ANNALS OF TRANSPLANTATION

ORIGINAL PAPER

e-ISSN 2329-0358
© Ann Transplant, 2026; 31: e952021
DOI: 10.12659/AOT.952021

Received: 2025.11.06
Accepted: 2026.04.01
Available online: 2026.05.02
Published: 2026.XX.XX

Immunosuppression Withdrawal in Pediatric Liver Transplant Recipients With Posttransplant Lymphoproliferative Disorder: A Single-Center Retrospective Study

Authors' Contribution:
Study Design A
Data Collection B
Statistical Analysis C
Data Interpretation D
Manuscript Preparation E
Literature Search F
Funds Collection G

ABCDEFG 1-5 Ying Liu*
ABCDEF 2-6 Ru-Zhou Cai*
ACDE 2-6 Wei Qu
ADE 2-6 Lin Wei
ADE 2-6 Zhi-Gui Zeng
ACDE 2-6 Zhi-Jun Zhu
ABCDEFG 2-5,7 Li-Ying Sun

1 International Medical Center, National Clinical Research Center for Digestive Diseases, Beijing Friendship Hospital, Capital Medical University, Beijing, PR China
2 Beijing Key Laboratory of Organ Cultivation and Organ Protection in Transplantation, Beijing Friendship Hospital, Capital Medical University, Beijing, PR China
3 Clinical Center for Pediatric Liver Transplantation, Capital Medical University, Beijing, PR China
4 State Key Laboratory of Digestive Health, Beijing, PR China
5 Laboratory for Clinical Medicine, Capital Medical University, Beijing, PR China
6 Liver Transplantation Center, National Clinical Research Center for Digestive Diseases, Beijing Friendship Hospital, Capital Medical University, Beijing, PR China
7 Department of Critical Liver Disease, National Clinical Research Center for Digestive Diseases, Beijing Friendship Hospital, Capital Medical University, Beijing, PR China

* Ying Liu and Ru-Zhou Cai contributed equally to this manuscript

Corresponding Author: Li-Ying Sun, e-mail: sunxlx@outlook.com

Financial support: The study was funded by the Chronic Disease Management Research Project of National Health Commission Capacity Building and Continuing Education Center (No. GWJJMB202510025098)

Conflict of interest: None declared

Background: Posttransplant lymphoproliferative disorder (PTLD) is a serious complication following pediatric liver transplantation. Reduction of immunosuppression is a cornerstone of PTLD management; however, the feasibility and safety of complete immunosuppression withdrawal (ISW) in this setting remain unclear.

Material/Methods: We retrospectively reviewed 6 pediatric liver transplant recipients diagnosed with PTLD who subsequently underwent complete ISW at our center between 2013 and 2019. Demographic characteristics, clinical features, pathological classification, treatments, and follow-up outcomes were analyzed.

Results: The cohort included 6 children (4 females and 2 males) who underwent liver transplantation at a median age of 8 months. PTLD subtypes included infectious mononucleosis-type (n=3), polymorphic PTLD (n=1), Burkitt lymphoma (n=1), and classical Hodgkin lymphoma–like PTLD (n=1). All patients achieved complete remission following multimodal therapy. The median interval from transplantation to initiation of ISW was 35 months. During a median follow-up of 52 months after ISW, 4 patients maintained stable graft function without biopsy-proven rejection, whereas 2 developed rejection-related complications, both of which resolved after restart of low-dose immunosuppressive therapy. No graft loss or PTLD recurrence occurred.

Conclusions: In carefully selected pediatric liver transplant recipients with PTLD, supervised ISW may be achieved without irreversible graft injury. However, a substantial risk of rejection remains, highlighting the importance of close clinical and histological monitoring.

Keywords: Posttransplant Lymphoproliferative Disorder • Immunosuppression • Liver Transplantation • Pediatrics • Retrospective Studies • Transplant Recipients

Full-text PDF: https://www.annalsoftransplantation.com/abstract/index/idArt/952021

Indexed in: [Science Citation Index Expanded] [Index Medicus/MEDLINE] [Chemical Abstracts] [Scopus]

Introduction

Posttransplant lymphoproliferative disorder (PTLD) represents a heterogeneous group of lymphoid proliferations that arise in the setting of immunosuppression after solid organ transplantation. PTLD is most commonly associated with Epstein-Barr virus (EBV)-driven proliferation of B lymphocytes and encompasses a spectrum ranging from benign polyclonal proliferations to aggressive lymphomas. Because PTLD can progress rapidly and is associated with significant morbidity and mortality, it is considered one of the most serious complications following solid organ transplantation.

The reported incidence of PTLD in pediatric liver transplant recipients ranges from approximately 5% to 15% [1-3]. Standard PTLD management typically includes reduction of immunosuppression, administration of the anti-CD20 monoclonal antibody rituximab, chemotherapy, surgical intervention, and radiotherapy when indicated [4-6]. T-cell-mediated immune surveillance plays a central role in controlling EBV-infected B cells. Immunosuppressive therapy impairs this surveillance, thereby predisposing transplant recipients to uncontrolled lymphoid proliferation and the development of PTLD [7,8].

The immunologic characteristics of the liver differ from those of other transplanted organs, and operational tolerance has been reported more frequently after liver transplantation than after kidney, heart, or lung transplantation. In selected long-term stable liver transplant recipients, complete immunosuppression withdrawal (ISW) has been successfully achieved [9, 10]. Factors favoring successful withdrawal include long-term stable graft function, absence of prior rejection, non-autoimmune primary liver disease, and pediatric recipient status. Previous studies have suggested that operational tolerance is achieved in approximately 20% to 40% of carefully selected pediatric liver transplant recipients, whereas the success rate appears to be lower in adult recipients [11]. However, the safety and long-term outcomes of complete ISW in pediatric patients with PTLD remain poorly characterized.

Material and Methods

Study Population and Data Collection

We retrospectively identified pediatric liver transplant recipients who subsequently underwent supervised complete ISW after being diagnosed with PTLD at Beijing Friendship Hospital between June 2013 and December 2019. Clinical, laboratory, and histopathologic data were extracted from medical records. Information regarding primary transplant diagnoses, sex, age at liver transplantation, donor types, donor-recipient ABO type, timing of PTLD onset, treatments received, and follow-up outcomes was collected. This study was approved by the Ethical Committee of Beijing Friendship Hospital after rigorous ethical review (No. 2022-P2-191), and was conducted in accordance with the Declaration of Helsinki. Written informed consent was obtained from the parents or legal guardians of all patients.

Immunosuppression Regimens and Withdrawal Protocol

All patients received a standard immunosuppressive regimen after liver transplantation. Induction therapy consisted of intravenous methylprednisolone, followed by maintenance therapy with tacrolimus and corticosteroids. Corticosteroids were tapered and discontinued within 3 months after transplantation in all cases. Following the diagnosis of PTLD, immunosuppressive therapy was initially reduced as part of standard PTLD management. During this period, patients underwent close monitoring of liver function and PTLD disease activity to ensure sustained remission.

Complete ISW was considered in carefully selected PTLD recipients. Recipients were considered eligible for complete ISW when the following criteria were met: (1) stable liver function after immunosuppression reduction; (2) absence of histological evidence of rejection on liver biopsy; and (3) persistent risk of PTLD recurrence requiring minimization of immunosuppressive exposure, such as high Epstein-Barr virus (EBV) replication. Patients with primary autoimmune liver diseases were generally not considered suitable candidates for complete ISW because of their higher immunological risk. The final decision to proceed with complete ISW was made after multidisciplinary team discussion involving transplant surgeons, hepatologists, pathologists, and oncologists. The time from immunosuppression adjustment to complete ISW was defined as the interval between initiation of immunosuppression tapering and complete discontinuation of all immunosuppressive agents.

During the period of immunosuppression tapering, liver function tests were monitored monthly. After complete ISW, patients underwent protocol liver biopsy at approximately 1 year after ISW. If no evidence of rejection was detected, follow-up intervals were gradually extended from every 2 to 3 months to every 6 months. Protocol liver biopsies were generally performed every 1 to 2 years, and additional biopsies were obtained whenever abnormal liver function tests or clinical suspicion of rejection arose.

PTLD Diagnosis and Classification

All PTLD diagnoses were confirmed by histopathological examination of tissue biopsy specimens. Histopathologic classification was assigned according to the 2016 revision of the World Health Organization classification of lymphoid neoplasms [12].

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Table 1. Baseline characteristics of the patients.

Case	Sex	Age at LT (months)	Primary disease	Donor type	Donor age (years)	ABO blood type (recipient/donor)	Time from transplantation to PTLD (months)	IS regimen at PTLD onset	Tacrolimus trough level (μg/L)
1	Female	8	BA	Living donor	29	B/B	32	Tacrolimus 0.625 mg q12h	4.3
2	Female	7	BA	Living donor	36	B/O	11	Tacrolimus 0.25 mg q12h	7.2
3	Female	5	BA	Living donor	31	A/A	9	Tacrolimus 0.5 mg q12h	7.1
4	Male	14	BA	DCD	1	A/A	27	Tacrolimus 1 mg q12h	1.3
5	Male	19	Cryptogenic cirrhosis	Living donor	32	AB/AB	11	Tacrolimus 0.375 mg q12h	1.1
6	Female	5	BA	Living donor	39	A/A	13	Tacrolimus 0.75 mg q12h	1.1

BA – biliary atresia; DCD – donation-after-circulatory-death; IS – immunosuppression; LT – liver transplantation; PTLD – post-transplant lymphoproliferative disorder.

Two experienced pathologists independently reviewed all biopsy specimens to confirm the diagnosis and subtype.

Statistical Analysis

Statistical analyses were performed using SPSS version 27.0. Given the small sample size and the descriptive nature of this study, only descriptive statistics were applied. Continuous variables are presented as median (range), and categorical variables are presented as numbers and percentages. No formal statistical comparisons were performed.

Results

Patient Characteristics

Between June 2013 and December 2019, a total of 38 pediatric liver transplant recipients were diagnosed with PTLD at Beijing Friendship Hospital, including patients who had undergone liver transplantation at other institutions. Among these patients, 6 patients (4 females and 2 males) subsequently underwent complete ISW. The median age at liver transplantation was 8 months (range 5-19 months). Primary liver disease was biliary atresia (n=5) and cryptogenic cirrhosis (n=1). Five patients received living donor liver transplants, and 1 received a donation-after-circulatory-death graft. All donor-recipient pairs in this cohort were ABO compatible. The median time from liver transplantation to PTLD onset was 12 months (range 9-32 months). None of the patients had a history of acute rejection prior to PTLD. At the time of PTLD diagnosis, all patients were on tacrolimus monotherapy; trough tacrolimus levels ranged from 1.1 to 7.2 μg/L (**Table 1**).

Diagnosis and Treatments of PTLD Patients

The 6 patients presented with clinical features including fever, lymphadenopathy, anemia, hypoproteinemia, hepatosplenomegaly, and various gastrointestinal manifestations (eg, abdominal pain, diarrhea, ascites, or intestinal perforation) (**Table 2**). Histologic classification of PTLD was infectious mononucleosis-type in 3 patients, polymorphic in 1 patient, Burkitt lymphoma in 1 patient, and classical Hodgkin lymphoma–like PTLD in 1 patient.

All patients underwent reduction or discontinuation of immunosuppression as part of PTLD therapy. Four patients (3 with infectious mononucleosis-type PTLD and 1 with polymorphic PTLD) received rituximab (anti-CD20) therapy. One of these patients (case 6) required a partial small-intestinal resection because of intestinal perforation. The patient with Burkitt lymphoma (case 1) was treated with R-CHOP chemotherapy (rituximab, cyclophosphamide, doxorubicin, vincristine, prednisone). The patient with Hodgkin-like PTLD (case 4) was initially treated with 2 cycles of ABVD chemotherapy (adriamycin, bleomycin, vinblastine, dacarbazine). Because this regimen failed to achieve remission, therapy was escalated to a more intensive multi-agent regimen (including cytarabine, etoposide, cyclophosphamide, doxorubicin, vincristine, procarbazine, and prednisone) combined with radiotherapy. All 6 patients ultimately achieved complete remission following these treatments.

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Table 2. Clinical features, immunosuppression management, and graft outcomes of patients with PTLD.

Case	Clinical features	Histological classification	IS adjustment	Anti-PTLD therapy	PTLD outcome
1	Fever, distension, ascites	Burkitt lymphoma	Tacrolimus tapered	R-CHOP, 4 cycles	CR
2	Fever, lymphadenopathy	Infectious mononucleosis PTLD	Methylprednisolone 8 mg/day, tapered	Rituximab, 2 cycles	CR
3	Fever, splenomegaly, anemia, liver dysfunction, multiple enlarged mesenteric lymph nodes	Polymorphic PTLD	Methylprednisolone 4 mg/day, tapered	Rituximab, 1 cycle	CR
4	Progressive enlargement of cervical lymph nodes	Classical Hodgkin lymphoma–like PTLD	Complete ISW	ABVD, 2 cycles; Cycle A: cytosine arabinoside, etoposide; COPP/ABV; Cycle C: cyclophosphamide, doxorubicin, vincristine and prednisone; radiation	CR
5	Enlarged cervical lymph nodes	Infectious mononucleosis PTLD	Methylprednisolone 6 mg/day, tapered	Rituximab, 1 cycle	CR
6	Fever, anemia, distention, intestinal perforation	Infectious mononucleosis PTLD	Methylprednisolone 4 mg/day, tapered	Partial small-intestinal resection; rituximab, 4 cycles	CR

Case	Time from IS adjustment to complete ISW (months)	Time between LT and complete ISW (months)	Duration of ISW (months)	Graft dysfunction (yes/no)	Liver biopsy	IS restart	Liver function of the last follow-up		
							ALT/AST (IU/L)	TB/DB (μmol/L)	ALP/GGT (IU/L)
1	6	38	39	No	Borderline rejection	Tacrolimus 0.5 mg/day	27/38	18/7	183/37
2	27	38	48	No	No ejection	-	15/25	6.1/3.3	175/10
3	29	38	56	No	No ejection	-	19/38	13.3/3.8	267/13
4	0	27	60	No	No ejection	-	28/42	11.7/2.6	228/13
5	22	33	36	No	No ejection	-	25/31	18.8/7.3	161/27
6	6	19	4	Yes	Mild rejection	Methylprednisolone 4 mg/day	34/47	6.7/2.5	176/54

IS – immunosuppression; ISW – immunosuppression withdrawal; ABVD – adriamycin, bleomycin, vinblastine and dacarbazine; COPP/ABV – cyclophosphamide, vincristine, procarbazine, prednisone, adriamycin, bleomycin and vinblastine; CR – complete remission; R-CHOP – rituximab, cyclophosphamide, doxorubicin, vincristine and prednisone; PTLD – post-transplant lymphoproliferative disorder.

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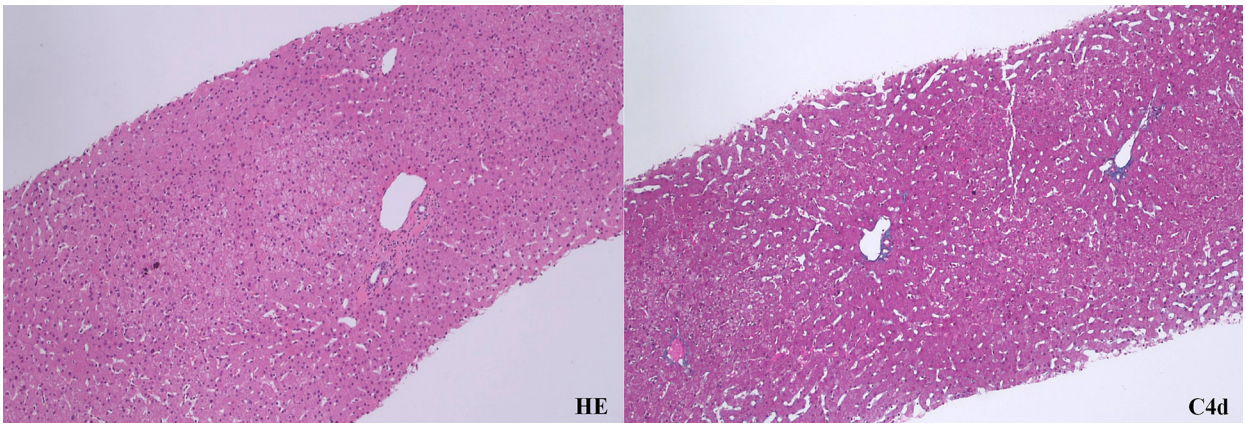

Figure 1. Liver biopsy in case 3 shows no evidence of rejection or graft fibrosis at 1 and 2 years after immunosuppression withdrawal.

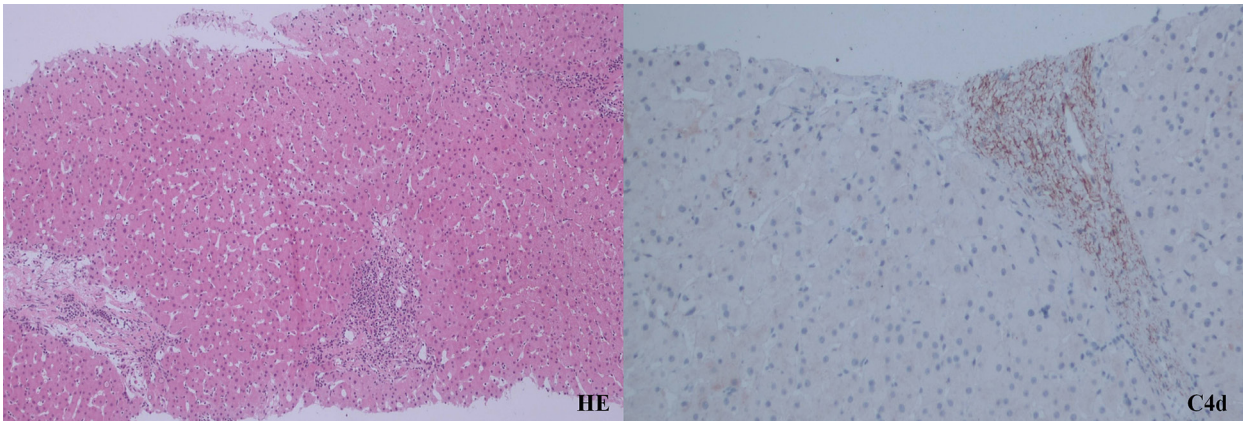

Figure 2. Liver biopsy in case 1 at 3 years after immunosuppression withdrawal shows a borderline rejection with stromal C4d positivity.

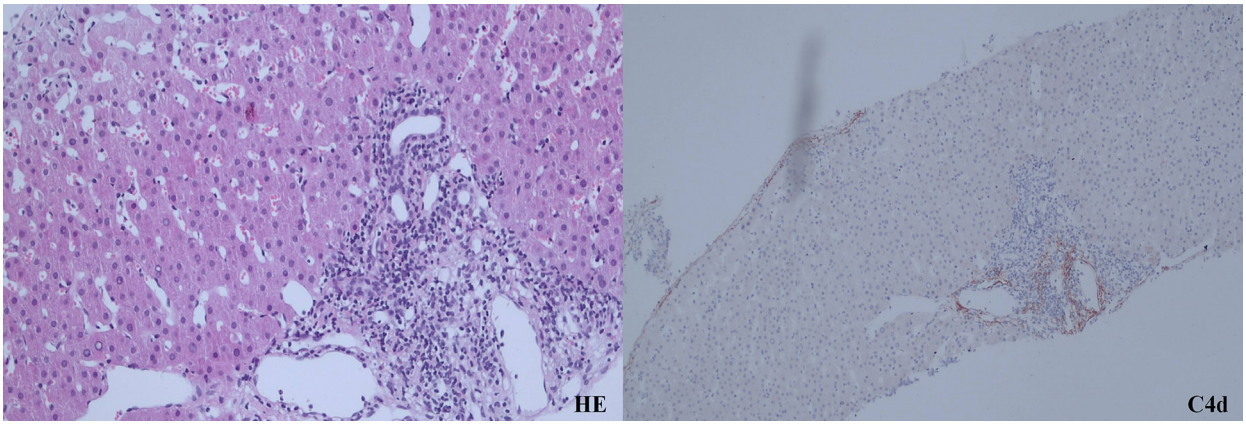

Figure 3. Liver biopsy shows mild rejection (Banff score=4) and C4d positivity in case 6 at 4 months after immunosuppression withdrawal.

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ISW and Graft Outcomes

The median interval from liver transplantation to complete ISW was 35 months (range 19–38). The duration from initiation of immunosuppression adjustment to complete ISW varied substantially among patients, ranging from 0 to 29 months (**Table 2**). Throughout the withdrawal process and during follow-up, graft status was closely monitored using serial liver function tests and protocol liver biopsies. During a median follow-up of 52 months after ISW, 4 of 6 patients maintained stable liver function without evidence of rejection (**Figure 1**).

Two patients developed rejection-related complications. In 1 patient (case 1), a protocol biopsy at 3 years after ISW revealed subclinical (borderline) rejection, characterized by mild portal inflammation and stromal C4d positivity (**Figure 2**). This patient was treated with low-dose tacrolimus (0.5 mg/day), after which graft function remained stable.

Another patient (case 6) developed biochemical graft dysfunction at 4 months after ISW. Liver biopsy demonstrated mild acute cellular rejection (Banff score 4) with stromal C4d positivity (**Figure 3**). Following reintroduction of immunosuppression with methylprednisolone (4 mg/day), liver enzyme levels normalized and graft function recovered. A repeat liver biopsy performed more than 1 year later revealed no evidence of rejection. Notably, no patient experienced graft loss, and no recurrence of PTLD was observed during follow-up.

Discussion

In this study, we found that complete withdrawal of maintenance immunosuppression was feasible in a carefully selected cohort of pediatric liver transplant recipients with PTLD, without irreversible graft injury in most cases. However, reversible rejection occurred in 2 patients, indicating that ISW carries a substantial risk of immune-mediated graft injury. These findings underscore the importance of rigorous biochemical and histological monitoring to allow early detection and timely treatment of rejection during and after ISW.

Our observations align with prior reports that only a minority of pediatric liver transplant recipients can achieve operational tolerance [10]. The liver is considered a relatively tolerogenic organ, and operational tolerance has been reported more frequently after liver transplantation than after other solid organ transplants. Previous studies have identified factors associated with successful ISW, such as young recipient age, living donor grafts, a prolonged posttransplant interval before withdrawal, and no history of rejection[9]. The favorable profile of our cohort across these parameters likely contributed to the relatively high success rate in achieving stable graft function. Unlike many ISW series in unselected recipients, our cohort consisted entirely of patients with PTLD, adding a unique context to these established favorable factors.

PTLD is recognized as a potentially life-threatening complication after liver transplantation. Calcineurin inhibitors (CNIs), such as tacrolimus, are known risk factors for PTLD development [8]. Adjusting immunosuppressive therapy, including reducing or discontinuing CNIs, is a standard component of PTLD management. In our study, all patients underwent immunosuppression reduction as part of their initial PTLD treatment. This strategy not only addressed the malignancy but also set the stage for eventual tolerance, as reducing CNI exposure may lessen oncogenic stimuli and promote immune regulation over time.

Operational tolerance occurs more frequently in liver transplantation than in other solid organ transplants, particularly in pediatric recipients. Predictive factors include younger patient age at transplant and the use of living-related donor grafts [13]. Our cohort, predominantly consisting of living donor grafts, may have benefited from the associated genetic similarity or favorable immunologic matching, which could promote tolerance. The interval between transplant and ISW initiation in our patients was substantial, consistent with the notion that longer engraftment time before withdrawal favors tolerance [11].

Histologic features may also inform graft tolerance status. Previous studies have shown that even mild portal inflammation or C4d deposition in biopsies can predict loss of tolerance [14]. In our cohort, surveillance biopsies from 2 patients revealed mild portal inflammation with stromal C4d positivity during the withdrawal period, suggesting subclinical immune activation despite overall graft stability. The dynamic nature of tolerance is illustrated by the patient in case 1 who had normal histology at 1 year after ISW but developed borderline rejection by year 3. This observation emphasizes that operational tolerance may not be permanent and reinforces the value of periodic biopsies even in the absence of laboratory abnormalities, as tolerance can be reversible [15]. Early histologic detection in our patients allowed prompt intervention before significant injury occurred, leading to full recovery of graft function. These experiences highlight that the potential benefit of ISW must be weighed against the necessity of being able to promptly reverse graft injury. Importantly, previous studies have demonstrated that grafts in operationally tolerant liver transplant recipients may develop progressive fibrosis despite normal liver biochemical parameters. In this context, protocol liver biopsy plays a critical role in the long-term surveillance of graft integrity, allowing early detection of fibrosis or immune-mediated injury even in clinically stable patients.

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Efforts to identify biomarkers of tolerance are ongoing. While certain peripheral blood gene expression signatures have been linked to tolerant states, no universally reliable biomarker currently exists [16,17]. All patients in our cohort had EBV-positive PTLD and most received rituximab. It is conceivable that B-cell depletion induced by rituximab contributed to an immunologic environment favoring graft tolerance, although this hypothesis requires further investigation [18]. Rituximab has been reported to potentially promote immune tolerance in other types of transplant patients, although the evidence remains limited and requires further validation through additional studies [19,20].

Importantly, operational tolerance should not be considered a permanent state, as immune activation can recur over time. Therefore, continued surveillance remains essential even in patients who successfully discontinue immunosuppression.

This study has several limitations. It is a retrospective review of a small, heterogeneous cohort from a single center, which limits generalizability. Because some transplants were performed during an earlier period, donor and recipient EBV serology data were not systematically available, and only EBV DNA viral load monitoring had been routinely performed. Moreover, the PTLD histologies and treatments varied, and we did not perform mechanistic immunologic studies. There is an inherent selection bias, as only patients judged to have very low rejection risk were offered ISW. Despite these limitations, our findings provide insight into the potential benefits and pitfalls of ISW in pediatric PTLD. Prospective multicenter studies are needed to better define which patients with PTLD may safely undergo ISW, optimize monitoring protocols, and identify noninvasive biomarkers of tolerance.

Conclusions

PTLD is a serious complication following pediatric liver transplantation. Reduction of immunosuppression remains a cornerstone of PTLD management, and in carefully selected patients, complete withdrawal can be achievable. Our findings suggest that supervised ISW can be performed in selected pediatric recipients without irreversible graft injury, although a risk of rejection persists. Careful patient selection, close clinical surveillance, and protocol liver biopsies are essential to ensure early detection and prompt management of rejection. Future studies are needed to refine selection criteria and identify biomarkers that may help guide safe ISW in this population.

Acknowledgements

We would like to thank the involved patients and their families for participating in the study. We also thank Siqi Liu, Junshan Liu, Rili Li, and Fanghui Cui for their invaluable efforts in maintenance of Beijing Friendship Hospital database of China Liver Transplant Registry.

Patient Permission/Consent Declarations

All the pediatric liver transplantation procedures were approved by the Ethics Committee of Beijing Friendship Hospital, and written informed consent was obtained from the legal guardians of all participants.

Declaration of Figures' Authenticity

All figures submitted have been created by the authors who confirm that the images are original with no duplication and have not been previously published in whole or in part.

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