Dissecting Epstein-Barr Virus-Specific T-Cell Responses After Allogeneic EBV-Specific T-Cell Transfer for Central Nervous System Posttransplant Lymphoproliferative Disease

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Epstein–Barr virus (EBV)-associated posttransplant lymphoproliferative disease (PTLD) with central nervous system (CNS) involvement is a severe complication after solid organ transplantation. Standard treatment with reduction of immunosuppression and anti-CD20 antibody application often fails leading to poor outcome. Here, we report the case of an 11-year-old boy with multilocular EBV-positive CNS PTLD 10 years after liver transplantation. Complete remission was achieved by repeated intravenous and intrathecal anti-CD20 antibody rituximab administration combined with intrathecal chemotherapy (methotrexate, cytarabine, prednisone) over a time period of 3 months. Due to the poor prognosis of CNS PTLD and lack of EBV-specific T-cells (EBV-CTLs) in patient's blood, we decided to perform EBV-directed T-cell immunotherapy as a consolidating treatment. The patient received five infusions of allogeneic EBV-CTLs from a 5/10 HLA-matched unrelated third-party donor. No relevant acute toxicity was observed. EBV-CTLs became detectable after first injection and increased during the treatment course. Next-generation sequencing (NGS) TCR-profiling verified the persistence and expansion of donor-derived EBV-specific clones. After two transfers, epitope spreading to unrelated EBV antigens occurred suggesting onset of endogenous T-cell production, which was supported by detection of recipient-derived clones in NGS TCR-profiling. Continuous complete remission was confirmed 27 months after initial diagnosis.

Keywords: posttransplant lymphoproliferative disease, adoptive T cell therapy, T cell receptor sequencing, transplantation, Epstein–Barr virus
INTRODUCTION

Posttransplant lymphoproliferative disease (PTLD) constitutes a heterogeneous group of lymphoproliferative disorders occurring as severe complications of immunosuppression after solid organ transplantation (SOT). Acquired by up to 15% of pediatric transplant recipients, most cases of childhood PTLD are of B-cell origin and associated with Epstein–Barr virus (EBV) infection or reactivation (1, 2). Long-lasting immunosuppressive therapy to prevent graft rejection as well as lack of EBV-specific immunity at the time of transplantation contribute to the high incidence and unfavorable prognosis of PTLD in children (1). Up to 20% of affected patients eventually succumb to the disease (1). While modulation of immunosuppressive therapy may be sufficient in some patients, multi-agent immuno-/chemotherapy serves as the primary treatment option for advanced stage PTLD in children resulting in 80% overall survival (3). In the PTLD-1 study, complete response to Rituximab conferred a favorable outcome in adults (4). Central nervous system (CNS) PTLD displays an unfavorable outcome with 30–50% overall survival (isolated disease) (5–7) and as low as 0–10% (combined systemic and CNS disease) (7, 8). For these high-risk patients, very limited treatment options are available. Intrathecal rituximab as a combination to intravenous immuno-/chemotherapy is a promising treatment option (9). In addition, transfer of EBV-specific T-cell lines manufactured from healthy volunteers has shown promise in some patients with CNS involvement (10). Here, we report the first case of treatment of an SOT patient with CNS PTLD receiving freshly isolated, partially HLA-matched EBV-specific T-cells (EBV-CTLs) from an unrelated third party donor in addition to intravenous and intrathecal chemono-/immunotherapy.

METHODS

Ethical Approval and Patient Informed Consent
The study was approved by the IRB of Hannover Medical School. The patient’s legal guardian gave written informed consent to both participation in the research project and publication of the case report.

Donor Pre-Testing, Production of EBV-CTLs, and Application

Frequencies of EBV-CTLs were determined in patient mother’s blood (not sufficient for transfer) as well as in five partially HLA-matched potential donors from the alloCELL T-cell donor registry (www.alloCELL.org, Tables 1 and 2) as described using EBV peptide pools EBV nuclear antigen 1 (ppEBNA-1) and EBV Select (ppSelect) (Miltenyi Biotec, Bergisch-Gladbach, Germany) (11).

Manufacturing of clinical-grade EBV-specific CD4+ and CD8+ T-cells from EBV-seropositive allogeneic 5/10 HLA-matched third party donor (TPD 1, Tables 1 and 2) was performed on a CliniMACS device using ppEBNA1 and ppSelect in combination and the IFN-γ Cytokine Capture System (Miltenyi Biotec). Quality control of the final T-cell product was done as described (11). Details on the T-cell manufacturing and product can be found in the Supplementary Material. The patient got one fresh and four cryopreserved EBV-specific T-cell products from a single manufacturing process.

Monitoring

Monitoring of viral load and EBV-specific T-cell frequencies in patient’s blood was done before and after T-cell transfer by IFN-γ ELISpot assay as described and using the following peptide pools: ppEBNA1, ppSelect, ppLMP2a, ppBZLF1 (all Miltenyi Biotec) (12, 13). If suitable numbers of PBMCs were obtained, EBV-CTLs were expanded over 7 days using the respective antigens ppEBNA1 and ppSelect in TexMACS media (Miltenyi Biotec) containing 50 U/ml IL-2 (Peprotec). After 7 days, IFN-γ ELISpot assay was repeated using the respective antigens. Expanded cells were used for TCR beta chain repertoire analysis.

TCR Beta Chain Repertoire Analysis

The stimulated and expanded PBMCs were stained with following antibodies: dead/alive (DAPI), hCD45, IFN-γ, ELISpot assay was repeated using the respective antigens.

TABLE 1: Donor selection: HLA characteristics and verification of donor’s Epstein–Barr virus (EBV)-specific memory T cells.

| Donor type | HLA-A | HLA-B | HLA-C | HLA-DR | HLA-DQ | IFN-γ Cytokine Capture System |
|------------|-------|-------|-------|--------|--------|-----------------------------|
| Patient    | 03    | 07/14 | 07/08 | 01/15  | 05     | EBNA1 + Select IFN-γ CSA [% IFN-γ CD3+ T cells] |
| Mother     | PMRD  | 03/11 | 07    | 07     | 15/16  | 06/06 IFN-γ CSA [% IFN-γ CD3+ T cells] |
| TPD 1      | PMUD  | 03/11 | 07/08 | 07     | 15/16  | 06/06 IFN-γ CSA [% IFN-γ CD3+ T cells] |
| TPD 2      | PMUD  | 03/11 | 07/08 | 07     | 15/16  | 06/06 IFN-γ CSA [% IFN-γ CD3+ T cells] |
| TPD 3      | PMUD  | 03/11 | 07/08 | 07     | 15/16  | 06/06 IFN-γ CSA [% IFN-γ CD3+ T cells] |
| TPD 4      | PMUD  | 03/11 | 07/08 | 07     | 15/16  | 06/06 IFN-γ CSA [% IFN-γ CD3+ T cells] |
| TPD 5      | PMUD  | 03/11 | 07/08 | 07     | 15/16  | 06/06 IFN-γ CSA [% IFN-γ CD3+ T cells] |

For ELISpot assay and CSA, EBV-specific T-cells were activated by 4 h in vitro restimulation with peptide pools EBNA1, Select and both in combination (EBNA1 + Consensus), respectively; TPD, third party donor; PMRD, partially matched related donor; PMUD, partially matched unrelated donor; HLA, human leukocyte antigen; IFN-γ, interferon-gamma; spw, spot per well; CSA, cytokine secretion assay; OF, original fraction, before enrichment; TCF, T-cell fraction, after magnetic enrichment; TNTC, too numerous to count.
FIGURE 1 | Continued

A

B

C

D

E

HE
CD20
EBER

LMP1
LMP2a
EBNA2
BZLF1

within viable lymphocytes
within viable leukocytes

Frequencies [%]

CD3+  CD19+  CD56+  CD3+CD56+  CD3+CD56-  CD33+  CD14+

LA
PreS
PF

relative numbers
absolute numbers

antigen-specific T cell [%]

CD3+IFN-γ within CD3
CD4+IFN-γ within CD4
CD8+IFN-γ within CD8
CD3+IFN-γ
CD4+IFN-γ
CD8+IFN-γ

total cell number

3.0x10^4
2.0x10^4
1.0x10^4
0
CASE PRESENTATION

An 11-year-old boy with Alagille syndrome received a related liver allograft during first year of life. Being EBV-negative at transplantation, seroconversion occurred 2 years later. Initial immunosuppression was based on tacrolimus, followed by a combination with mycophenolate mofetil. Ten years after transplantation, he suffered from severe headache, nausea, vomiting, and photophobia without B symptoms. Funduscopic examination revealed bilateral papilledema. Magnetic resonance imaging (MRI) studies of the brain demonstrated multifocal lesions in the left hemisphere (Figure 1A). After initial treatment for suspected toxoplasmosis, biopsy of the lesion revealed a monomorphic EBV-associated PTLD with features matching the patient's or donor's HLA-type are not contained (Figure 1B). Immunohistochemistry showed expression of CD20 and CD30. Most lymphoma cells expressed EBERs (Epstein–Barr encoded RNAs), LMP1 (EBV latent membrane protein 1), and LMP2a while EBNA2 (Epstein–Barr nuclear antigen 2) and BZLF1 (EBV immediate-early protein) were detected in a low number of neoplastic cells (Figure 1C). EBV PCR was negative in cerebrospinal fluid and weakly positive in peripheral blood (<1,000 copies/ml). Therefore, the diagnosis of EBV-related primary CNS PTLD was made.

Total body imaging and bone marrow aspirate histology displayed no evidence for systemic disease. During initial treatment with dexamethasone, symptoms rapidly improved. Immunosuppression was stopped and immune-/chemotherapy was initiated with six doses of intravenous (i.v.) rituximab (375 mg/m^2) and weekly intrathecal (i.th.) therapy with rituximab (40 mg), methotrexate (12 mg), cytarabine (30 mg), and prednison (10 mg) over 10 weeks (9). A partial response by MRI was observed after 3 weeks evolving to complete remission at the end of immuno-/chemotherapy. Due to poor prognosis and the lack of EBV-specific T cells in the patient's peripheral blood, we decided to consolidate treatment by transfer of partially HLA-matched EBV-CTLs.

RESULTS AND DISCUSSION

The patient received five doses of 2.5 × 10^4 EBV-CTLs/kg body weight from a 5/10 HLA-matched third party donor (TPD; Table 1). During the production process, CD3+ T-cells were enriched to >80% in the T-cell product with a predominance of CD8+ T-cells (Figures 1D,E; Data Sheet S1 in Supplemental Material). T-cells were administered every 3 weeks in the absence of graft-versus-host disease. After the second injection, the patient developed a skin rash around the neck, which turned out to be atopic dermatitis on histology and responded well to topical steroids without recurrence after subsequent T-cell injections. No other acute or chronic side effects were observed. EBV-PCR remained negative in peripheral blood throughout the whole course. After the end of treatment, immunosuppression was re-introduced with everolimus. At the last follow-up, 2 years after end of cellular therapy, the patient is in continuous remission of PTLD with good organ graft function.

No EBV-CTLs were detectable in patient blood on two occasions before adoptive immunotherapy (Figure 2A). In contrast, EBV-CTLs against ppEBNA1 and ppSelect became immediately and constantly detectable 4 days after the first T-cell transfer. While total numbers of CD3+, CD4+, and CD8+ T-cells remained stable throughout the treatment course, EBV-CTLs increased to a maximum of 40 per 250,000 PBMC before the second adoptive transfer. Over time, the target antigens of T-cell response broadened from initially EBNA1 and ppSelect to a broader response including T-cells against LMP2a and BZLF1, respectively (Figure 2A). Since epitopes from these two proteins matching the patient's or donor's HLA-type are not contained...
in the peptide pools used for manufacturing, this suggests that transfer of EBV-specific TPD cells induced an endogenous EBV-directed immune response in the patient, which was absent prior to immunotherapy. Frequency of EBV-CTLs increased during a 7-day in vitro restimulation and expansion demonstrating proliferative capacity (Figure 2B).
Figure 3: TCR beta chain sequencing of Epstein–Barr virus-stimulated T-cells before and after adoptive transfer. TCR beta chain sequencing was performed on blood samples at different timepoints before and after adoptive T-cell transfer and on the input sample itself. The left panel shows the samples enriched by stimulation with the ppEBNA1 peptide pool, whereas the right panel shows the ones after stimulation with ppSelect. Expansion of different shared clones is shown in both panels for exogenous (A) and endogenous (B) origin. Clones are labeled according to the antigen, origin (D, donor; R, recipient) and number. TCR sequences can be found in Table S1 in Supplementary Material.
TABLE 2 | T-cell receptor CDR3 sequences of clones displayed in Figures 3A,B.

cdr3 clones selectively detected in T cell product (donor = D) and post transfer | cdr3 clones detected in recipient (R) before transfer and post transfer
--- | ---
EBNA.D1 | EBNA.R1
EBNA.D2 | EBNA.R2
EBNA.D3 | EBNA.R3
EBNA.D4 | EBNA.R4
EBNA.D5 | EBNA.R5
EBNA.D6 | EBNA.R6
EBNA.D7 | EBNA.R7
EBNA.D8 | EBNA.R8
EBNA.D9 | EBNA.R9
EBNA.D10 | EBNA.R10
EBNA.D11 | EBNA.R11
EBNA.D12 | EBNA.R12
EBNA.D13 | EBNA.R13
EBNA.D14 | EBNA.R14
EBNA.D15 | EBNA.R15
EBNA.D16 | EBNA.R16
EBNA.D17 | EBNA.R17
EBNA.D18 | EBNA.R18
EBNA.D19 | EBNA.R19
EBNA.D20 | EBNA.R20
EBNA.D21 | EBNA.R21
EBNA.D22 | EBNA.R22
EBNA.D23 | EBNA.R23
EBNA.D24 | EBNA.R24
EBNA.D25 | EBNA.R25
EBNA.D26 | EBNA.R26
EBNA.D27 | EBNA.R27
EBNA.D28 | EBNA.R28
EBNA.D29 | EBNA.R29

Occasionally, transferred cells could be detected in patient material after transfer, but most authors were unable to retrieve TPD cells on analysis (14). We aimed at dissecting EBV-directed T-cell responses in the T-cell graft and the patient on a clonal molecular level. We performed TCR beta chain (TRB) repertoire analyses by NGS to follow-up the transferred cells and to monitor their expansion to EBV-associated antigens. Investigating the 77 shared clonotypes 41 were identified as expanding clones in CD8+ T cells after the transfer (Figures 3A,B). Four clones could be detected in both follow-up samples at 6 and 7 months after T-cell transfer, while the remaining 37 clones were picked up only once. Notably, the most abundant clone (EBNA. D8 = CASSAGPATNEKLFF, Figure 3A; Table 2) in the enriched T-cell product was not recovered at high abundance while two other clones that made up only 0.001% each of the donor’s T-cell product was not recovered at high abundance while two other clones that made up only 0.001% each of the donor’s T-cell product were detected in two patient samples obtained 7 months after transfer (EBNA. D1 = CASSSKRQVPD7TYF; Select.D6 = CASSPVRSSSETQYF, Figure 3A and Table 2). These findings suggest that at least a fraction of the transferred TPD cells were expanding and presumably contributing to EBV-specific T-cell responses in the patient. At the same time, we observed a sustained EBNA1-specific expansion of endogenous TRB sequences that were already present in the recipient’s CD8+ T-cell pool before TPD T-cell treatment (Figure 3B). This is consistent with the idea that exogenous T-cells stimulated an efficient endogenous anti-EBV T-cell response and may explain the finding that EBV-T-cell responses against unrelated antigens (LMP2, BZLF1) newly arise after T-cell transfer. Due to limited material availability, we performed the analyses on expanded cells after one in vitro peptide pool restimulation, which leaves the possibility of ex vivo TCR skewing. These limitations need to be considered in future clinical trials.

Prognosis of CNS PTLD is very poor with 30% overall survival (7, 8). We and others have reported successful administration of intrathecal rituximab; however, efficacy has not been validated in larger series (9, 15). Several studies and case reports show an effect of adoptive T-cell transfer in PTLD (10, 16–19). In particular, patients with CNS PTLD with poor outcome may benefit from this new treatment strategy (8, 9). Haque and colleagues reported responses in 3/5 patients with CNS PTLD after SOT using in vitro expanded EBV-specific TPD T-cell lines and lymphoma regression in CNS B-cell lymphoma in an immunodeficiency patient (10, 20). The efficacy of directly isolated EBV-CTLs in CNS PTLD after SOT is still unknown. Studies from patients after stem cell transplantation indicate that these cells are effective in CNS PTLD (19). In the case reported here, combined therapy with intrathecal chemotherapy and rituximab led to sustained complete remission of CNS PTLD. Transfer of partially HLA-matched EBV-CTLs provoked a robust anti-EBV T-cell response containing both exogenous and endogenous TRB signatures; the contribution of T-cell induction to ongoing remission remains uncertain.
Partially HLA-matched TPDs are an attractive source of virus-specific T-cells readily available if pre-screened and registered in T-cell donor registries (13). We did not observe any side effects of TPD T-cell transfer similar to other studies employing virus-specific T-cell therapy, which supports their feasibility and safety. Prospective studies are warranted to prove safety and efficacy of freshly isolated EBV-CTLs from TPDs in this vulnerable patient population.

ETHICS STATEMENT

This case study was carried out in accordance with the Declaration of Helsinki. Treatment was provided on a compassionate use basis. The monitoring protocol was approved by the “ethics committee of Hannover Medical School.” Patient and legal representatives gave written informed consent to the diagnostic program.

AUTHOR CONTRIBUTIONS

BM-K and BE-V designed research. RS-F, ST, LK, SR, and CS-F performed research. PH, LG, RB, UK, H-GH, and BE-V manufactured cell product and treated the patient. AK and IA performed histological analysis. ST, LK, SR, CK, IP, BE-V, and BM-K analyzed and interpreted data. RS-F, ST, LK, IP, PH, BE-V, and BM-K wrote the manuscript. All authors read the manuscript and approved the final version.

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SUPPLEMENTARY MATERIAL

The Supplementary Material for this article can be found online at https://www.frontiersin.org/articles/10.3389/fimmu.2018.01475/full#supplementary-material.

REFERENCES

1. Mynarek M, Schober T, Behrends U, Maeker-Kollhoff B. Posttransplant lymphoproliferative disease after pediatric solid organ transplantation. Clin Dev Immunol. 2013;2013:814973. doi:10.1155/2013/814973
2. Wistinghausen B, Gross TG, Bollard C. Post-transplant lymphoproliferative disease in pediatric solid organ transplant recipients. Pediatr Hematol Oncol. 2013;30:520–31. doi:10.3109/08880018.2013.798844
3. Gross TG, Orjuela MA, Perkins SL, Park JR, Lynch JC, Cairo MS, et al. Low-dose chemotherapy and rituximab for posttransplant lymphoproliferative disease (PTLD): a Children’s Oncology Group Report. Am J Transplant (2012) 12:3269. doi:10.1111/j.1600-6143.2012.04206.x
4. Trappe R, Oertel S, Leblond V, Mollep P, Sender M, Reinke P, et al. Sequential treatment with rituximab followed by CHOP chemotherapy in adult B-cell post-transplant lymphoproliferative disorder (PTLD): the prospective international multnicentre phase 2 PTLD-1 trial. Lancet Oncol (2012) 13:196–206. doi:10.1016/S1470-2045(11)70300-X
5. Evens AM, Choquet S, Kroll-Desrosiers AR, Jagadeesh D, Smith SM, Morschhauser F, et al. Primary CNS posttransplant lymphoproliferative disease (PTLD): an international report of 84 cases in the modern era. Am J Transplant (2013) 13:1512–22. doi:10.1111/ajt.12211
6. Cavaliere R, Petroni G, Lopes MB, Schiff D, O’Neill BP, Plotkin SR, et al. Primary central nervous system post-transplantation lymphoproliferative disorder: an international primary central nervous system lymphoma collaborative group report. Cancer (2010) 116:863–70. doi:10.1002/cncr.24834
7. Buell JE, Gross TG, Hanaway MJ, Trofe J, Roy-Chaudhury P, First MR, et al. Posttransplant lymphoproliferative disorder: significance of central nervous system involvement. Transplant Proc (2005) 37:59–65. doi:10.1016/j.transproceed.2004.12.130
8. Maeker B, Jack T, Zimmermann M, Abdul-Khalig H, Burdelki M, Fuchs A, et al. CNS or bone marrow involvement as risk factors for poor survival in post-transplantation lymphoproliferative disorders in children after solid organ transplantation. J Clin Oncol (2007) 25:4902–8. doi:10.1200/JCO.2006.10.2392
9. van de Gindt G, de Graaf S, Klein C, Cornelissen M, Maeker B, Loeften J. Intrahepatic rituximab treatment for pediatric post-transplant lymphoproliferative disorder of the central nervous system. Pediatr Blood Cancer (2008) 50(4):886–8. doi:10.1002/pbc.21297
10. Haque T, Willie GM, Jones MM, Higgins CD, Urquhart G, Wingate P, et al. Allogeneic cytotoxic T-cell therapy for EBV-positive posttransplantation lymphoproliferative disease: results of a phase 2 multicenter clinical trial. Blood (2007) 110:1123–31. doi:10.1182/blood-2006-12-063008
11. Tischer S, Priesner C, Heuft H-G, Goudeva L, Mende W, Barthold M, et al. Rapid generation of clinical-grade antiviral T cells: selection of suitable T-cell donors and GMP-compliant manufacturing of antiviral T cells. J Trans Med (2014) 12:336. doi:10.1186/s12976-014-0336-5
12. Tischer S, Dieks D, Sukdolak C, Bunse C, Figueiredo C, Immenschuh S, et al. Evaluation of suitable target antigens and immunoassays for high-accuracy immune monitoring of cytomegalovirus and Epstein-Barr virus-specific T cells as targets of interest in immunotherapeutic approaches. J Immunol Methods (2014) 408:101–13. doi:10.1016/j.jim.2014.05.011
13. Sukdolak C, Tischer S, Dieks D, Figueiredo C, Goudeva L, Heuft HG, et al. CMV-, EBV- and ADV-specific T cell immunity: screening and monitoring of potential third-party donors to improve post-transplantation outcome. Biol Blood Marrow Transplant (2013) 19:1480–92. doi:10.1016/j.bbmt.2013.07.015
14. Uhlin M, Gertow J, Uzunel M, Okas M, Berglund S, Watz E, et al. Rapid salvage treatment with virus-specific T cells for therapy-resistant disease. Clin Infect Dis (2012) 55:1064–73. doi:10.1093/cid/cis625
15. Rubenstein JL, Combs D, Rosenber J, Levy A, McDermott M, Dameron L, et al. Rituximab therapy for CNS lymphomas: targeting the leptomeningeal compartment. Blood (2003) 101:466–8. doi:10.1182/blood-2002-06-1636
16. Moosmann A, Bigalke I, Tischer J, Schirrmann L, Kasten J, Tippmann S, et al. Effective and long-term control of EBV PTLD after transfer of peptide-selected T cells. Blood (2010) 115:2960–70. doi:10.1182/blood-2009-08-236356
17. Doubrovina E, Oflaz-Sozmen B, Prokop SE, Kerman NA, Abramson S, Teruya-Feldstein J, et al. Adoptive immunotherapy with unselected or EBV-specific T cells for biopsy-proven EBV lymphomas after allogeneic hematopoietic cell transplantation. Blood (2012) 119:2644–56. doi:10.1182/blood-2011-08-371971
18. Heapel H, Slowod K, Pule M, Hale G, Rousseau A, Smith C, et al. Long term outcome of EBV specific T-cell infusions to prevent or treat EBV-related lymphoproliferative disease in transplant recipients. Blood (2009) 115:925–36. doi:10.1182/blood-2009-08-239186
19. Icheva V, Kayser S, Wolff D, Tuve S, Kyzirakos C, Bethge W, et al. Adoptive transfer of Epstein-Barr virus (EBV) nuclear antigen 1-specific T cells as treatment for EBV reactivation and lymphoproliferative disorders after allogeneic stem-cell transplantation. J Clin Oncol (2013) 31:39–48. doi:10.1200/JCO.2011.39.8495
20. Wynn RF, Arkwright PD, Haque T, Gharib MI, Wilkie G, Morton-Jones M, et al. Treatment of Epstein-Barr-virus-associated primary CNS B-cell lymphoma with allogeneic T-cell immunotherapy and stem-cell transplantation. Lancet Oncol (2005) 6:334–6. doi:10.1016/S1470-2045(05)70717-6
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