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EBV-Associated Lymphoproliferative Disorders: Classification and Treatment

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Key Words. EBV • EBV-associated lymphomas • Post-transplant-associated lymphoproliferative disorders • HIV-associated lymphoproliferative disorders • EBV cell–based immunotherapy

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LEARNING OBJECTIVES

After completing this course, the reader will be able to:
1. Assess patients with EBV-associated lymphoproliferative disorders.
2. Describe the pathogenesis of the lymphoproliferative disorders linked to EBV infection.

ABSTRACT

Since its discovery as the first human tumor virus, Epstein-Barr virus (EBV) has been implicated in the development of a wide range of B-cell lymphoproliferative disorders, including Burkitt’s lymphoma, classic Hodgkin’s lymphoma, and lymphomas arising in immunocompromised individuals (post-transplant and HIV-associated lymphoproliferative disorders). T-cell lymphoproliferative disorders that have been reported to be EBV associated include a subset of peripheral T-cell lymphomas, angioimmunoblastic T-cell lymphoma, extranodal nasal type natural killer/T-cell lymphoma, and other rare histotypes. EBV encodes a series of products interacting with or exhibiting homology to a wide variety of antiapoptotic molecules, cytokines, and signal transducers, hence promoting EBV infection, immortalization, and transformation. However, the exact mechanism by which EBV promotes oncogenesis is an area of active debate. The focus of this review is on the pathology, diagnosis, classification, and pathogenesis of EBV-associated lymphomas. Recent advances in EBV cell–based immunotherapy, which is beginning to show promise in the treatment of EBV-related disorders, are discussed. The Oncologist 2008;13:577–585

**INTRODUCTION**

Epstein-Barr virus (EBV) is a member of the herpesvirus family. As with other herpesviruses, EBV is an enveloped virus that contains a DNA core surrounded by an icosahedral nucleocapsid and a tegument. Family members include herpes simplex I and II and varicella zoster virus (α-herpesvirus subfamily), cytomegalovirus and human herpesvirus (HHV)-6 and HHV-7 (β-herpesvirus subfamily), and HHV-8 and EBV (γ-herpesvirus subfamily) [1].

Human tumors have been attributed to both HHV-8 (Kaposi’s sarcoma, primary effusion lymphoma [PEL], and multicentric Castleman’s disease) and EBV (Burkitt’s lymphoma, nasopharyngeal carcinoma, and Hodgkin’s and non-Hodgkin’s lymphomas) [2–5].

In this review we discuss only the EBV-associated lymphoproliferative disorders, dividing them into B-cell and T/natural killer (NK)-cell lymphomas. The focus is on the pathology, diagnosis, and classification, pathogenesis, and treatment of EBV-associated lymphomas.

**EBV PRODUCTS AND PATTERNS OF EBV GENE EXPRESSION**

The EBV genome encodes a series of products interacting with or exhibiting homology to a wide variety of antiapoptotic molecules, cytokines, and signal transducers, hence promoting EBV infection, immortalization, and transformation [6–8].

Based on patterns of expression of the EBV genome, three types of latent gene expression have been described: latency I, II, and III (Table 1). During latency I, Epstein–Barr nuclear antigen 1 (EBNA-1) and the two small non-coding Epstein–Barr RNAs (EBERs) are expressed [9]. EBV gene expression in latency II is usually limited to EBNA-1, the EBERs, latent membrane protein (LMP)-1, and LMP-2A and LMP-2B [10]. Latency III usually involves the unrestricted expression of all EBNAs, EBERs, and LMPs [10]. Latency I is generally associated with the EBV-related Burkitt’s lymphoma [9], latency II has been associated with classic Hodgkin’s lymphoma (cHL) and T-cell non-Hodgkin’s lymphoma (NHL) [11], and latency III occurs mainly in immunocompromised individuals suffering from post-transplant lymphoproliferative disorders (PTLDs) and HIV-associated lymphoproliferative disorders and in lymphoblastoid cell lines [12].

Furthermore, EBV encodes the following important proteins that show sequence and functional homology to diverse human proteins: BCRF-1 and interleukin (IL)-10, BHRF-1 and BCL-2, BARF-1 and intracellular adhesion molecule 1 [13].

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**Table 1. Latent EBV-encoded genes**

<table>
<thead>
<tr>
<th>EBV-encoded genes</th>
<th>Location</th>
<th>Latency type</th>
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<tbody>
<tr>
<td>EBNA-1</td>
<td>Nucleus</td>
<td>I, II, III</td>
</tr>
<tr>
<td>EBNA-2</td>
<td>Nucleus</td>
<td>III</td>
</tr>
<tr>
<td>EBNA-3</td>
<td>Nucleus</td>
<td>III</td>
</tr>
<tr>
<td>LMP-1</td>
<td>Membrane</td>
<td>II, III</td>
</tr>
<tr>
<td>LMP-2</td>
<td>Membrane</td>
<td>II, III</td>
</tr>
<tr>
<td>EBER-1 and EBER-2</td>
<td>Nucleus</td>
<td>I, II, III</td>
</tr>
</tbody>
</table>

Abbreviations: EBV, Epstein–Barr virus; EBNA, Epstein–Barr nuclear antigen; LMP, latent membrane protein; EBER, Epstein–Barr RNA.

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**Table 2. EBV-associated lymphoproliferative disorders**

**EBV-associated B-cell lymphoproliferative disorders**

<table>
<thead>
<tr>
<th>Disorder</th>
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<tbody>
<tr>
<td>Burkitt’s lymphoma</td>
</tr>
<tr>
<td>Classic Hodgkin’s lymphoma</td>
</tr>
<tr>
<td>Post-transplant lymphoproliferative disorders</td>
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<tr>
<td>HIV-associated lymphoproliferative disorders</td>
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<tr>
<td>Primary central nervous system lymphoma</td>
</tr>
<tr>
<td>Diffuse large B-cell lymphoma, immunoblastic</td>
</tr>
<tr>
<td>HHV-8–positive primary effusion lymphoma and its solid variant</td>
</tr>
<tr>
<td>Plasmablastic lymphoma</td>
</tr>
<tr>
<td>Other histotypes (rare)*</td>
</tr>
</tbody>
</table>

*Other histotypes include: lymphomatoid granulomatosis, pyothorax–associated lymphoma, senile EBV-associated B-cell lymphoproliferative disorders.

Abbreviations: EBV, Epstein–Barr virus; HHV-8, human herpesvirus 8.

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**EBV-ASSOCIATED B-CELL LYMPHOPROLIFERATIVE DISORDERS**

Since its discovery as the first human tumor virus, EBV has been implicated in the development of a wide range of B-cell lymphoproliferative disorders (Table 2).

**Burkitt’s Lymphoma**

Burkitt’s lymphoma is a highly proliferative B-cell tumor that includes three variants: endemic (affecting children in equatorial Africa and New Guinea), sporadic (affecting children and young adults throughout the world) (Fig. 1), and immunodeficiency related (primarily in association with HIV infection). EBV has been detected in virtually all cases of the endemic variant, 15%–20% of cases of the sporadic variant, and 30%–40% of cases of the immunodeficiency-related variant [1]. In all variants, irrespective of EBV status, constitutive activation of the c-myc oncogene through its translocation into one of the immunoglobulin
loci is clearly the key factor in the oncogenesis of Burkitt’s lymphoma [1]. The detection of somatic hypermutations in the V region of the immunoglobulin genes and the phenotype of the Burkitt’s lymphoma cells indicate a germinal center (GC) cell origin of the lymphoma [14]. The tumors isolated from nonendemic Burkitt’s lymphoma patients are usually from different stages of B-cell development than those isolated from patients with endemic Burkitt’s lymphoma [15, 16].

Most EBV-positive cases exhibit a highly restrictive pattern of expression of latent encoded proteins, only expressing EBNA-1 and the EBERs (latency I) [1]. However, it was recently reported that some cases, in addition to EBNA-1 and the EBERs, express EBNA-3A, EBNA-3B, EBNA-3C, and EBNA leader protein but still lack EBNA-2 and the latent membrane proteins [17]. EBNA-1 plays a crucial role in the maintenance and replication of the viral genome, but its oncogenic potential is highly controversial [1, 18]. Conversely, as the EBERs are believed to possess antiapoptotic activity, it has been postulated that they may play an essential role in the oncogenesis of Burkitt’s lymphoma [19].

cHL
Not all subtypes of cHL harbor EBV to the same degree [20–22]. There are also data that suggest that the incidence of EBV-positive cHL is age related [23].

Hodgkin’s Reed-Sternberg (RS) cells of cHL represent transformed B cells. The detection of destructive somatic mutations in the rearranged immunoglobulin genes of RS cells of cHL indicates that they originate from preapoptotic GC B cells [24]. In RS cells, constitutive nuclear factor (NF)-κB activation, which is essential for RS cell survival, seems to result from various mechanisms. Several receptors, such as CD30, CD40, receptor activator of nuclear factor κB (RANK), and RANK ligand, which activate the classic NF-κB pathway, are expressed by RS cells, and ligands for these receptors are frequently expressed on activated CD4+ T cells and other bystander cells surrounding RS cells, including eosinophils, neutrophils, and B-cell subsets. In EBV-positive cHL cases, the LMP-1 gene also contributes to NF-κB activation because it mimics activated CD40 receptors [25–27].

In HIV/AIDS persons with severe immunosuppression, the acquisition of an antiapoptotic phenotype by RS cells is not a result of CD40–CD40L interactions, which are lacking, but rather is induced by EBV-encoded LMP-1, which is functionally homologous to activated CD40. Thus, EBV is thought to play a pivotal role in the pathogenesis of lymphoma in patients with immunosuppression [28].

EBV-Associated Lymphomas in Immunocompromised Individuals
There exist several distinct classes of EBV-associated lymphoproliferative disorders in immunocompromised individuals. First, there is a disorder resulting from an inherited immunodeficiency known as X-linked lymphoproliferative disorder. Second, there are lymphomas associated with immunosuppressive drugs given to transplant recipients. Finally, there are AIDS-related lymphoproliferative disorders. The most common gene-expression pattern in these disorders is latency III.

PTLDs
Since the original report in 1969 [29, 30], it has been well established that there is a higher incidence of lymphoproliferative disorders in transplant recipients of both a solid organ and bone marrow. According to the World Health Organization (WHO) classification [31], PTLDs may be classified into: (a) early lesions, generally represented by EBV-driven polyclonal lymphoproliferations and (b) true monoclonal diseases, including polymorphic PTLD and monomorphic PTLD; the latter is further distinguished into Burkitt’s lymphoma/Burkitt’s-like lymphoma, diffuse large B-cell lymphoma (DLBCL), and cHL.

Early-onset PTLDs are mainly regarded as EBV-driven lymphoproliferations that are frequently, though not always, polyclonal or oligoclonal, whereas most late-onset PTLDs are true monoclonal lymphoid malignancies that are
not necessarily associated with EBV infection. Correlative studies of the morphologic and molecular features of PTLDs have contributed to the recognition of specific disease categories and have provided prognostic indicators for these disorders [32]. Among monoclonal B-cell PTLD, derivation from GC-related B cells occurs independently of the type of organ transplanted, the interval between transplant and lymphoma, the histology, and the site of origin of the lymphoma.

Oncogenic viruses known to be involved in PTLD pathogenesis include EBV and HHV-8. Both EBV and HHV-8 act predominantly through direct mechanisms, that is, the virus is able to directly infect the tumor clone and exerts a transforming effect upon B cells. Viral infection in PTLD exploits several strategies to ensure persistent infection, namely, prevention of death of infected cells, enhancement of their proliferation to maintain the infected reservoir, and evasion of the immune system [33–35]. Several lines of evidence suggest that EBV infection has a major pathogenetic role in PTLDs: (a) EBV infects 60%–80% PTLD patients, including 100% of early-onset PTLD patients [6]; (b) in many cases of monomorphic PTLD, EBV infection is monoclonal, consistent with the hypothesis that the virus has been present in the tumor progenitor cell since the early phases of clonal expansion; (c) a decrease in EBV-specific cytotoxic T lymphocytes (CTLs) and an increase in the EBV viral load are strongly associated with PTLD development [36]; and (d) treatment of PTLDs with autologous EBV-specific CTLs results in viral load control and tumor size reduction.

**HIV-Associated Lymphoproliferative Disorders**

HIV-associated lymphoproliferative disorders are a heterogeneous group of diseases that arise in the presence of HIV-associated immunosuppression, a state that permits the unchecked proliferation of EBV-infected lymphocytes. Traditionally, these aggressive disorders include both central nervous system and systemic lymphomas. PEL also occurs and often involves EBV in addition to HHV-8.

The categories of HIV-associated NHL (HIV-NHL) included in the latest WHO proposal [37] are grouped as follows. (a) Lymphomas also occurring in immunocompetent patients. The vast majority of these HIV-NHLs belong to three high-grade B-cell lymphomas: Burkitt’s lymphoma, DLBCL with centroblastic features, and DLBCL with immunoblastic features. According to the site of involvement, the present spectrum of HIV-NHL includes extranodal/nodal lymphomas and primary central nervous system lymphomas. (b) Unusual lymphomas occurring more specifically in HIV-positive patients. These lymphomas include two rare entities, namely, PEL and plasmablastic lymphoma of the oral cavity [38, 39]. (c) Lymphomas also occurring in other immunodeficiency states.

EBV-associated HIV-NHL includes primary central nervous system lymphoma, systemic lymphomas having a DLBCL immunoblastic morphology (Fig. 2), HHV-8–positive PEL and its solid variant with and without serous effusions (Fig. 3), and plasmablastic lymphoma [7, 40–46] (Table 2).

**EBV-ASSOCIATED T/NK-CELL LYMPHOPROLIFERATIVE DISORDERS**

EBV is known primarily for its ability to infect B cells, but it can also infect other cells. Several types of non-B-cell NHL are associated with EBV (Table 3). T-cell lymphoproliferative disorders that have been reported to be EBV associated include a subset of peripheral T-cell lymphomas, angioimmunoblastic T-cell lymphoma (AITL), extranodal nasal type NK/T-cell lymphoma, enteropathy-type T-cell lymphoma, γδ T-cell lymphomas (hepatosplenic and non-hepatosplenic), T-cell lymphoproliferative disorders after chronic EBV infection, EBV-associated cutaneous T-cell lymphoproliferative disorders (especially in Asia), and aggressive NK-cell leukemia/lymphoma [8]. This section focuses on the two types in which EBV has been most directly implicated: AILT and nasal T/NK-cell lymphoma.
Peripheral T-Cell Lymphomas

Peripheral T-cell lymphomas (PTCLs) represent 10%–15% of all lymphoid neoplasms [47]. They are a heterogeneous group of tumors that in the Revised European-American Lymphoma/WHO classification are subdivided into specified and unspecified forms [47, 48].

AILT

AILT is a PTCL characterized by systemic disease, a polymorphous infiltrate primarily involving lymph nodes, and prominent proliferation of high endothelial venules and follicular dendritic cells [49]. AILT is the second most common PTCL subtype, accounting for 15%–20% of cases [49]. Laboratory findings include polyclonal hyper-globulinemia, circulating immunocomplexes, cold agglutinins with hemolytic anemia, positive rheumatoid factor, and anti–smooth muscle antibodies. The clinical behavior is very aggressive, the response to therapy is scarce, and the long-term outcome is dismal. The differential diagnosis with other PTCLs, and in particular with unspecified PTCL, is puzzling and mainly based on different features of the reactive components (i.e., prominent vascular structures and abundant follicular dendritic cells) and phenotype (CD10 and BCL-6 expression) [50]. Notably, no unique markers can reliably discriminate between these two entities. Recently, chemokine (C-X-C motif) ligand 13 was proposed as a possible candidate to distinguish the two diseases [51].

The molecular pathogenesis of AILT, as in general for all peripheral T-cell neoplasms, is poorly understood. The karyotype is often characterized by complex abnormalities, but specific alterations have not been identified, and only few reports focused on the gene-expression profile of nodal PTCLs [51, 52]. In one study, an AILT molecular signature was identified together with a molecular link between AILT and T follicular helper lymphocytes [52].

AILT is a lymphoma in which expanding B-cell clones are often present beside the T-cell clones. EBV infection is seen mainly in the B lymphocytes and B immunoblasts, although the virus also occurs in rare neoplastic and non-neoplastic T cells. The presence of EBV in only a subpopulation of cells suggests that EBV infection is secondary to malignancy or that the viral genome has been lost from the malignant cell [6].

Nasal T/NK-Cell Lymphoma

Nasal T/NK-cell lymphoma cells exhibit several unique genotypic and phenotypic features. These features include an absence of T-cell antigens, the expression of the NK cell marker CD56, and the absence of T-cell receptor gene rearrangement [15]. Clinically, these tumors occur in the nasal and upper aerodigestive area. EBV is consistently associated with these lymphomas, regardless of geographical location [53, 54] (Fig. 4).

TREATMENT

This section focuses on the treatment of patients with EBV-associated lymphomas in which EBV cell–based immunotherapy has been most directly applied.

EBV Cell–Based Immunotherapy

Treatment of EBV-Related PTLDs

Because the great majority of PTLDs occurring in patients receiving a solid organ transplant (SOT) or allogeneic stem cell transplant are a result of EBV infection/reactivation that follows the impairment of the immune system, modulation of the immunosuppressive treatment is recom-

Figure 3. HHV-8-associated extracavitary solid lymphoma without serous effusion. Large tumor cells express EBV infection as detected by EBER in situ hybridization. Tumor cells are pleomorphic and of heterogeneous size. (EBER in situ hybridization, hematoxylin counterstain, original magnification ×400.)

Abbreviations: EBER, Epstein–Barr RNA; EBV, Epstein–Barr virus; HHV-8, human herpesvirus 8.

Table 3. EBV-associated lymphoproliferative disorders

<table>
<thead>
<tr>
<th>EBV-associated T/NK-cell lymphoproliferative disorders</th>
</tr>
</thead>
<tbody>
<tr>
<td>Peripheral T-cell lymphoma, unspecified</td>
</tr>
<tr>
<td>Angioimmunoblastic T-cell lymphoma</td>
</tr>
<tr>
<td>Extranodal nasal T/NK-cell lymphoma</td>
</tr>
<tr>
<td>Other histotypes (rare)</td>
</tr>
</tbody>
</table>

Other histotypes include hepatosplenic T-cell lymphoma, nonhepatosplenic γδ T-cell lymphomas, enteropathy-type T-cell lymphoma.

Abbreviations: EBV, Epstein–Barr virus; NK, natural killer.

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mended as a first-line approach in patients with PTLDs [55]. This approach induces tumor regression in 25%–50% of patients [55]. Importantly, modulation of immunosuppression is currently applied by many transplant groups in patients with a high EBV-DNA viral load in order to prevent the occurrence of a PTLD [56].

The management of patients with PTLDs who fail the reduction in immunosuppression is complex and remains controversial. Chemotherapy and monoclonal antibodies are the therapeutic options most frequently offered to these patients. The use of rituximab has significantly changed the therapeutic approach to PTLDs. The administration of the antibody is generally well tolerated and rapidly induces depletion of mature B lymphocytes in the peripheral blood, thus reducing the compartment of EBV-infected cells, with an associated normalization of the viral load [57]. In the case of PTLDs occurring in SOT patients, very enthusiastic results have been reported in small uncontrolled studies, with a complete remission rate in the range of 30%–70% [58, 59]. However, more recent phase II studies including larger cohorts of patients have shown an overall median survival time of 15 months, suggesting that rituximab should be integrated with chemotherapy in patients with poor prognostic factors, such as a high tumor burden, an EBV-negative PTLD, and PTLD occurring late after transplantation [60].

In our own experience, the duration of remission in SOT patients with EBV-related PTLD treated with rituximab can be relatively short, because the recovery of B cells in the presence of impaired T-cell immunity is often accompanied by a new increase in the EBV viral load and eventually relapse of the PTLD [61].

Chemotherapy still remains the only option for many patients with aggressive PTLDs. The response rate and overall survival time were widely variable on retrospective analysis, and few prospective studies have been reported so far [62–64]. The best time and the best chemotherapy schedule for these patients remain controversial. Low-dose chemotherapy has been prospectively used in order to reduce the morbidity and mortality associated with standard protocols. Although this approach significantly reduces toxicities, it is also associated with a high relapse rate, suggesting that more conventional chemotherapy regimens are indicated in patients with aggressive PTLD [65]. CHOP-like regimens are currently recommended by the European guidelines at least for renal transplantation [66].

Is there a role for cellular immunotherapy in patients with EBV-related PTLD? Adoptive transfer of donor-derived EBV-specific CTLs (EBV-CTLs) has been successful when used as prophylaxis and treatment of EBV-related PTLDs in the context of allogeneic stem cell transplantation [67]. Analogously, adoptive transfer of EBV-CTLs may help to restore the balance of the immune compartment in patients receiving a SOT, while avoiding the risk for graft rejection that is frequently associated with the withdrawal of immunosuppression.

Several groups recently reported their preliminary experiences with adoptive EBV-CTL transfer in SOT recipients as pre-emptive treatment in cases of high EBV DNA viral load or as treatment in the presence of clinical evidence of a PTLD [68, 69]. Overall, these studies indicate that CTL infusions are well tolerated. No graft toxicity was reported. Although EBV DNA did not decrease uniformly in all patients, in several studies EBV-CTLs prevented the development of PTLDs, and in small series tumor regression after CTL therapy was also reported.

The main limitations to the extensive application of CTL therapies in these patients remain the cost and the time required for EBV-CTL line generation. PTLDs can be very aggressive and rapidly fatal, while the generation of autologous EBV-CTL lines requires several weeks. The use of partly HLA-matched EBV-CTLs seems a promising alternative to the generation of autologous EBV-CTLs [70]. In our first experience, we generated EBV-CTL lines for patients with a high EBV DNA viral load in the peripheral blood (>4,000 DNA copies). However, because the EBV DNA viral load is not always predictive of PTLD occurrence in SOT recipients, we are currently generating EBV-CTL lines only for patients who have received previous treatment for a PTLD. Because the relapse rate remains high in these patients, we are proposing the generation and

Figure 4. Nasal T/NK-cell lymphoma. EBV-infected tumor cells display an angiocentric pattern of growth. The figure shows several vascular structures of heterogeneous size. (EBER in situ hybridization, hematoxylin counterstain, original magnification ×400.)

Abbreviations: EBER, Epstein–Barr RNA; EBV, Epstein–Barr virus; NK, natural killer.
infusion of EBV-CTLs as a salvage treatment, thus reducing the cost of manufacturing and decreasing the risk–benefit ratio for this procedure.

**Treatment of EBV-Associated cHL**

Modern radiotherapy and/or chemotherapy regimens have dramatically improved the cure rate of patients with cHL [71]. However, despite the identification of clinical prognostic factors and the optimal use of primary and secondary treatments, cHL remains fatal for >15% of patients [71]. Therefore, new therapeutic agents are required for primary refractory patients and biological strategies are also desirable to maintain remission in high-risk patients after conventional treatment. In addition, biological treatments may reduce many of the serious long-term side effects correlated with radiation and chemotherapy [71].

Therapy with monoclonal antibodies, and in particular with anti-CD30 monoclonal antibodies, has been used in patients with cHL. Although the great majority of patients enrolled in these studies were heavily resistant to previous therapies, clinical responses were minimal and not comparable with the response rate obtained with rituximab in B-cell–derived lymphomas [72].

The association between EBV and a subset of cHL and the expression of the latent proteins LMP-1 and LMP-2 by tumor cells constitute the rationale to assess the feasibility of EBV-CTL adoptive transfer for patients with EBV-related cHL. We have conducted several phase I clinical trials in patients with cHL using both polyclonal EBV-CTLs and CTLs enriched in precursors targeting LMP-2 [73]. CTL infusions were well tolerated, with objective and sustained clinical responses in several patients.

**EBV-CTL Therapies and Future Directions**

These studies demonstrate both the feasibility and efficacy of CTL therapies in patients with PTLDs after SOT and in patients with EBV-associated malignancies like cHL. However, other important factors need to be considered to improve the clinical benefit of these approaches.

SOT patients require continuous immunosuppression to prevent graft rejection. Although adoptive transfer of EBV-CTLs may help in restoring immunocompetence, CTL function is still partially impaired by the immunosuppressive drugs [73]. We and others are currently evaluating strategies to genetically modify EBV-CTLs to make them resistant to the effects of immunosuppressive drugs.

A more robust in vivo expansion of adoptively transferred CTLs may help to increase the antitumor effects. This concept was proven in previous clinical trials in melanoma patients in which adaptive T-cell transfer followed the administration of nonmyeloablative chemotherapy regimens to create a lymphodepleted environment that would favor the expansion of CTLs [74]. The tumor microenvironment may be particularly hostile for antitumor immunity. Indeed, many tumor cells, such as HL tumor cells, produce factors, including transforming growth factor β, thymus and activation-regulated chemokine, IL-10 and Fas ligand, that significantly impair the function or survival of antitumor CTLs. Genetic modification of CTLs to overcome tumor escape mechanisms and increase their survival in the tumor microenvironment may help in arming CTLs and increasing their antitumor effects [75]. Finally, the capacity of tumor cells to downregulate tumor-associated antigens needs to be carefully evaluated. Modern technology allows the genetic modification of CTLs to expand their antigen specificity. In particular, gene transfer of chimeric antigen receptors targeting crucial molecules expressed on the cell surface of cHL cells, such as CD30, may improve their antitumor effects. Phase I clinical studies will help in clarifying the role of these mechanisms to improve the treatment of these malignancies.

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Final approval of manuscript: Antonino Carbone, Annunziata Gloghini, Giampietro Dotti

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