Review paper

# Challenging cases of Epstein-Barr virus positive lymphomas

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Epstein-Barr virus (EBV)-associated lymphomas can develop in both immunocompetent and immunocompromised patients, such as those with human immunodeficiency virus (HIV) infection, other primary or acquired immunodeficiencies, or *post* transplantation. The current World Health Organization classification consolidated immunodeficiency/dysregulation-related lymphoproliferative disorders into three major histopathological categories: hyperplasia, lymphoproliferative disorder of varied malignant potential, and lymphoma; and reporting the histopathological category, the presence or absence of causally associated viruses (EBV, KSHV/HHV-8), and the immune deficiency/dysregulation setting is recommended. In hematopathology practice, EBV is best confirmed by EBV-encoded RNA *in situ* hybridization, while immunohistochemistry is used as a complementary test for the presence of EBV-related proteins. Often, additional analyses such as fluorescence *in situ* hybridization and polymerase chain reaction for B- and T-cell clonality have to be applied for the final classification. This review, summarizing a lecture given at the 10th Course of the Academy of Immunohistochemistry in Cracow, Poland (June 2026), presents some challenging examples of EBV-related lymphomas. Cases include EBV+ mucocutaneous ulcer with subsequent EBV+ diffuse large B-cell lymphoma, EBV+ diffuse large B-cell lymphoma with plasma cell differentiation, *post* therapy EBV+ Burkitt lymphoma and cutaneous NK/T-cell lymphoma.

**Key words:** lymphoma, B-cell, T-cell, Epstein-Barr virus, EBER, immunohistochemistry.

## Introduction

Epstein-Barr virus (EBV, human herpesvirus 4 – HHV-4) is a double-stranded DNA virus belonging to the Gammaherpesvirinae subfamily of the Herpesviridae family. Epstein-Barr virus was first identified in Burkitt lymphoma (BL) cells in 1964 [1]. Subsequently, a wide spectrum of lymphomas has been associated with EBV infection (Table I), as well as 85% of nasopharyngeal carcinoma cases and approximately 10% of gastric cancer cases [2].

Epstein-Barr virus associated lymphomas can develop in both immunocompetent and immunocompromised patients, such as those with human immunodeficiency virus (HIV) infection, other primary or acquired immunodeficiencies, or *post* transplantation. The current World Health Organization (WHO) Classification of Tumors (2022, 5th ed.) [3] consolidated the immune deficiency/dysregulation (IDD)-related lymphoproliferative disorders (LPD) into three major histopathological categories: hyperplasia, lymphoproliferative disorder of varied malignant potential, and lymphoma. The World Health Organization Classification of Tumors, 5th ed. recommends reporting the histopathological category, the presence or absence of causally associated viruses (EBV, KSHV/HHV-8), and the IDD setting. Immunodeficiency-related tumors are more often EBV-positive than the same type of lymphoma in immunocompetent patients. There are genetic differences



 

**Table I.** Most common EBV-related lymphoproliferative disorders and lymphomas

| Entity* | Immunophenotype | EBV latency and viral load | Molecular characteristics |
|---|---|---|---|
| EBV+ mucocutaneous ulcer | Large cells CD20+, CD30+, CD15+/– | II (50%) or III (50%) Viral load negative | Clonal *IGH*/*IGK* in 50% of cases |
| EBV+ polymorphic LPD | Large cells CD20+, CD30+, CD15 –/(+) | II or III High viral load | Clonal *IgH*/*IGK* in 50–70% of cases |
| Lymphomatoid granulomatosis | Large cells CD20+, CD30+, CD15– Three grades bases on numbers of EBER+ cells | Mostly II (III in pts with immunosuppression) Viral load low or negative | Clonal *IGH*/*IGK* in grade 2 and 3 |
| Burkitt lymphoma | CD20+, CD10+, BCL-2-, BCL-6+, C-MYC+ | I High viral load | Clonal *IGH*/*IGK* t(8;14) |
| EBV+ diffuse large B-cell lymphoma NOS | Often HRS-like cells positive for CD20, CD30, CD15–/(+), MUM-1+, CD10–, PDL1+ | Mostly II ((III in 10% pts mostly with immunosuppression) High viral load | Clonal *IGH*/*IGK* Alterations of *NF-α B*, WNT and IL6/JAK/STAT pathways |
| Classical Hodgkin lymphoma | HRS cells CD20– (+), PAX5+, CD30+, CD15+ (–), PDL1+, EBER+ in 40% | II IgG antibody titers against VCA and EA complex higher in EBV+ cases | Monoclonal *IGH* rearrangements in microdissected HRS cells, alterations of *NF-α* B pathway, recurrent chromosomal imbalances |
| Extranodal NK/T-cell lymphoma | Positive for cCD3, CD2, TIA-1, GranzB, perforin and CD56 (80%), negative for sCD3. CD7 +/–, neg for CD4, CD8, CD16, CD57 | I/II Viral load correlates with stage | *JAK*/*STAT* pathway alterations (*STAT3*, *JAK3*, *STAT5B*), epigenetic regulators (*BCOR*, *KMT2D*, *ARUD1A*, *EP300*) |
| Angioimmunoblastic T-cell lymphoma | Large B-cell immunoblasts positive | II | Mutations in *TET2*, *IDH2*, *DNMT3A*, *RHOA*, and TCR |

*EBV – Epstein-Barr virus, LPD – lymphoproliferative disorder, NK – natural killer, TCR – T-cell receptor*
** In World Health Organization 5th ed. {3, 26} included in lymphoid proliferations and lymphomas associated with immune deficiency and dysregulation if appears in this setting*

between EBV-positive and EBV-negative IDD-lymphomas. This indicates the possibility of biological differences between lymphomas in immunocompetent patients and IDD patients, as well as between EBV-positive and EBV-negative lymphomas in the IDD setting [4].

Epstein-Barr virus related lymphomas are generally more common in Asia than in Europe. The prevalence of EBV+ diffuse large B-cell lymphomas (DLBCL) is 7.9% (95% CI: 6.2–10.0%) [5] with a significantly higher incidence in Asia and South America (28% of DLBCL) compared to Western countries (< 5%). In Poland, EBV+ cases accounted for 12% of DLBCL [6]. Endemic BL occurs mainly in equatorial Africa and Papua New Guinea and is closely associated with *Plasmodium falciparum* (malaria) infection, which is thought to impair immune control of EBV [7]. Epstein-Barr virus associated natural killer (NK)/T-cell lymphomas account for 30% of NK/T-cell lymphomas in Asia and only 8% in Europe and North America. Approximately 25–30% of classic Hodgkin lymphoma cases are EBV-positive [5].

Epstein-Barr virus infection in early childhood is usually asymptomatic. In adolescents and young adults, it can manifest as symptoms including fever, painless swollen glands, and sore throat, known as infectious mononucleosis. Epstein-Barr virus infected B-cells proliferate and differentiate *via* an atypical germinal center-type reaction. The virus then establishes a persistent and antigenically silent infection in memory B-cells. Latent virus is maintained in a circular, chromatinized (episomal) state within the host cell nucleus but does not integrate with host DNA. The host cell provides the machinery that regulates the viral genome's replicative capability during the cell cycle. The lifelong latent infection of B-cells is characterized by its ability to immortalize these cells *in vitro*. There are four distinct latency programs, each characterized by a specific pattern of viral gene expression. These programs directly induce B-cell

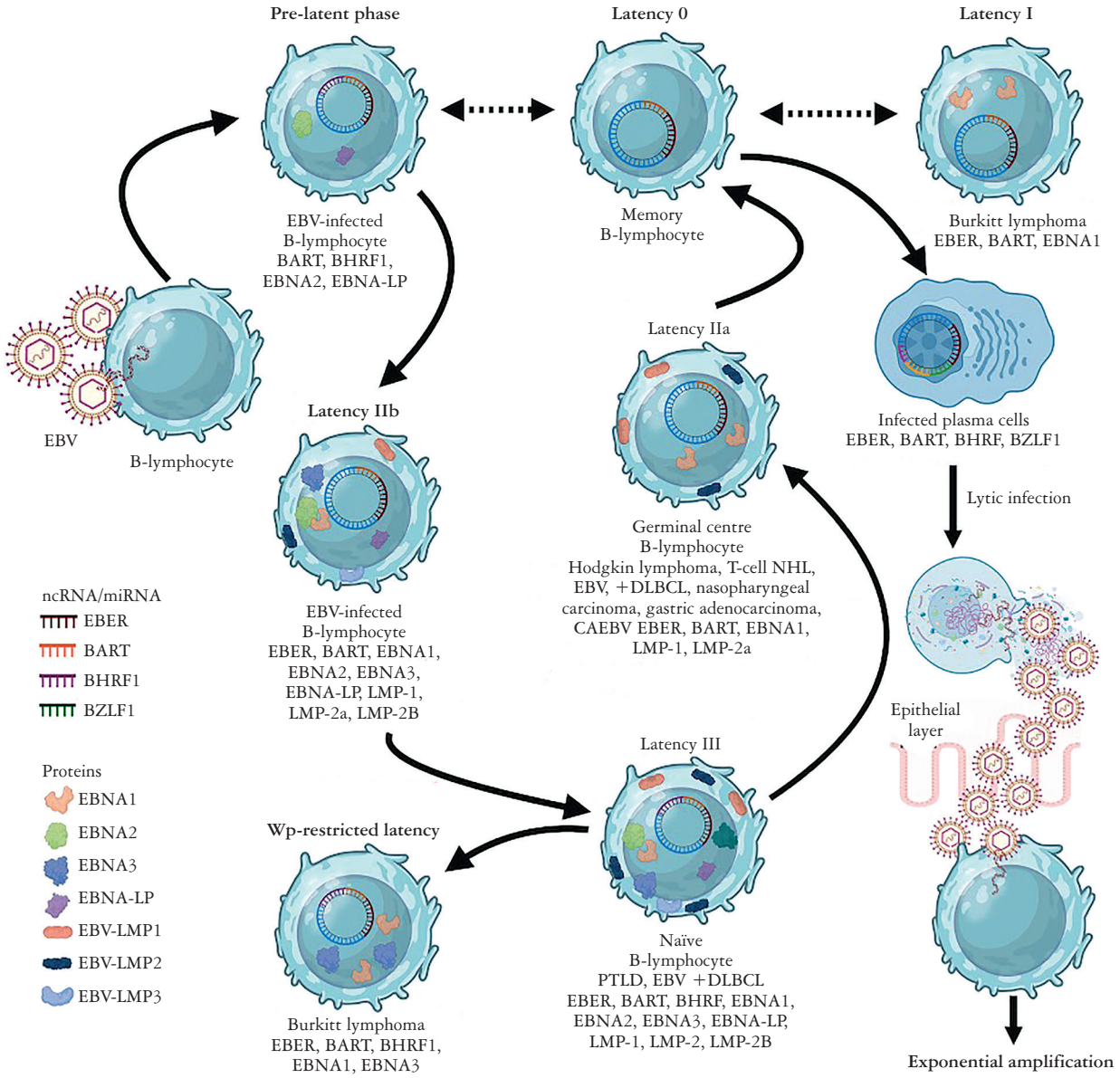


**Figure 1.** Cycle in B-cells, beginning with viral entry through the oropharyngeal epithelium and progression from lytic replication to multiple latency states (0, I, IIa, IIb, III) [9]

*BARTs – BamHI-A rightward transcripts, BHRF – BamHI H rightward open reading frame, BZLF – BamHI Z fragment leftward open reading frame, EBER – Epstein-Barr virus encoded RNA, EBV – Epstein-Barr virus, lmp – latent membrane protein, EBNA – Epstein-Barr nuclear antigen, EBNA-LP – Epstein-Barr nuclear antigen leader protein*
*Color-coded symbols represent EBV proteins and non-coding RNAs, including Epstein-Barr nuclear antigen 1-6, latent membrane protein (LMP) 1, LMP2A/2B, EBER, BHRF1, BARTs, and EBV miRNAs. Arrows show movement of infected cells from naïve B-cells through germinal centers into memory – chronic active EBV disease or lymphoblast stages. Each latency phase is linked to specific diseases (Table I) such as Burkitt lymphoma, Hodgkin lymphoma, nasopharyngeal carcinoma, CAEBV, post-transplant lymphoproliferative disorders, and EBV-positive diffuse large B-cell lymphomas, with lytic reactivation shown during plasma cell differentiation.*

transformation through host interactions and utilize numerous mechanisms to control B-cell activation and differentiation, alter cellular gene transcription, and constitutively activate key cell-signaling pathways (Figure 1) [8, 9].

Epstein-Barr virus encoded RNA (EBER) is a small, non-coding RNA expressed at high copy numbers in virtually all EBV latency programs, making it an excellent molecular marker for the presence of EBV-infected cells. Epstein-Barr nuclear antigen 1 (EBNA1) helps maintain the viral genome in infected cells and is one of the few latent proteins consistently seen across EBV-associated tumors. There are two major EBV genotypes in humans: EBV1 and EBV2 [10], distinguished by the differences in the *EBNA2* gene. Epstein-Barr virus 1 and EBV2 differ in their ability to transform, with EBV1 being more efficient in immortalizing B-cells *in vitro* than EBV2.

Latent membrane protein 1 (LMP1) is the major transforming protein in many EBV-associated lymphomas. By clustering and forming higher-order oligomers in the plasma membrane, LMP1 enables

the efficient recruitment of downstream signaling proteins, specifically tumor necrosis factor receptor-associated factors (1–5), and it activates signaling pathways that promote growth and survival. Latent membrane protein 1 is especially relevant in classical Hodgkin lymphoma (cHL) and some DLBCL. Latent membrane protein 2A mimics B-cell receptor signaling, helping infected B-cells avoid apoptosis and supporting lymphomagenesis, particularly in cHL and other B-cell lymphomas. Latent membrane protein 2B can modulate LMP2A activity and is less often emphasized as a direct oncogenic driver [8, 9].

Latency I is the most restrictive latency program, expressing only EBER1, EBER2, and EBNA1, and is typically associated with BL. Latency II displays EBER1, EBER2, EBNA1, LMP1, and LMP2A and is exemplified by cHL. Latency III exhibits the most complex viral expression pattern, characterized by EBER1, EBER2, EBNA1, EBNA-LP, EBNA2, EBNA3A/B/C, LMP1, LMP2A, and LMP2B. Latency III pattern is seen *in vitro* in EBV-transformed lymphoblastoid cell lines and is observed in many cases of B-cell *post* transplant lymphoproliferative disorders (PTLD). Persistence of EBV in the quiescent memory B-cell pool without viral protein expression (hence antigenically silent) is termed latency 0, which is a feature of healthy B-cells but not lymphomas, except for plasmablastic lymphoma (PBL). Epstein-Barr virus itself does not directly cause gene mutations; however, its chronic presence and the activity of its latent proteins can indirectly contribute to genomic instability [8, 9].

Despite the activation of a robust host T-cell response upon primary infection and the capacity for a memory T-cell response thereafter, EBV persists for life even in immunocompetent individuals. Chemical agents, external stress stimuli, and certain infectious agents may reactivate EBV. In addition, acute infection with other pathogens may enable EBV reactivation [11]. Upon reactivation, during the new lytic phase, many viral antigens are available for proteasomal processing and human leukocyte antigen Class I presentation at the cell surface. Subsequently, memory CD8+ T-cells capable of eliminating the infected cells are recruited. In immunocompromised individuals, reactivation occurs primarily due to a breakdown in T-cell surveillance (specifically CD8+ and Vδ2+ T-cells), allowing latent episomes in B-cells to enter the lytic cycle. Moreover, EBV expresses proteins that directly interfere with major histocompatibility complex (MHC) presentation. The viral protein BILF1 (BamHI I fragment Leftward Open reading frame 1) acts as a functional homolog of the viral interleukin-10 receptor, suppressing antigen presentation and reducing immune recognition. Additionally, EBV's EBNA proteins can downregulate MHC Class I molecules on infected B-cells, preventing cytotoxic T lymphocyte recognition. The BNLF2a protein specifically inhibits transporter associated with antigen processing (TAP), blocking the transport of antigenic peptides to the MHC Class I pathway. Other immune evasion mechanisms include inhibition of apoptosis, promoting cell proliferation, reducing NK and myeloid cell activity, suppressing cytokine expression, and disrupting key host immune recognition molecules [12].

In hematopathology practice, EBV is best confirmed by EBER *in situ* hybridization (ISH), while immunohistochemistry (IHC) is used as a complementary test for the presence of EBV-related proteins. In this review, summarizing a lecture given at the 10th Course of the Academy of Immunohistochemistry in Cracow, Poland (22–24 June 2026), some challenging examples of EBV-related lymphomas are presented and discussed.

## Examples of Epstein-Barr virus-related B-cell lymphomas

### Case 1

A 44-year-old male presented with an ulceration of approximately 5 cm in diameter in the gingiva. The patient had been suffering from ulcerative colitis for more than 10 years. Six months ago, treatment with infliximab every 8 weeks was started. Previous history included mononucleosis at the age of 15 and recurrent pneumonias, the latest a month ago. The biopsy was consistent with EBV-related mucocutaneous ulcer, and the lesion regressed after infliximab treatment was stopped. However, 3 months later, positron emission tomography showed a cavitating lesion in the left lower lung lobe. Bronchoscopy was nondiagnostic, but a transbronchial biopsy revealed EBV-related diffuse large B-cell lymphoma. The patient received cytostatics (R-CHOP) treatment and is in complete remission 3 years later.

Microscopically, the gingival biopsy (Figure 2A) showed varying numbers of plasma cells and histiocytes with admixed large, atypical cells, including Hodgkin Reed-Sternberg (HRS)-like cells. The large, abnormal cells were positive for CD20, CD30, EBER, and MUM1, and were highly proliferative on Ki-67 immunostaining (> 80%).

The lung biopsy (Figure 2B) was small but revealed a heterogeneous infiltrate including groups of large cells, partly CD20 positive, positive for CD30, and EBER. A clonal *IGK* gene rearrangement was detected in the gingiva, but *IGH* showed an oligoclonal pattern (Figures 2A, B). In the lung biopsy, both *IGH* and IGK were clonally rearranged (Figures 2C, D), showing a different product size in *IGK*.

Epstein-Barr virus positive mucocutaneous ulcer (EBV+ MCU) was first described by Dojcinov *et al*. [13] and included in WHO 2016 (4th ed.) classification as a provisional entity [14]. In WHO 2022 (5th ed.) classification, EBV+ MCU is included as an entity

within *Lymphoid proliferations and lymphomas associated with immune deficiency and dysregulation* [3]. Epstein-Barr virus positive MCU are solitary, mucosal, or cutaneous ulcers (mainly in the oral mucosa, tonsils, palate, and gastrointestinal tract), occurring in immunosuppressed subjects (primary immunodeficiency, solid organ transplantation, HIV, and iatrogenic immunosuppression). In elderly patients, EBV+ MCU may be due to immune senescence. The median age at diagnosis is 70 years, but EBV+ MCU can also occur in younger immunosuppressed patients. Patients do not have generalized lymphadenopathy, organomegaly, or bone marrow involvement.

The ulcers are shallow and sharply circumscribed. The underlying infiltrate is polymorphous, displaying a mixture of lymphocytes and immunoblasts with many large, pleomorphic Hodgkin and Reed-Sternberg-like cells. Scattered plasma cells, histiocytes, and eosinophils are generally present. Angioinvasion, thrombosis, and necrosis are seen. The large, atypical cells are positive for CD30 and MUM1 with co-expression of CD15 in half of the cases, reduced or negative CD20 in one-third of cases, negative CD10, and variable expression of PAX5, OCT2, Bob-1, and BCL6. Other lymphoid cells positive for CD20, CD30, OCT2, and PAX5 may vary in size.

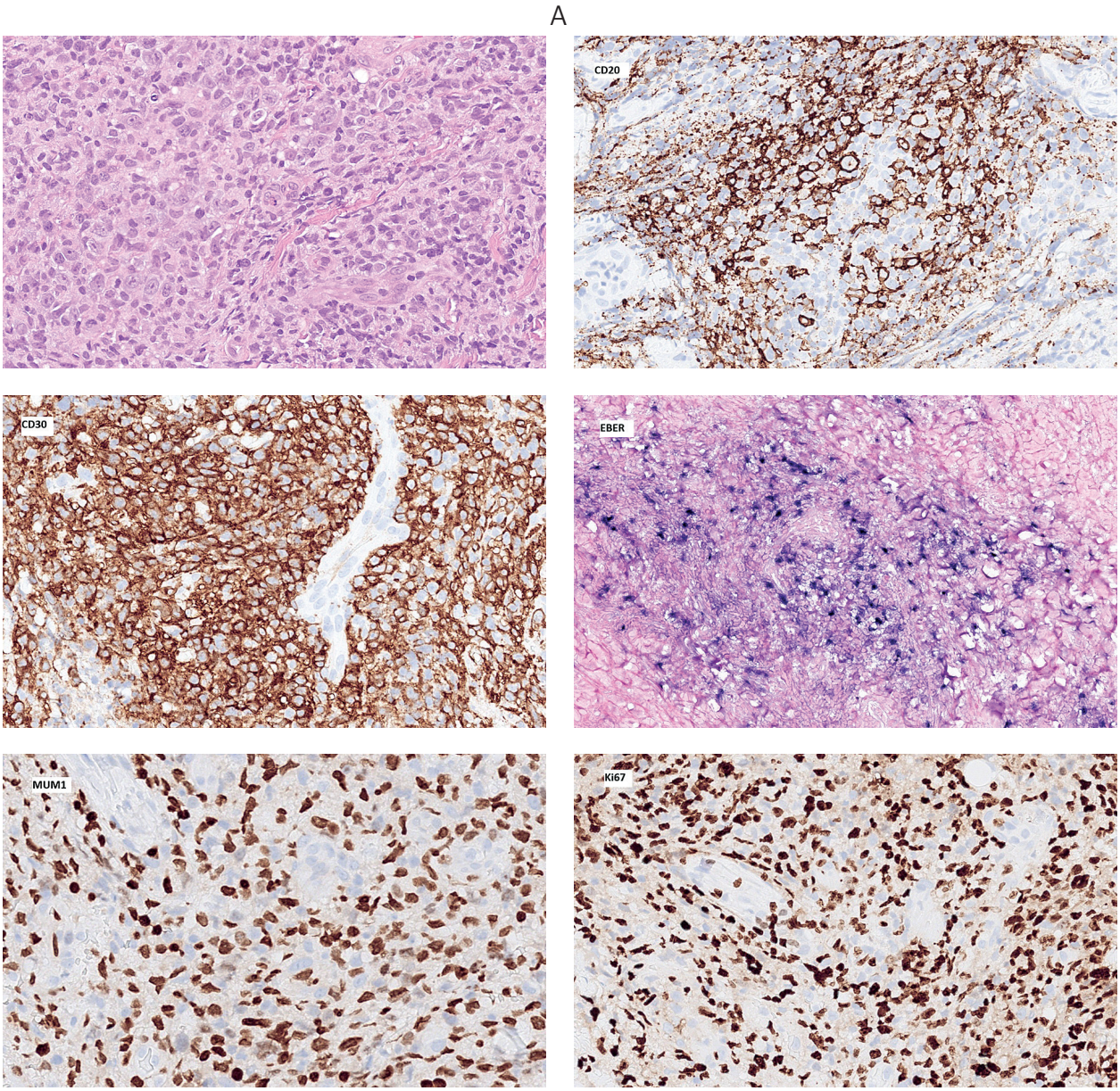


**Figure 2.** Epstein-Barr virus (EBV)-positive mucocutaneous ulcer and subsequent EBV-positive diffuse large B-cell lymphoma in a patient treated with tumor necrosis factor α inhibitor (Case 1). **A**) Mucocutaneous ulcer with large, atypical cells that are highly proliferating (Ki-67) and positive for CD20, CD30 and Epstein-Barr virus encoded RNA (EBER)

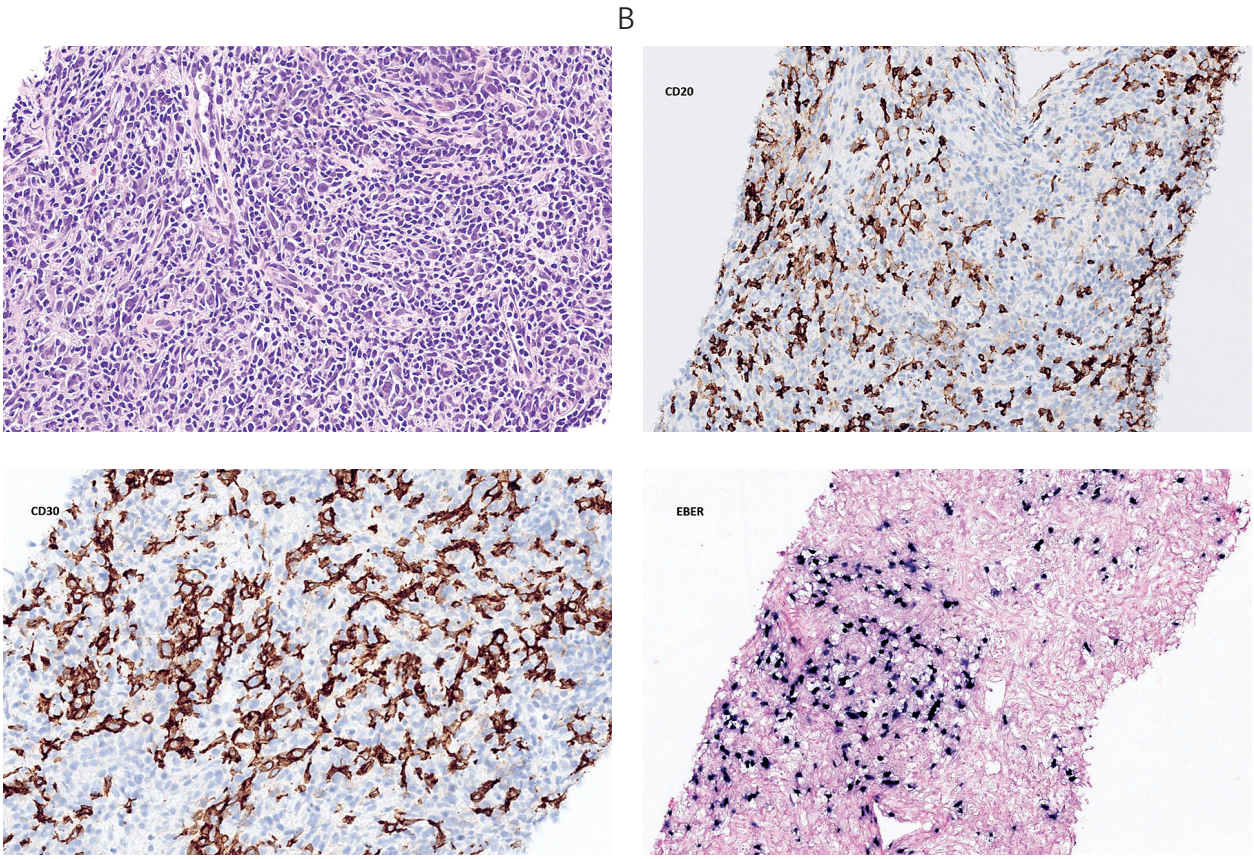


**Figure 2.** Cont. **B**) Small biopsy from lung tissue showing presence of large atypical cells positive for CD20, CD30 and EBER

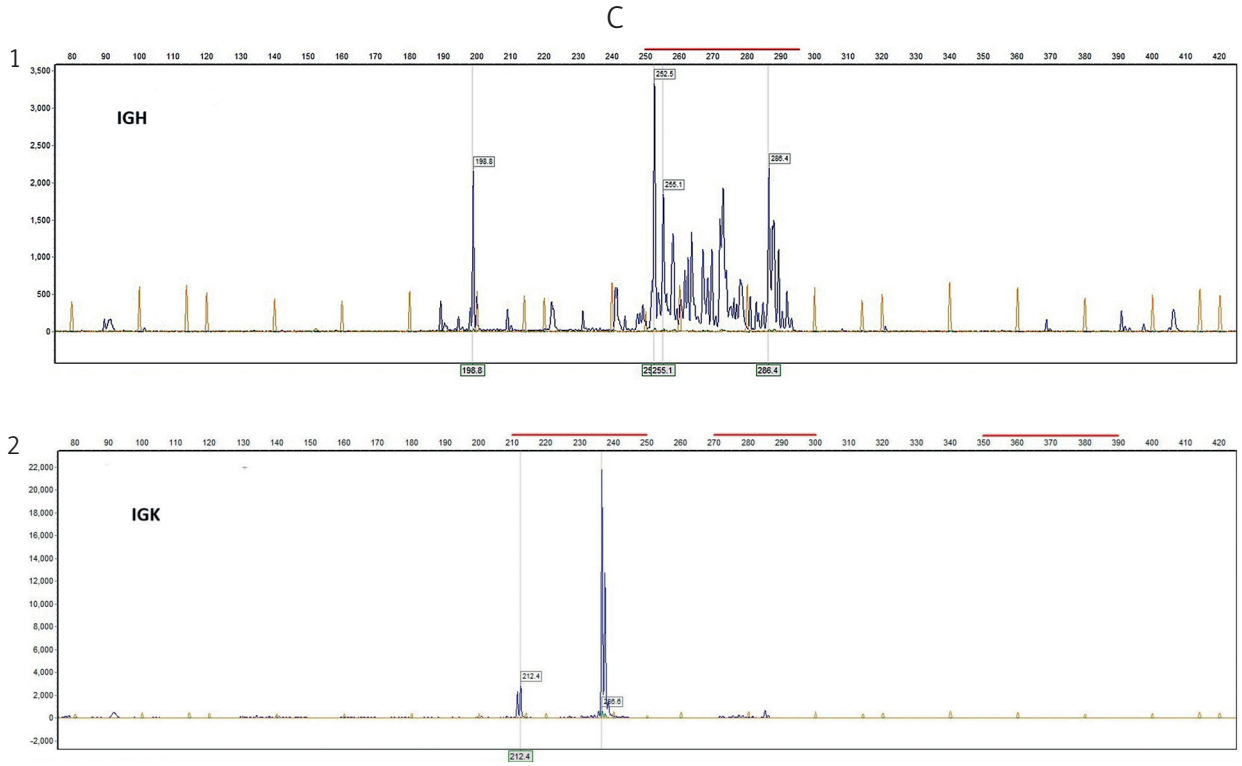


**Figure 2. C1, 2**) Polymerase chain reaction analysis of mucocutaneous ulcer

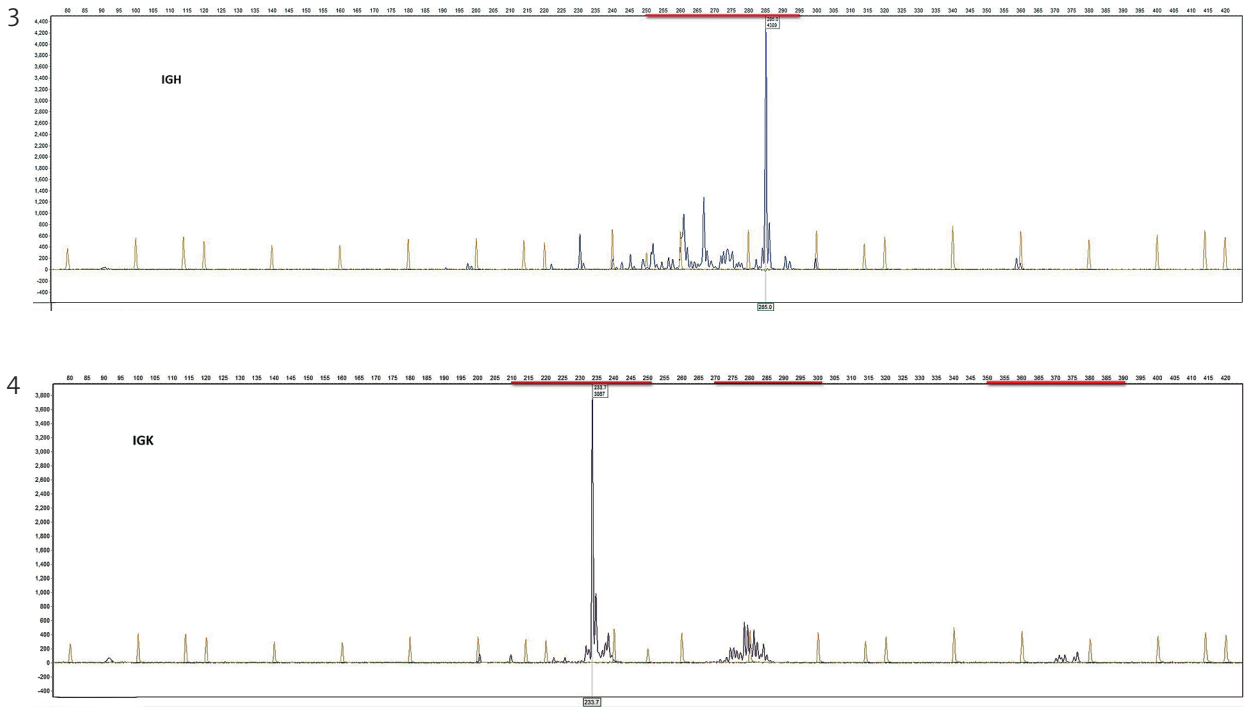


**Figure 2. C3,** 4) Polymerase chain reaction analysis of diffuse large B-cell lymphoma

*Figure C1, C2 and lung biopsy (C3, C4) showed a different pattern of IGH and IGK rearrangements.*

Both CD4+ and CD8+ T-cells are present with a rim of T-cells at the base of the ulcer. Epstein-Barr virus encoded RNA *in situ* hybridization is positive in large, pleomorphic cells and in many cases also in small lymphocytes. Epstein-Barr virus latency is type II in 50% and type III in 50% of cases. B-cell clonality can be demonstrated in 50% of cases. Oligoclonal or restricted patterns of T-cell receptor (TCR) rearrangement may be seen, indicating compromised immune surveillance. In most patients, spontaneous regression, or regression after reduction of immunosuppression is observed. A very good effect of single rituximab therapy is well-documented. In patients with primary immune deficiency, a more aggressive course requiring chemotherapy has been described. Development of a subsequent EBV-driven malignancy after regression of an $EBV^+$ MCU is rare, and only a few cases have been reported [15, 16]. As in our patient, the subsequent LPD were not clonally related to the primary $EBV^+$ MCU in reports with available molecular studies.

Epstein-Barr virus related DLBCL in a setting of immune deficiency or dysregulation are morphologically similar to those occurring in immunocompetent individuals. Epstein-Barr virus positive DLBCL are more often of non-germinal center cell than germinal center cell origin. Extranodal localization is a common feature [3].

## Case 2

An 82-year-old male underwent transurethral resection of a urinary bladder tumor. The patient had complex comorbidities related to advanced diabetes, nephropathy, and cardiovascular disease, but no previous history of malignancy or known immunodeficiency. Histology of urinary bladder biopsies showed an invasive papillary cancer G2 pT1m indicating early-stage disease that invades the bladder wall but has not reached the muscle layer at the left ostium, as well as non-invasive papillary cancer G2 pT1a at other sites. In several areas, stromal infiltrates of mature, CD138+, MUM1+, CD20–, EBER – plasma cells with polytypic expression of κ and lambda were present. Additional areas contained EBER-positive pleomorphic infiltrates of plasma cells, plasmablasts, immunoblasts, and centroblasts with monoclonal lambda expression (Figures 3A–C). Most cells in these infiltrates were MUM1- and CD79a-positive but showed heterogeneous expression of CD20, PAX5, CD30, and CD138 (Figures 3D–G) suggesting EBV-positive diffuse large B-cell lymphoma with plasma cell differentiation. Proliferation was high (> 90%) (Figure 3H). The cells were negative for HHV8, CD10, CD5, CD56, CD117, cyclin D1, BCL6, BCL2, and C-MYC. Molecular analysis confirmed monoclonal rearrangement of IGH and IGK. The patient received only palliative treatment and died 5 months later.

Only a few EBV-positive DLBCL of the urinary bladder have been reported to date [17]. A comprehensive immunohistochemical panel is necessary for differential diagnosis with EBV-positive PBL, which can occur in both immunocompetent patients and those with immunodeficiency. Although PBL most commonly presents in the oral cavity and gastroin-

testinal tract, other extranodal and rare nodal presentations, including those in the genitourinary tract, have been documented. Histologically, PBL consists of sheets of immunoblasts and plasmablasts, typically expressing plasma cell-associated markers such as CD138, CD38, BLIMP1, and MUM1/IRF4. Unlike DLBCL, NOS, CD20 and PAX5 are usually not expressed in PBL. Plasmablastic lymphoma shows a type I latency pattern, whereas EBV-positive DLBCL, NOS typically shows predominance of type II and III latency patterns. Unlike EBV-positive DLBCL, which usually lacks MYC overexpression, PBL often demonstrates MYC overexpression and rearrangement [3].

**Case 3**

A 72-year-old male previously treated with chemotherapy (4 courses of daunorubicin and cytarabine) for NPM1-positive acute myeloid leukemia (AML) has been in remission for the last 5 years. He presented with hypercalcemia, lymphadenopathy, splenomegaly, and tumor infiltration in the spinal canal. Laboratory findings showed pancytopenia (Hb 91 g/l, white blood cell count $2.4 \times 10^9$/l, platelets $100 \times 10^9$/l), but bone marrow aspiration and biopsy revealed no evidence of AML relapse or lymphoma infiltrates. Multiple core biopsies from a 9 cm tumor in the left pelvis were obtained. Flow cytometry on one sample demonstrated a κ-monoclonal B-cell population expressing CD19+, CD20+, CD10+, CD79b+, and CD81+, with negative staining for CD5, CD23, CD25, and CD200. A subset of cells expressed CD11c (Figure 4). Core biopsies showed diffuse infiltration by CD19+ B-cells with blast morphology that were positive for C-MYC and EBER (Figure 4). The proliferation index was very high (Ki-67 > 90%) (Figure 4). Immunohistochemistry revealed BCL-6 positivity but negativity for BCL-2, MUM1, TdT, CD34, CD117, CD30, CD21, and CD23. Fluorescence *in situ* hybridization (FISH) confirmed t(8;14)(q24;q32) IGH::-MYC (Figure 4). The diagnosis of BL was established.

Burkitt lymphoma was the first neoplasm shown to be associated with a chromosomal translocation involving *IGH*, subsequently demonstrated to be partnered with the *MYC* oncogene [3]. Less than 10% of pediatric sporadic cases of BL in the Western world are EBV-associated, compared to approximately 20–30% of adult cases [18]. Immune deficiency-associated BL, seen most frequently in patients with HIV/AIDS, *post*-transplant recipients, and those with primary immune deficiencies, is EBV-associated in up to 40% of cases. Burkitt lymphoma typically exhibits type I latency of EBV infection; EBV-positive BL-cells are positive for EBER and EBNA1 but negative for LMP1. By IHC, BL is positive for B-cell markers CD19, CD20, CD22, CD79a, and PAX-5, as well as germinal center-associated markers such as CD10, BCL-6, HGAL, and MEF2B, while LMO2 is often negative [7]. BCL-2 and GCET1 may be weakly expressed in 20% of cases, and weak MUM1/IRF4 expression is found in 10–40%. SOX11 is negative in EBV-positive BL cases, whereas approximately half of EBV-negative cases are positive. SOX11 expression demonstrates mutual exclusivity with EBV positivity.

Based on mutation patterns identified through somatic mutation analysis and antigen selection studies, EBV-negative BL reflects early centroblasts that underwent an initial mutation round and reached the subsequent mutation-silent phase prior to antigen selection, characterized by a low number of somatic mutations, lack of ongoing mutations, and absence of antigen selection evidence. In contrast, EBV-positive BL reflects late germinal center B-cells that have begun differentiation into memory B-cells, with a higher number of somatic mutations and evidence of antigen selection without ongoing mutations. Epstein-Barr virus positive BL frequently harbors ataxia telangiectasia mutated/ataxia telangiectasia and Rad3-related kinase pathway alterations, while EBV-negative cases often demonstrate *TCF3*/*ID3*/*CCND3* pathway mutations, *TP53* mutations, and *CDKN2A* homozygous deletions [7].

*Post* therapy BL is rare, representing less than 10% of PTLD, with 40% being EBV-related [19]. Only isolated cases of EBV-positive diffuse large B-cell lymphoma after chemotherapy for AML without stem-cell transplant have been described. However, some patients treated with chemotherapy may experience long-term immune defects after treatment ends; therefore, this case should be considered as belonging to the category *Lymphomas associated with immune deficiency and dysregulation* [3].

**Case 4**

An 83-year-old male patient with chronic venous insufficiency presented with nodules on the left lower leg. The nodules were not painful but developed ulcerations. A 3 mm punch biopsy was taken and showed intact epidermis with slight lymphocyte exocytosis and pleomorphic lymphoid infiltrate reaching deep in the dermis (Figure 5A). The lymphoid cells were predominantly medium sized with irregular nuclei and prominent nucleoli (Figure 5B). Most cells were positive for CD3 and CD56 (Figures 5C, D) but negative for CD2, CD5 (Figure 5E), CD4, CD8, CD7, CD30, TCR, and CD57, as well as for the B-cell markers. Cytotoxic markers TIA1 and granzyme B were positive (Figure 5F). Epstein-Barr virus encoded RNA was positive in > 80% of cells (Figure 5G). Proliferation was > 90% (Figure 5H). CD4 was positive in many histiocytes. Polymerase chain reaction (PCR) analysis showed monoclonal rearrangement of TCR β and γ. A diagnosis of a cutaneous variant of extranodal NK/T-cell lymphoma (ENKTL) with dermal nodular infiltration pattern was made [3, 20].

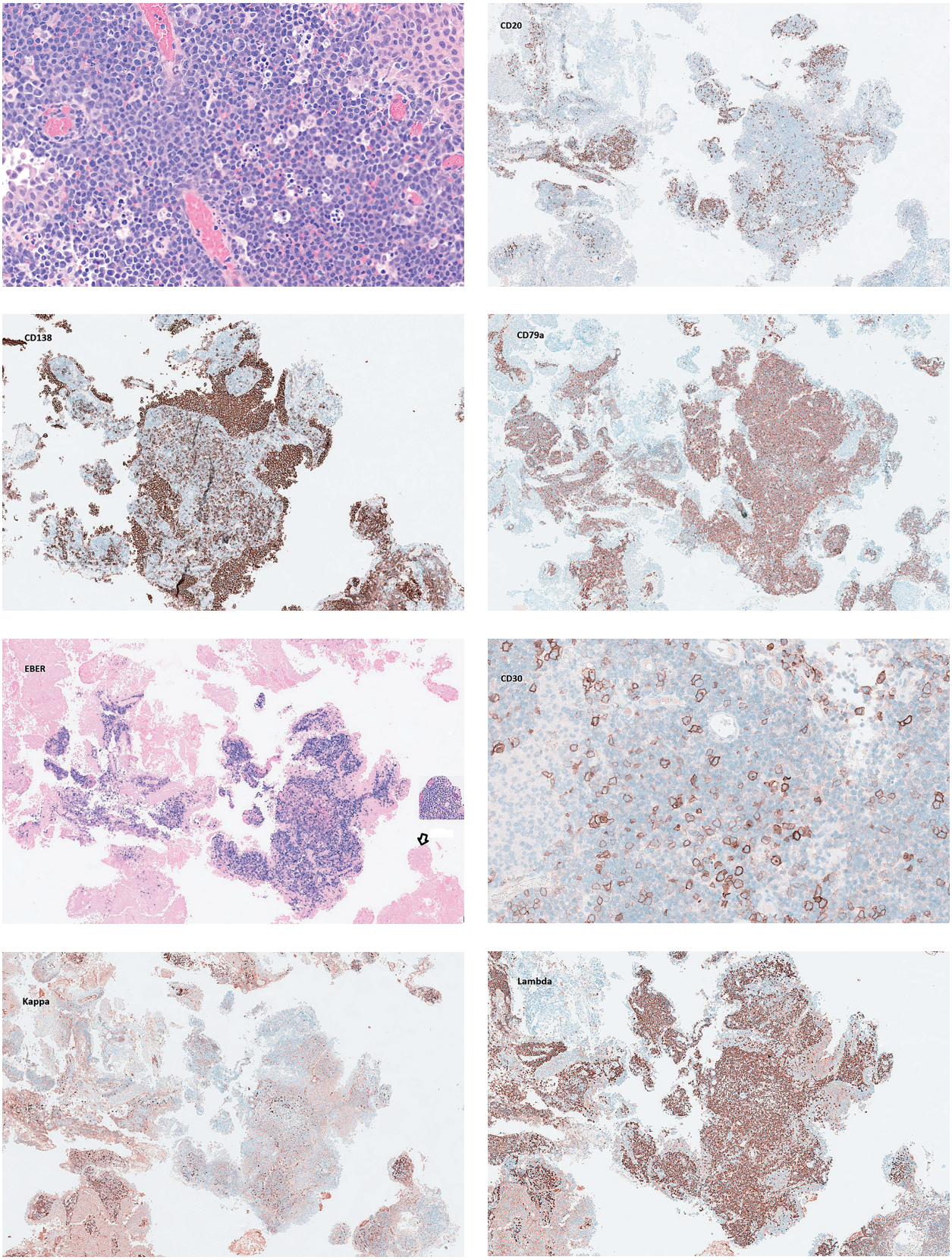


**Figure 3.** Epstein-Barr virus positive diffuse large B-cell lymphoma with plasma cell differentiation in the urinary bladder (Case 2)

*HE sections shows "starry sky" pattern and many plasmablasts. The lymphoma cells are partly CD20+, CD30+, CD138+, positive for CD79a, Epstein-Barr virus encoded RNA (EBER) and monoclonal for lambda. The arrow points out EBER negative areas with predominance of mature polyclonal plasma cells.*

Extranodal NK/T-cell lymphoma was first described by McBride in 1897 as causing rapid destruction of the nose and face at the midline, with progressing necrotic granuloma. This disease was subsequently termed "rhinitis gangrenosa progressiva" in Europe or "lethal midline granuloma" in the United States. Finally, in the late 1980s, the entity was called extranodal NK/T-cell lymphoma, nasal type, and EBV association was reported [21]. In the current WHO classification [3], this entity is called ENKTL with two clinical subtypes: nasal and non-nasal ENKTL. Extranodal NK/T-cell lymphoma predominantly occurs in eastern Asia and among the indigenous peoples of Central and South America, while the incidence in Europe and North America is low (< 1% of lymphoma cases). Hemophagocytic lymphohistiocytosis is a recognized and severe complication of ENKTL, reported in approximately 7–13% of patients [22].

Primary skin presentation of ENKTL is rare (10–20% of non-nasal cases). Occult involvement of the nasopharynx has been reported in some cases.

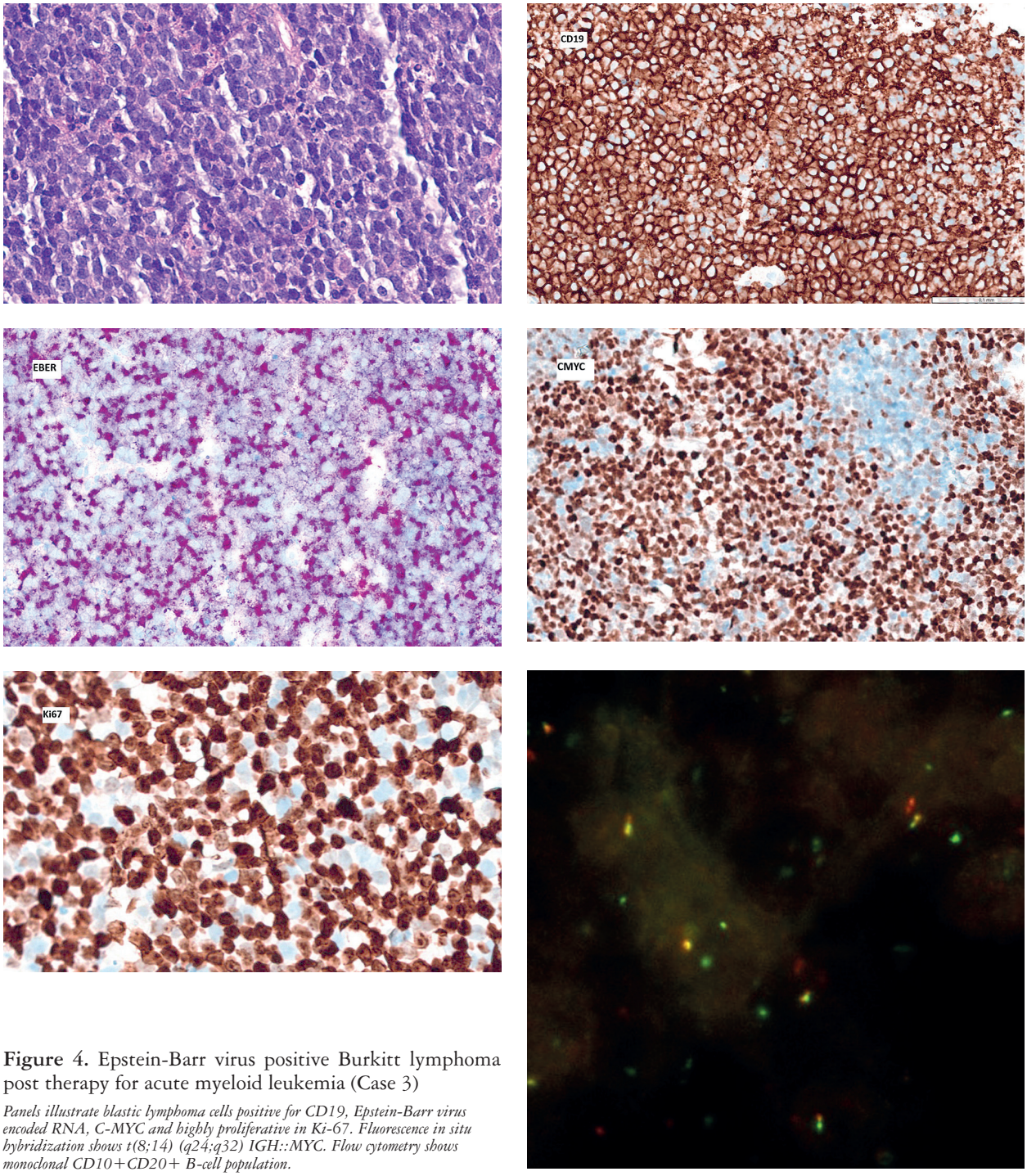


**Figure 4.** Epstein-Barr virus positive Burkitt lymphoma post therapy for acute myeloid leukemia (Case 3)

*Panels illustrate blastic lymphoma cells positive for CD19, Epstein-Barr virus encoded RNA, C-MYC and highly proliferative in Ki-67. Fluorescence in situ hybridization shows t(8;14) (q24;q32) IGH::MYC. Flow cytometry shows monoclonal CD10+CD20+ B-cell population.*

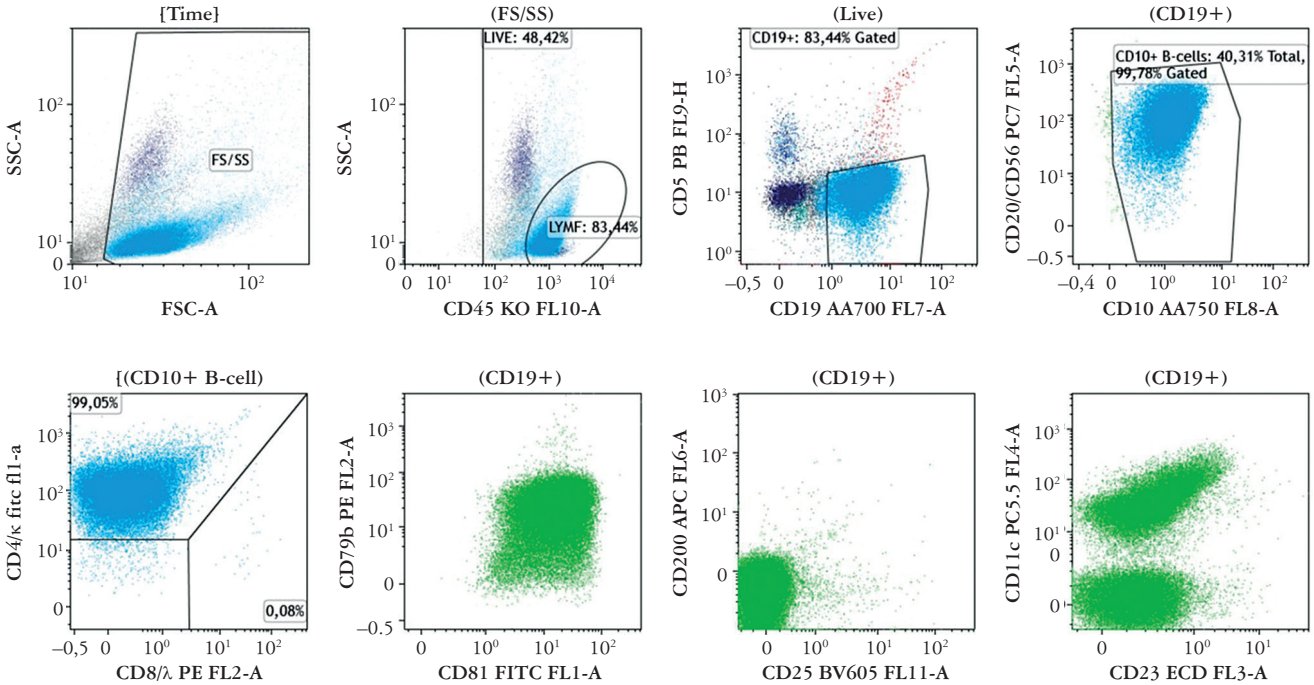


**Figure 4.** Cont. Flow cytometry findings (Case 3)

Four histopathological patterns of primary cutaneous ENKTL have been described [20]. The lobular panniculitis pattern is characterized by infiltration of subcutaneous fat by atypical cells surrounding fat lobules. The interface dermatitis pattern is defined by dermal infiltration of lymphoma cells with the overlying epidermis showing basal vacuolar changes and some necrotic keratinocytes. The granulomatous dermatitis pattern requires the presence of granulomas. In the dermal and subcutaneous nodular infiltration pattern, there is atypical lymphatic infiltration deep in the dermis or subcutaneous fat. All cases of ENKTL are positive for EBER. Published latency studies are mostly molecular-based, and a proportion of cases appears to exhibit latency I based on high levels of transcripts from EBNA1 only [23]. Another fraction of cases shows latency II with high levels of transcripts from EBNA1, LMP1, LMP2A, LMP2B, BNRF1, BILF1, BALF2, BALF3, BALF4, BALF5, and BNLF2b, and a minor number of cases appears to be in latency III due to identification of high levels of transcripts from EBNA1, LMP1, LMP2A, and EBNA2. Immunohistochemically, ENKTL cells are usually CD56+, TIA1+, granzyme B+, perforin+, mCD3–, cyt-CD3+, CD4–, CD5–, CD8–, TCRαβ–, and TCRγδ–, with some cases positive for CD8 and/or TCR. CD2 and CD7 expression varies. Single cases with aberrant CD20 expression have been reported. CD30 is expressed in approximately 70% of cases and has been associated with better survival. Successful therapy with anti-CD30 antibody-based therapy has been reported [24].

## Conclusions

An integrated diagnostic approach to LPD now incorporates imaging, pathology, and molecular testing to construct a complete disease profile. Pathological diagnosis and classification of lymphomas require a comprehensive evaluation of morphological and immunophenotypic findings, and in some cases, must be complemented by FISH. Molecular analysis (such as next-generation sequencing) is increasingly being applied for mutation detection, gene expression analysis, and copy-number variation assessment. Epstein-Barr virus encoded RNA testing is essential for specific EBV-associated lymphoma subtypes and can be selectively used based on clinical suspicion and histopathology findings. *In situ* hybridization for EBER is the gold-standard technique for determining EBV status and may be complemented by a less sensitive method, such as IHC for the viral proteins LMP1 or EBNA1. More sensitive methods, such as qPCR, RNAscope, and single-cell droplet digital PCR, have revealed viral traces that could not be detected by conventional ISH in biopsies from lymphoma patients [25]. Therefore, the methodology and indications for EBV-testing may soon be revisited.

## Disclosures

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2. Assistance with the article: None.
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4. Conflicts of interest: None.

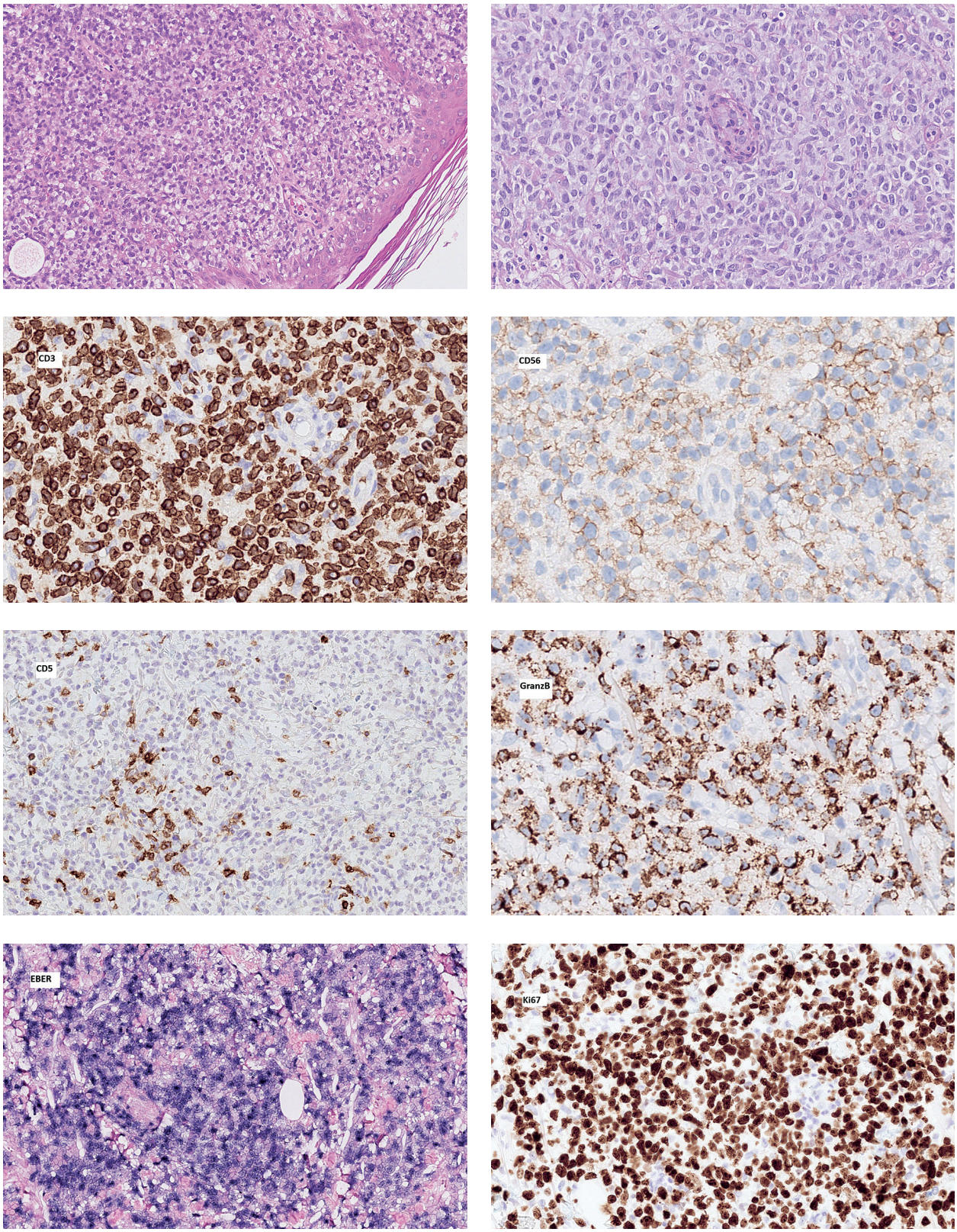


**Figure 5.** Extranodal NK/T-cell lymphoma, primary cutaneous variant (Case 4)

*Histology shows atypical medium sized cells with a sign of angioinvasion. The cells are CD3+, CD56 weakly+, CD5 negative, Epstein-Barr virus encoded RNA positive and highly proliferative.*

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