after an approximately 60% hepatectomy. Liver volume returned to 74% of baseline by day 10 but remained only 83% of baseline after follow-up for one year. GEC at 10 d was only 50% of baseline, despite the rapid return of volume, implying significant impairment of hepatocyte function at this early time point. At subsequent time points GEC exceeded baseline values suggesting complete restoration of function or over-compensation for diminished hepatocyte mass.

No study has used multiple quantitative tests or assessment of functional hepatocyte mass by SPECT liver scan to measure changes in hepatic function longitudinally in donors after LDLT.

Many factors, including the amount of residual liver mass after resection, patient age, and hepatic steatosis, may affect human hepatic regeneration. In addition, regeneration after resection is linked to a number of neurohumoral factors and metabolic events. Despite the wide breadth of knowledge concerning potential determinants and mediators of regeneration, none have been critically evaluated or correlated with return of hepatic function or mass in humans after resection. Living donors undergoing right lobe hepatectomy for LDLT are healthy without systemic or hepatic disease. For this reason, study of living liver donors provides a unique opportunity to investigate the normal regenerative and restorative pathways necessary for recovery following resection of the human liver.

3 Study Objectives/Specific Aims

3.1 Overall Aim

The primary objective of this study is to define the rate and degree of return of hepatic function and mass in living liver donors using multiple quantitative liver function tests (QLFT) and hepatic imaging. Hepatic function and hepatic mass after donor right hepatic lobectomy will be compared to baseline measurements prior to lobectomy.

3.2 Specific Aims

The specific aims of this study are to:

3.2.1 Specific Aim I

Assess changes in the following quantitative measures of hepatic metabolic capacity in living liver donors prior to and at 5 to 10 days, 3 months, and 6 months after donor hepatic lobectomy:

a) Salivary caffeine clearance
b) Erythromycin breath test
c) Galactose Elimination Capacity

3.2.2 Specific Aim II

Assess changes in the following quantitative measures of hepatic blood flow and portal shunt in living liver donors prior to and at 5 to 10 days, 3 months, and 6 months after donor hepatic lobectomy:

a) Hepatic blood flow: intravenous cholate clearance
b) Portal shunt: oral and intravenous cholate (dual isotope method)

3.2.3 Specific Aim III

Assess changes in post-resection regeneration volume by hepatic imaging in living liver donors prior to and at 5 to 10 days, 3 months, and 6 months after donor hepatic lobectomy:

a) Volume: magnetic resonance imaging
b) Functional hepatic mass: SPECT liver-spleen scan

3.2.4 Specific Aim IV

Determine the relationship among several measures of hepatic recovery after resection, including hepatic function as measured by QLFT, liver volume and liver functional mass, standard laboratory tests of liver function, and clinical events

3.2.5 Specific Aim V

Collect serum to be stored for subsequent analyses of factors related to liver regeneration that could be correlated to hepatic function, hepatic volume, and functional hepatic mass.

Living liver donors will be invited to undergo restudy at 12 months if hepatic metabolic capacity, hepatic blood flow or shunt, or hepatic mass is not within 85% of baseline values at 6 months.

GRITS Study Synopsis

The increase in the number of patients with end stage liver disease has led to rapid growth of the waitlist for transplantation and increased efforts to enlarge the donor pool with and adult-to-adult living donor (LD) liver transplantation. Fundamental differences between LD and deceased donor (DD) grafts are related to the differential magnitude of the proinflammatory response to ischemic injury, and the need for graft regeneration. During recovery and regeneration the liver must maintain metabolic and synthetic homeostasis, and if unable, the graft will express various degree of dysfunction. Although detailed models of liver injury and regeneration have been defined in rodents, the molecular specifics of how a human liver initiates mechanisms of recovery and regeneration following transplantation have yet to be clearly defined. Our first Aim is to determine and validate the molecular pathways that associated with the recovery within LD liver grafts in comparison to DD grafts. We hypothesize that early molecular proinflammatory and regeneration profiles may be diagnostic and predictive of subsequent graft function. Qualitative and quantitative failure to initiate and support the progression of molecular pathways of recovery will result in a spectrum of liver graft dysfunction. This Aim will utilize high density DNA microarrays to identify gene expression profiles linked through clinical outcome data to successful liver regeneration and graft recovery. The second Aim intends to determine the interrelation of donor and recipient clinical variables with the successful initiation and progression of recovery pathways in the allograft. Our hypothesis is that specific clinical conditions may contribute to the intensity of the proinflammatory response, impact on its resolution, affect cell cycle activation and subsequent regeneration, and present an overload on metabolic pathways in the allograft. The clinical expression of graft function under a given clinical condition may be diagnosed and predicted by gene expression profiles of proinflammatory and regenerative pathways in early and subsequent liver biopsies. Our hypothesis will be tested in the setting of DD and LD liver transplantation as an Ancillary study to the ongoing NIDDK multi-center Adult to Adult Living Donor Liver Transplant (A2ALL) consortium. This prospective cohort study will be used to recruit patients and follow clinical outcomes. The results of this study may be used for the diagnosis and management of clinical conditions in the immediate post liver transplant period. Understanding the mechanistic biology of graft recovery and pathways of liver regeneration will provide important data for designing treatment modalities that will prevent injury and enhance recovery and regeneration.

This information can be found at:
http://crisp.cit.nih.gov/crisp/CRISP_LIB.getdoc?textkey=7477974&p_grant_num=5R01DK073192-03&p_query=&ticket=79052824&p_audit_session_id=370224298&p_keywords=