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UDCA　Randomized control study

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UDCA　Cohort study

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UDCA　Meta-analysis

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UDCAの投与量の設定

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Clinical Guideline of Primary Biliary Cirrhosis 2012

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Predicting Operational Tolerance in Pediatric Living-Donor Liver Transplantation by Absence of HLA Antibodies

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Background. The role of anti–human leukocyte antigen (HLA) antibodies in operational tolerance (OT) after pediatric living-donor liver transplantation (LDLT) remains inconclusive. We investigated whether the presence of HLA antibodies impeded the development of OT.

Methods. We retrospectively examined the prevalence of anti-HLA antibodies in pediatric LDLT recipients before transplantation and at 3 weeks after transplantation and analyzed the significance of those antibodies in relation to later OT. Forty pediatric LDLTs were performed between April 1996 and December 2000 and followed up through July 2011, with sera available for measurement of HLA antibodies. Seventeen patients achieved OT (mean follow-up, 4571.9±544.7 days) and 23 patients did not achieve OT (mean follow-up, 4532.0±425.4 days). Protocol liver biopsy was done for 14 OT patients and 16 non-OT patients. Their sera were tested for anti-HLA class I and II antibodies using the LABScreen single antigen beads test, in which a 1000 mean fluorescence value was considered positive.

Results. The prevalence of antibodies after transplantation in non-OT patients was higher than in OT patients (95.2% vs. 73.3%; $P<0.001$). The highest mean fluorescence intensity of antibodies was significantly higher in non-OT patients than in OT patients. The prevalence of HLA-B, HLA-C, HLA-DQ, and HLA-DR antibodies was significantly higher in non-OT patients than in OT patients. The highest mean fluorescence intensity of HLA-A, HLA-B, and HLA-DQ observed in non-OT patients was significantly higher than those in OT patients.

Conclusions. In our study, posttransplantation HLA antibodies were associated with the future absence of OT. A prospective study with more patients is necessary to confirm the predictive value of HLA antibodies for OT.

Keywords: Operational tolerance, Human leukocyte antigen antibodies, Pediatric, Living-donor liver transplantation.

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Despite continued improvements in controlling rejection by immunosuppressive drugs, their serious side effects persist, including increased risk of infection, diabetes, renal dysfunction, and malignancy. Thus, the posttransplantation attainment of operational tolerance (OT) is highly desirable, with OT defined as prolonged survival of a transplanted organ without immunosuppression and without graft rejection, a state especially desirable for pediatric patients (*1, 2*). Unfortunately, although immunomodulatory strategies efficiently induce tolerance in animal models (*3–9*), reaching OT is difficult after clinical organ transplantation in general. The liver, however, is believed to have immunomodulatory

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TABLE 1. Characteristics of LDLT recipients

	OT	Non-OT	P
No. transplants (%)	17 (42.5)	23 (57.5)	
Recipient			
Mean age, yr	2.2±2.7	4.3±4.6	NS
Male (%)	11.8	52.2	0.008
Primary disease, biliary atresia (%)	94.1	87.0	NS
Donor			
Mean age, yr	33.3±7.4	35.9±8.0	NS
Male (%)	47.1	43.5	NS
Blood parent (%)	94.1	86.9	NS
Transplant factor			
PELD score	15.0±7.5	14.3±10.9	NS
Cold ischemia time, min	55.8±14.4	48.6±23.6	NS
Warm ischemia time, min	43.6±9.4	52.5±25.1	NS
AR (%)	33.3	50.0	NS
No. HLA mismatches	2.2±1.0	2.3±1.0	NS
Positive crossmatch (%)	5.9	8.7	NS
Time from LDLT, days	4571.9±544.7	4532.0±425.4	NS

AR, acute rejection; HLA, human leukocyte antigen; LDLT, living-donor liver transplantation; NS, not significant; OT, operational tolerance; PELD, pediatric end-stage liver disease.

properties, and there is growing evidence that OT can be achieved in a proportion of liver transplant recipients (*10–13*) significantly higher than that usually seen in recipients of other types of solid organ transplantation, perhaps as high as 20% (*14*).

Plainly, any factors that might impede the development of OT, especially in pediatric liver transplantation, should be identified so they can be dealt with clinically. Our study found that antibodies specific to human leukocyte antigen (HLA) constitute just such potentially deleterious factors.

We focused on these antibodies because other factors that may impede OT are uncertain; the mechanisms involved in developing and maintaining OT have not been sufficiently elucidated. Several mechanisms have been proposed, including production of donor-strain soluble MHC antigen by the transplanted liver, induction of donor-derived microchimerism by stem cells transferred with the graft, mass effect attributed to passenger leukocytes from the donor, and elevated incidence of circulating regulatory T cells (Tregs). Nevertheless, it is not yet clear why OT occurs, which recipients stand the best chance of developing OT, when it will develop, or what strategies are best to help achieve and monitor OT (*2, 10, 15–23*).

The role of HLA-specific antibodies in liver transplantation remains similarly unclear. Nevertheless, some studies strongly indicate their negative impact on graft survival. Several of these studies retrospectively demonstrated an increased rate of graft loss in patients with preformed HLA-specific antibodies or de novo antibodies developed within the first year after transplantation (*24–27*). Another analysis showed the association between HLA-specific antibodies and early acute rejection (AR) during the first month after liver transplantation (*28*). Recent retrospective studies showed the association between HLA-specific antibodies and chronic rejection (CR) (*29, 30*). In one of these studies, 92% of the patients who experienced CR had detectable HLA-specific antibodies before that CR induced graft loss, whereas only 61% of the non-CR patients had such antibodies (*30*). The same group recently reported a successful treatment of antibody-mediated rejection in liver transplant recipients by using bortezomib (*31*). Finally, it was reported that preformed class I donor-specific HLA antibodies markedly decreased graft survival after liver retransplantation (*32*).

Furthermore, reports have linked OT with the HLA-specific antibodies associated with graft failure (*13, 33*). Patients with high concentrations of circulating HLA-specific antibodies had a higher incidence of steroid-resistant rejection than did patients with low concentrations (*26*). In a retrospective analysis, patients successfully weaned from immunosuppression were negative for HLA-specific antibodies (*33*). Still, no study had examined the possible negative impact on achieving liver allograft OT of the different HLA-specific antibodies (HLA-A, HLA-B, HLA-C, HLA-DR, HLA-DQ, and HLA-DP), nor had any study addressed the significance of mean fluorescence intensity (MFI) combined with these antibodies.

Therefore, we investigated whether, retrospectively, in 40 pediatric living-donor liver transplantation (LDLT) recipients, the presence of HLA-specific antibodies impeded OT development and, conversely, whether the absence of HLA-specific antibodies predicted OT.

RESULTS

Table 1 summarizes the characteristics of both OT and non-OT patients. Most factors were similar between the groups. OT patients were more likely to be female than were non-OT patients (88.2% vs. 47.8%; P=0.008). Although not statistically significant, OT patients had a lower AR rate than non-OT patients (33.3% vs. 50.0%).

Table 2 shows the pretransplantation and posttransplantation HLA-specific antibody profile for OT and non-OT patients. The prevalence of pretransplantation HLA antibodies was high in both groups and slightly higher in non-OT than in OT patients, although the difference was

TABLE 2. Prevalence of HLA-specific antibodies

	OT	Non-OT	P
Pre-LT, n	16	19	
Class I only, n (%)	8 (50.0)	6 (31.6)	0.056
Class II only, n (%)	2 (12.5)	0 (0.0)	
Class I and II, n (%)	2 (12.5)	10 (52.6)	
None	4 (25.0)	3 (15.8)	
Post-LT, n	15	21	
Class I only, n (%)	7 (46.7)	6 (28.6)	<0.001
Class II only, n (%)	4 (26.7)	0 (0.0)	
Class I and II, n (%)	0 (0.0)	14 (66.7)	
None	4 (26.7)	1 (4.8)	

HLA, human leukocyte antigen; LT, liver transplantation; OT, operational tolerance.

not statistically significant. However, after transplantation, a significantly higher percentage of non-OT patients had antibodies: 95.2% versus 73.3% OT patients ($P<0.001$). More than half of the non-OT patients had both class I and II antibodies before transplantation, and two thirds had both classes after transplantation, whereas the percentage of OT patients who had both classes was much lower before transplantation (12.5% vs. 52.6%) and even lower after transplantation (0.0% vs. 66.7%). Almost none of the HLA antibodies detected both before and after transplantation were donor specific. Indeed, only two OT patients had detectable donor-specific antibodies (DSA) before transplantation. Similarly, after transplantation, DSA were detected in no OT patients and in only three non-OT patients.

Because many non-OT patients had more than one anti-HLA antibody (class I or II), we assessed the highest MFI. Figure 1 shows the highest log-transformed MFI for each patient in the OT and non-OT groups, with the values compared. The highest log-transformed MFI of both class I and II antibodies was significantly higher after transplantation in non-OT patients than in OT patients; however, before transplantation, the highest log-transformed MFI did not significantly differ between class I and II antibodies.

Table 3 shows that the prevalence of HLA-B, HLA-C, HLA-DQ, and HLA-DR antibodies was significantly higher in non-OT patients than in OT patients (66.7% vs. 20.0% for HLA-B, $P=0.006$; 76.2% vs. 33.3%, $P=0.01$; 42.9% vs. 6.7%, $P=0.017$; and 57.1% vs. 20.0%, $P=0.026$, respectively). Because HLA antibody MFI appears relevant, the highest log-transformed MFI of each antibody was compared. With HLA-A, it was significantly higher for non-OT patients than in OT patients (7.05±1.09 vs. 6.08±1.03; $P=0.0067$); the same applied to HLA-B (7.31±1.11 vs. 6.15±0.81; $P=0.0014$), HLA-DQ (6.92±1.32 vs. 5.95±0.63; $P=0.0023$), and HLA-DR (22,244.3±76,143.5 vs. 1112.5±3160.4; $P=0.043$).

TABLE 3. HLA antibody and specificities

	OT (n=15)	Non-OT (n=21)	*P*
Post-LT class I, n (%)	7 (46.7)	20 (95.2)	0.001
HLA-A, n (%)	3 (20.0)	9 (42.9)	NS
HLA-B, n (%)	3 (20.0)	14 (66.7)	0.006
HLA-C, n (%)	5 (33.3)	16 (76.2)	0.01
Post-LT class II, n (%)	4 (26.7)	14 (66.7)	0.018
HLA-DQ, n (%)	1 (6.7)	9 (42.9)	0.017
HLA-DP, n (%)	1 (6.7)	1 (4.8)	NS
HLA-DR, n (%)	3 (20.0)	12 (57.1)	0.026

HLA, human leukocyte antigen; LT, liver transplantation; NS, not significant; OT, operational tolerance.

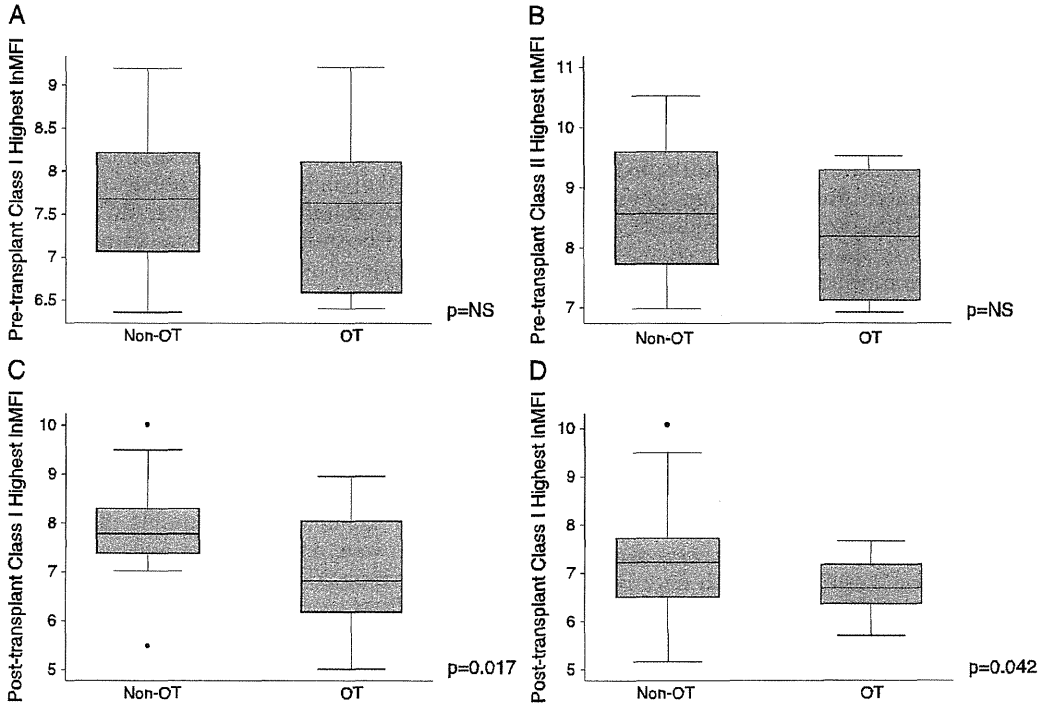

FIGURE 1. Highest log-transformed MFI for each patient in the OT and non-OT groups. A, highest log-transformed MFI HLA antibodies for pretransplant HLA class I antibodies (mean highest log-transformed MFI, OT 7.48±0.87 vs. non-OT 7.70±0.80). B, highest log-transformed MFI for pretransplant HLA class II antibodies (mean highest log-transformed MFI, OT 8.20±1.28 vs. non-OT 8.68±1.28). C, highest log-transformed MFI for posttransplant HLA class I antibodies (mean highest log-transformed MFI, OT 6.98±1.14 vs. non-OT 7.85±0.94). D, highest log-transformed MFI for posttransplant HLA class II antibodies (mean highest log-transformed MFI, OT 6.70±0.55 vs. non-OT 7.43±1.24). MFI, mean fluorescence intensity; OT, operational tolerance.

DISCUSSION

An increasing body of evidence has highlighted the significant impact of HLA antibodies in liver transplantation, but the importance of HLA antibodies of all specificities has not been well described. We believe that this is the first study to examine the association of the presence or absence of early posttransplantation HLA class I and II antibodies with OT development by evaluating and comparing MFI values, especially HLA-B and HLA-DQ antibodies at their highest MFI values.

Because our study involved very young patients, the level of HLA-specific antibodies was much higher than expected (95.2% non-OT and 73.3% OT). These levels were close to those reported in adults (*13, 30*). This phenomenon was likely due to preoperative and perioperative use of blood products and the antigen-antibody reaction the liver graft caused, impelling us to evaluate the differences of the specificities and their MFI values between OT and non-OT patients.

Surprisingly few of our patients had DSA; almost all HLA antibodies were non-DSA. This is partially because preoperative HLA typing for our cohort's donors and recipients was done only for the A, B, and DR loci, so it could not be determined whether HLA-DQ antibodies, present in nearly half our recipients, were DSA or non-DSA. A portion of those HLA-DQ antibodies was likely DSA, which would increase the cohort's overall DSA level. Although the evidence is limited for liver transplantation, previous kidney transplantation studies showed that both DSA and non-DSA were detected and that both were associated with rejection and lower graft survival (*34–36*). This suggests that non-DSA in liver transplantation could be associated with rejection, which would account for the higher prevalence of non-DSA in our non-OT group. In addition, a kidney transplantation study reported that the majority of non-DSA detected in recipients resulted from sharing an epitope with a donor antigen (*37*). This also may be applicable to liver transplantation. Non-DSA resulting from a shared epitope could cause graft rejection, which could impede OT.

Although, in previous studies, AR was one of the factors with negative impact on immunosuppression withdrawal after liver transplantation, we found that AR did not significantly differ between OT and non-OT patients (*38, 39*). This difference may be due to recipient ages and primary liver diseases. One of the previous studies involved adult patients with various primary diseases; our cohort had a mean age of 2.4±2.8 and mostly biliary atresia. The other study showed no difference in gender composition; in our study, approximately 90% of OT patients were female. In another study, a positive T-cell crossmatch negatively impacted graft survival free of AR, although AR, per se, did not significantly influence overall patient or graft survival (*40*). Thus, the effect of recipient age and the underlying liver diseases on the graft's antigenic stimulus may result in different consequences. The relatively small sample size of our study may also explain the insignificant difference in AR between OT and non-OT patients.

The mechanism of OT and rejection (i.e., direct hepatocyte injury or vascular injury mediated by antibody binding to hepatic sinusoidal endothelial cells) has not been elucidated. Although hepatic sinusoidal endothelial cells express both MHC class I and II molecules, hepatocytes and biliary epithelial cells express only MHC class I molecules, so they can act as antigen-presenting cells only for MHC class I-restricted T cells (*32, 41, 42*), nor do those cells express MHC class II molecules constitutively, and MHC class II molecules become up-regulated after inflammation. This parallels the study showing that class I DSA is more detrimental to early graft function, and that preformed persistent and de novo class II DSA [is] are more associated with CR (*30*). Early graft injury caused by class I antibodies may trigger up-regulation of class II molecules after release of proinflammatory cytokines, perhaps resulting in chronic damage to the liver graft and consequent absence of OT.

Our cohort's unexpectedly high prevalence of HLA antibodies, together with a previous report, make it unsurprising to find HLA antibodies in patients with well functioning and tolerated grafts (*30*). However, we found the significantly highest MFI values for each antibody in non-OT patients, and only when those MFI values reached high titers were they associated with liver damage, although our study's 1000 MFI cutoff may not be ideal for considering antibodies in OT positive or negative. We cannot rule out possible progression of pathologic changes, which could not be detected by liver chemistry test. Further studies should clarify the association between MFI values and OT.

Our results suggest that HLA-B and HLA-DQ antibodies may impede OT more than HLA-A, HLA-C, HLA-DP, and HLA-DR. Although data are limited, recent kidney transplantation studies showed that HLA-DQ antibodies were the most common type detected after kidney transplantation and may contribute to inferior survival (*43, 44*). The detrimental effects of HLA-DQ antibodies are not limited to kidney transplantation. Class II antibodies (especially HLA-DQ) are reportedly associated with graft failure in cardiac, lung, and liver transplantation (*45–47*). Further studies are needed to understand the impact of HLA-DQ antibodies on OT; for example, one such study would be HLA typing for HLA-DQ antibodies and association between them and the presence of other HLA antibodies such as HLA-B and-DR in relation to OT.

Other factors in addition to HLA antibodies are also involved in achieving tolerance. Non-HLA antibodies, such as anti-angiotensin type 1 receptor antibodies and other autoantibodies, might help induce graft rejection. Recently, several autoantibodies were reported to be associated with graft dysfunction (*48, 49*). Furthermore, specific antibody characteristics other than MFI threshold may affect tolerance. For example, donor-specific HLA antibodies of IgG subclasses were reported to be associated with CR and graft loss after liver OT (*50*). Other immunomodulatory factors, such as Tregs specific to donor antigens, may have played a critical role in the induction and maintenance of OT in our cohort. A possible association between HLA-A matches and the predominance of Tregs after liver OT has been reported (*39*), with possible linkage between HLA antibodies and Tregs suggested. They may, together, influence OT.

Our study's limitations stem from its retrospective nature. Because HLA typing of Cw and DG loci were unavailable for the cohort, DSA analysis was not fully performed. The effects on tolerance and rejection of the timing and duration of exposure to DSA and non-DSA cannot be evaluated. The frequent collection of sera samples is

necessary to determine the precise time of antibody exposure; some antibodies could be transient with no definite impact on tolerance, particularly OT, which may be dynamic (*13*). For example, we examined the association between early posttransplantation HLA antibodies and OT; however, later posttransplantation HLA antibodies (e.g., during immunosuppression weaning and when graft function stabilized with low-dose immunosuppression) along with early posttransplantation HLA antibodies may be more meaningful in determining the association between HLA antibodies and OT. Furthermore, we had to depend on liver chemistry tests—with no protocol liver biopsy (PLB) for 10 patients. The relevant Banff Working Group strongly recommends PLB of patients for whom immunosuppression withdrawal is planned and for patients under and after weaning (*51*). Our cohort had completed weaning before the recommendation was published so misclassification of OT and non-OT could have occurred in our study. Finally, we could not evaluate HLA antibodies by graft function at 3 weeks after transplantation due to the small sample size of the study. At the time of HLA antibody measurement, nine patients had developed AR, which could affect the presence of HLA antibodies and the development of OT.

In summary, in our study, posttransplantation HLA class I and II antibodies were associated with the future absence of OT. Specifically, the level of HLA-B and HLA-DQ antibodies was lower in OT patients than in non-OT patients. We also found that the highest MFI of antibodies in OT patients was significantly lower than in non-OT patients. Further study is needed to confirm our findings that anti-HLA antibodies are associated with impeding OT and to identify threshold levels and characteristics of HLA antibody specificities in relation to the development or absence of OT.

MATERIALS AND METHODS

Study Population

We retrospectively studied 52 pediatric LDLT recipients at the University of Tokyo Hospital between April 1996 and December 2000, followed up through July 2011 at the Jichi Medical University Hospital, where they underwent immunosuppression withdrawal (*52*). One graft failed during this period. Sera of 40 (17 OT and 23 non-OT) of the 51 (78.4%) with functioning grafts were available for the measurement of anti-HLA antibodies; for 31 patients, sera taken before and after transplantation (at 3 weeks) were also available. Nine patients (2 OT and 7 non-OT) had biopsy-proven AR when the posttransplantation sera were stored. The university's research ethics committee approved this study.

Immunosuppression and Weaning Protocol

Tacrolimus and methylprednisolone were used (*52*). The target trough serum tacrolimus level was 15 to 20 ng/mL on the first week after transplantation and gradually decreased 6 months after transplantation to 8 to 5 ng/mL. Methylprednisolone (20 mg/kg) was given before the anhepatic phase of LDLT, the dose subsequently reduced to the maintenance level. Cyclosporine replaced tacrolimus in patients who suffered tacrolimus side effects. When AR developed, regardless of severity, patients were treated with bolus intravenous methylprednisolone, the starting dose 20 mg/kg per day, as described previously (*53*). With a parent's consent, patients were administered the weaning protocol regardless of their primary liver disease when they met the following criteria: being more than 2 years past liver transplantation; normal graft function and no episodes of rejection for more than 1 year; taking only tacrolimus for immunosuppression, dose less than 0.05 mg/kg per day (*52*); and having no autoantibodies. (Patients taking cyclosporine, with a dose less than 1.0 mg/kg per day, are considered for weaning, including a few in our cohort.) Liver chemistry test results were considered normal when serum alanine aminotransferase, γ-glutamyl transpeptidase, and direct bilirubin were all within normal ranges. OT was defined as stable normal graft function for more than 1 year off immunosuppression.

Protocol Liver Biopsy

Percutaneous transhepatic liver biopsy was performed under analgesia and sedation using ultrasonographically guided 14G Monopty (C.R. Bard, Murray Hill, NJ). Manual compressive hemostasis was performed for 20 min, with compressive bandage hemostasis until the following day. Preventive cefoperazone and sulbactam were administered. We assessed the histopathologic features of the PLB samples using the Metavir scoring system, which grades activity, that is, the amount of inflammation (specifically, the intensity of necroinflammatory lesions), on a four-point scale from A0 to A3 (*54*). Fibrosis is graded on a five-point scale, from 0 to 4. We defined abnormal biopsy histology as more than A2 or more than F2. Fourteen of 17 OT patients and 16 of 23 non-OT patients received PLB.

Human Leukocyte Antigen Typing

The pretransplantation HLA typing for A, B, and DR loci was performed routinely for all donors and recipients. HLA-A and HLA-B typing was performed by standard complement-dependent microcytotoxicity assay using a Terasaki HLA tray (One Lambda, Canoga Park, CA). HLA-DR typing was performed by two-color fluorescence. All typing for our cohort was performed serologically.

Lymphocytotoxic Crossmatch

Lymphocytotoxic crossmatch testing followed standard National Institutes of Health technique for all donors and recipients. The recipient serum obtained immediately before LDLT was tested for cytotoxic antibodies against donor T or B lymphocytes. Donor lymphocytes were isolated from peripheral blood, and 1 μL of the patient's serum was added for 30 min at room temperature. Rabbit complement (5 μL) was added for an additional hour at room temperature, and ethidium bromide and acridine orange were added to stain the cells. The crossmatch was considered positive when more than 20% of the donor lymphocytes were killed by the recipient serum.

Detection of Anti-Human Leukocyte Antigen Antibodies and Determination of Donor-Specific Antibody Specificity

The samples stored before and after transplantation were sent to the Terasaki Foundation Laboratory for evaluation. Sera were screened using LABScreen mixed beads (One Lambda). Sera with a positive screen result had the specificity of their anti-HLA antibody identified using LabScreen single antigen class I (Lot 6) and class II (Lot 8) beads. Assays followed the manufacturer's protocol. Trimmed mean values of MFI (i.e., normalized MFI) were obtained from the output file generated by the flow analyzer, normalized using the formula: ([sample #N bead]-[sample negative control bead])-([negative control serum #N bead]-[negative control serum negative control bead]), with normalized values over 1000 MFI considered positive. To identify DSA specificity, donor-recipient mismatched HLAs were compared with the antibody profile for each patient's sample.

Statistical Analysis

Patient characteristics were compared using the chi-square test for categorical variables and Wilcoxon rank-sum tests for continuous variables. Each MFI was analyzed after log transformation because of their nonnormal distribution. We used Stata version 10.0 (Stata, College Station, TX) for all statistical analyses. Data were expressed as mean±standard deviation. P=0.05 was considered statistically significant.

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Original Article

Role of 6-month abstinence rule in living donor liver transplantation for patients with alcoholic liver disease

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Aim: Although alcoholic liver disease (ALD) is an accepted indication for liver transplantation (LT), there are several controversial issues. The aim of this study is to examine the applicability of the 6-month abstinence rule prior to LT and to evaluate the results in living donor LT for patients with ALD.

Methods: A retrospective study of 102 patients with ALD referred for LT was performed. Clinical data, including alcohol consumption history, were analyzed. A period of abstinence from drinking alcohol of at least 6 months was strictly required.

Results: Among 102 patients, 21 abstained from drinking for at least 6 months. Of these, 13 patients (12%) underwent LT, five patients (5%) recovered without LT and three patients (3%) were listed for deceased donor LT. LT was not indicated for the remaining 81 patients (80%). Eight patients died within 6 months of referral to our program. The Child–Pugh score was higher in these eight patients than in the 21 who achieved 6-month abstinence, although the alcohol consumption history variables did not significantly differ between the two groups. The 5-year overall survival rates after LT in 13 patients with ALD (91%) were similar to those in 387 non-ALD patients (83%). The rate of alcohol consumption relapse after LT was 8% (n = 1/13).

Conclusion: Living donor LT for patients with ALD who complied with the 6-month abstinence rule provides sufficient survival benefit with good compliance, compensating for the potential risks to the donors.

Key words: abstinence period, alcohol recidivism, alcoholic liver disease, liver transplantation

INTRODUCTION

ALCOHOLIC LIVER DISEASE (ALD) is an increasingly important cause of end-stage liver disease, and a recognized indication for liver transplantation (LT), accounting for approximately 2% of all primary transplants in Japan,[1] 40% in Europe[2] and 20% in the USA.[3] The proportion of ALD patients undergoing LT remains small in Japan compared to the latter two regions, but the number of ALD patients who underwent living donor LT (LDLT) in Japan is increasing annually based on a report by the Japanese Liver Transplantation Society.[1] A fair therapeutic strategy is necessary before considering patients with ALD for LDLT, because deceased donor organs remain scarce in East Asian regions, including Japan.

The outcome of the long-term prognosis of patients transplanted for ALD is at least as good as that of patients transplanted for most other diagnoses.[2,4–6] Although post-LT drinking impairs the long-term survival of ALD patients after LT,[7] late graft loss due to recurrence of the original disease, such as viral hepatitis and cholestatic disease, is uncommon. A fixed period of abstinence from drinking alcohol prior to transplantation allows some patients to recover their liver function to the extent that LT is no longer needed and should be adopted as inclusion criteria for LT.[8] There have been no studies in the published work focusing on the treatment of ALD, including LT and patient outcome, in regions in which deceased donor organs are scarce.

In the present study, we performed a retrospective analysis of ALD patients to examine the applicability of

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the 6-month abstinence rule prior to LT and to evaluate the results of LDLT for patients with ALD.

METHODS

Patients

BETWEEN JANUARY 1996 and September 2011, 102 patients with chronic ALD or alcoholic liver cirrhosis were referred to the University of Tokyo Hospital for LT; patients presenting with severe alcoholic hepatitis were not included. The diagnosis of ALD was based on a history of habitual and excessive alcohol consumption in the absence of other causes of liver cirrhosis. The clinical records of these patients were retrospectively reviewed. A history of alcohol intake was also obtained from the clinical records, including duration of heavy drinking, types and amount of alcohol consumed, and previous treatment history. A high-risk alcoholism relapse score was calculated according to Yates *et al.*[9]

Indication criteria of LT for ALD

The selection criteria for LT at our institution are described elsewhere.[10] In addition to our general criteria, patients with ALD are required to fulfill additional criteria as follows: period of abstinence from drinking alcohol of at least 6 months prior to LT; participation in Alcoholics Anonymous or an equivalent rehabilitation program; consultation with a psychiatrist; and signed agreement indicating intention of lifetime abstinence. ALD patients meeting our criteria were considered candidates for LDLT or deceased donor LT (DDLT), irrespective of a high-risk alcoholism relapse score. The indications for LDLT and the type of liver graft were determined according to the ratio of the remnant liver volume to total liver volume in living donors, and that of the graft volume to the standard liver volume[11] in recipients.[12]

Surgical treatment and management

Our LT procedure has been described elsewhere.[13] For the follow-up evaluation, blood test and ultrasonography findings were examined at every outpatient clinic (usually every 1–2 weeks) beginning immediately after the patients were discharged. Alcohol relapse after LT was defined as re-drinking on the basis of self-report questionnaires and interviews with patients and/or family members.

Statistical analysis

Continuous data are expressed as the median values (with range). Quantitative and categorized variables were compared using the Wilcoxon rank sum test and Fisher's exact test, respectively. Long-term survival was measured from the time at which patients underwent LT. Overall survival curves were constructed using the Kaplan–Meier method, and compared using the log-rank test. A *P*-value of less than 0.05 was considered to indicate statistical significance. Statistical analysis was performed with JMP software ver. 9.0.2 (SAS Institute, Cary, NC, USA).

RESULTS

THE FLOW OF study participants is shown in Figure 1. Among 102 patients, 13 (12%) underwent LDLT, patients (5%) were recognized as recovering from liver failure and three patients (3%) were listed for DDLT after an abstinence period lasting at least 6 months. LT was not indicated for 81 patients (80%) and eight of these (8% of total) died within 6 months of referral to our program. The reasons for rejection are shown in Table 1. Fifty-five patients (68%) were rejected for reasons related to recipient issues, including not abstaining from drinking alcohol in 15 patients (21%).

Demographic data of 21 patients who achieved 6 months of abstinence ("abstinence group") are shown in Table 2 and compared with the eight patients who died within 6 months ("mortality group"). The Child–Pugh score was significantly higher in the Mortality group than in the abstinence group (median [range], 12

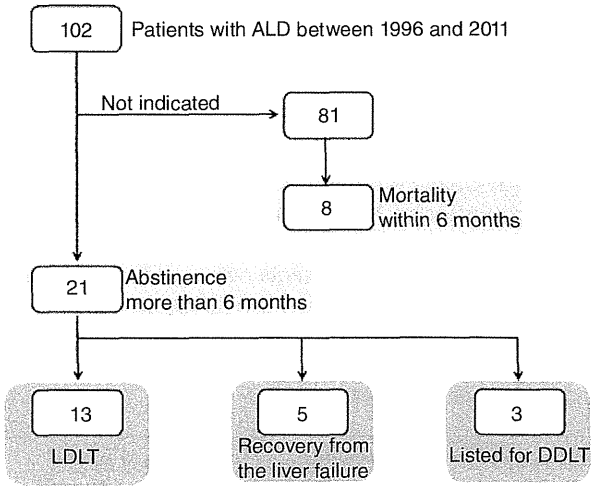

Figure 1 Flow of study participants. ALD, alcoholic liver disease; DDLT, deceased donor living transplantation; LDLT, living donor liver transplantation.