Journal of clinical and experimental hematopathology
Vol. 61 No.3, 173-179, 2021

Case report

Lymphocyte-depleted classic Hodgkin lymphoma with primary extranodal disease: Two cases that highlight the combination of immunodeficiency and immune escape in the pathogenesis

Yuta Tsuyuki,[1)] Kei Kohno,[1,2)] Yuichiro Inagaki,[3)] Yu Sakai,[4)] Hiroshi Kosugi,[5)] Emiko Takahashi,[6)] Yuka Suzuki,[1)] Satoko Shimada,[1)] Seiichi Kato,[7)] Taishi Takahara,[6)] Akira Satou,[6)] Yoshie Shimoyama,[1)] Shigeo Nakamura,[1)] Naoko Asano,[8)] and Ayako Sakakibara[1)]

Neoplastic programmed cell death ligand 1 (PD-L1) expression, activated by *PD-L1* gene alterations, is strongly associated with classic Hodgkin lymphoma (CHL). This association enabled a diagnostic consensus for lymphocyte-depleted CHL (LD-CHL), a previously enigmatic disease. We describe two patients with LD-CHL and primary extranodal disease. One patient was a 92-year-old female (Case #1) with a large mass that involved the uterus combined with swollen lymph nodes in the pelvic cavity. The second patient was a 76-year-old female (Case #2) with human T-cell leukemia virus type 1 (HTLV-1) who initially exhibited massive bone marrow involvement without peripheral lymphadenopathies. Biopsies of these tumors from the cervix uteri and bone marrow, respectively, revealed lesions rich in Hodgkin and Reed-Sternberg (H-RS) cells and diminished populations of other cell populations. Immunohistochemistry demonstrated that these H-RS cells expressed CD30, BOB1, and fascin, but not CD15, CD20, PAX5, or OCT2. They also expressed PD-L1, which led to our preferred diagnosis of LD-CHL in both patients. Epstein-Barr virus was associated with LD-CHL in Case #1, but not in Case #2. Both patients were deemed too frail for treatment. They died of disease at 1 (Case #1) and 15 months (Case #2) after the diagnosis. These findings highlight the abnormal biological behavior of this immune-escape-related lymphoid neoplasm in patients with immunodeficiency due to immune senescence and HTLV1 infection.

Keywords: programmed cell death ligand 1, lymphocyte-depleted classic Hodgkin lymphoma, primary extranodal disease, fragile elderly

INTRODUCTION

The lymphoma classification is evolving with the continuously deepening understanding of the biology of programmed death ligand 1 (PD-L1). This understanding is exemplified by the great success of immunotherapies that block PD-L1 in classic Hodgkin lymphoma (CHL).[1-3] Alterations in the *PD-L1* gene and protein expression are now regarded as a defining feature of CHL. This feature led to a diagnostic consensus that also includes lymphocyte-depleted CHL (LD-CHL). LD-CHL is an enigmatic disease with a highly variable histopathological appearance and widespread involvement.[2,4] We describe two patients with LD-CHL and primary extranodal disease that involved, in one case, the uterus, and in the second case, the bone marrow. The description of this abnormal disease pattern can shed light on the unique biological behavior of this immune-escape-related lymphoid neoplasm in hosts with immunodeficiencies.

Received: February 6, 2021. Revised: May 20, 2021. Accepted: June 21, 2021. Online Published: September 10, 2021
DOI: 10.3960/jslrt.21008

[1)]Department of Pathology and Laboratory Medicine, Nagoya University Hospital, Nagoya, Japan, [2)]Department of Pathology, Kurume University, School of Medicine, Kurume, Japan, [3)]Department of Hematology and Oncology, Anjo Kosei Hospital, Anjo, Japan, [4)]Department of Diagnostic Pathology, Anjo Kosei Hospital, Anjo, Japan, [5)]Department of Hematology, Ogaki Municipal Hospital, Ogaki, Japan, [6)]Department of Surgical Pathology, Aichi Medical University Hospital, Nagakute, Japan, [7)]Department of Pathology and Molecular Diagnostics, Aichi Cancer Center Hospital, Nagoya, Japan, [8)]Department of Clinical Laboratory, Nagano Prefectural Suzaka Hospital, Suzaka, Japan
Corresponding author: Yuta Tsuyuki, MD, Department of Pathology and Laboratory Medicine, Nagoya University Hospital, 65 Tsurumai-cho, Showa-ku, Nagoya, Aichi 466-8560, Japan. E-mail: tsuyukiyuta@med.nagoya-u.ac.jp

CASE REPORT

Case 1

A 92-year-old female initially presented with abnormal vaginal bleeding. Biopsy demonstrated polymorphous infiltration of lymphocytes with scattered Hodgkin and Reed-Sternberg (H-RS) cells, which was diagnosed as LD-CHL. Magnetic resonance imaging and other imaging methods, including transvaginal ultrasonography, revealed a tumor mass that measured 60 x 46 mm and affected the cervix uteri, corpus uteri, and parametrium. In addition, swollen lymph nodes were observed in the pelvic cavity and retroperitoneum (Fig. 1). At the physical examination, the superficial lymph node was unremarkable. Blood tests yielded a white blood cell count (WBC) of 8,400/μL, a hemoglobin (Hb) level of 10.8 g/dL, a platelet count of 26,100/μL, a ferritin level of 392 ng/mL, and a soluble interleukin-2 (IL-2) receptor level of 8,080 U/mL. The patient was deemed too frail for treatment and she died of the disease within 1 month of the diagnosis.

Case 2

A 76-year-old female complained of appetite loss with pleural effusion and ascites. Peripheral lymphadenopathy was not detected. Blood tests yielded a WBC of 16,810/μL, a Hb level of 8.8 g/dL, a platelet count of 168,000/μL, a ferritin level of 1,862 ng/mL, and a soluble IL-2 receptor level of 59,529 U/mL. She also carried human T-cell leukemia virus, type 1 (HTLV-1), based on the detection of anti-HTLV-1 antibodies in the blood. Positron emission tomography in tandem with computed tomography demonstrated hypermetabolic foci in the bone marrow and hepatic hilar lymph nodes. Atypical lymphocytes were not detected in the peripheral blood smear. The bone marrow biopsy sample exhibited a large lesion rich in tumor cells, which led to a definitive diagnosis of LD-CHL. The patient was deemed too frail for treatment and received only prednisolone without multi-agent chemotherapy, which resulted in clinical remission. However, she gradually developed multiple swollen lymph nodes of 21 x 16-60 x 41 mm in size and died of the disease after a clinical course of 15 months.

PATHOLOGICAL FINDINGS

Case 1

The biopsy specimen from the vagina exhibited diffuse infiltration of Hodgkin and Reed-Sternberg (H-RS) cells and/or pleomorphic large cells. Moreover, we observed an inflammatory background comprising a small-to-moderate number of eosinophils, small lymphocytes, and histiocytes distributed in a vaguely nodular pattern (Fig. 2A). These tumor cells featured a high degree of nuclear pleomorphism and/or irregular nuclear contours, with frequent binucleate and multi-lobed nuclear forms, and abundant cells with a pale cytoplasm (Fig. 2B). Necrosis was not observed.

Based on immunohistochemical analysis, the H-RS cells strongly expressed CD30 (Fig. 2C), BOB1, and fascin, but not CD15, CD20, PAX5 (Fig. 2D), OCT2, MUM1, or BCL6. PD-L1 (clones SP142, 28-8, and E1J2J) was also detected in >90% of the tumor cells (Fig. 2E). In addition, *in situ* hybridization with Epstein-Barr virus (EBV)-encoded small RNA probes (EBER) revealed that the tumor cells harbored EBV in the nuclei (Fig. 2F). The background cells mainly consisted of CD3+ small lymphocytes with a scattered distribution of FoxP3+ T regulatory cells (Fig. 2G).

No clonal rearrangement in the immunoglobulin heavy locus gene was found on polymerase chain reaction (PCR) analysis of available paraffin tissue sections, but indeterminate peaks were observed for the T-cell receptor γ gene. We assessed the copy number status of the *CD274/PD-L1* gene by fluorescence *in situ* hybridization using a Spec CD274 PDCD1LG2/CEN9 Dual Color Probe (Zytovision,

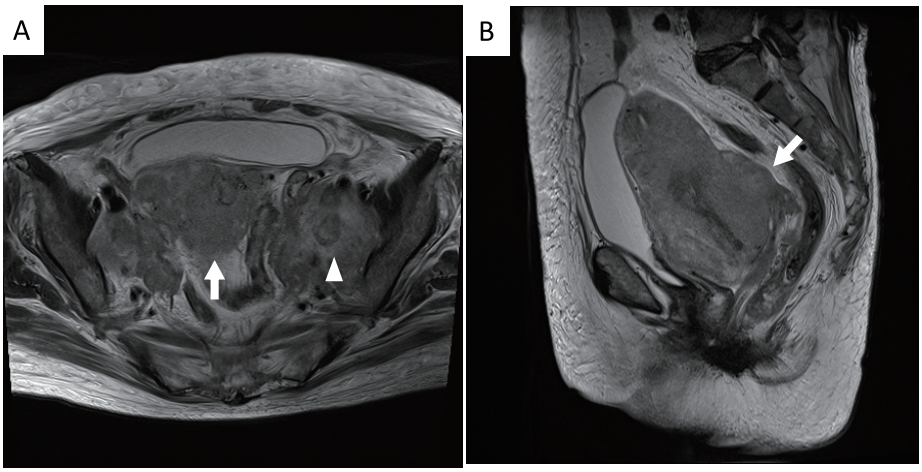

Fig. 1. T2-weighted magnetic resonance images show (***A***) axial and (***B***) sagittal views of a 92-year-old patient (Case #1). A tumor mass (arrows) affected the (*left panel*) cervix uteri, corpus uteri, and (*right panel*) parametrium, and (*left panel*) a swollen lymph node (arrowhead) was present in the pelvic cavity.

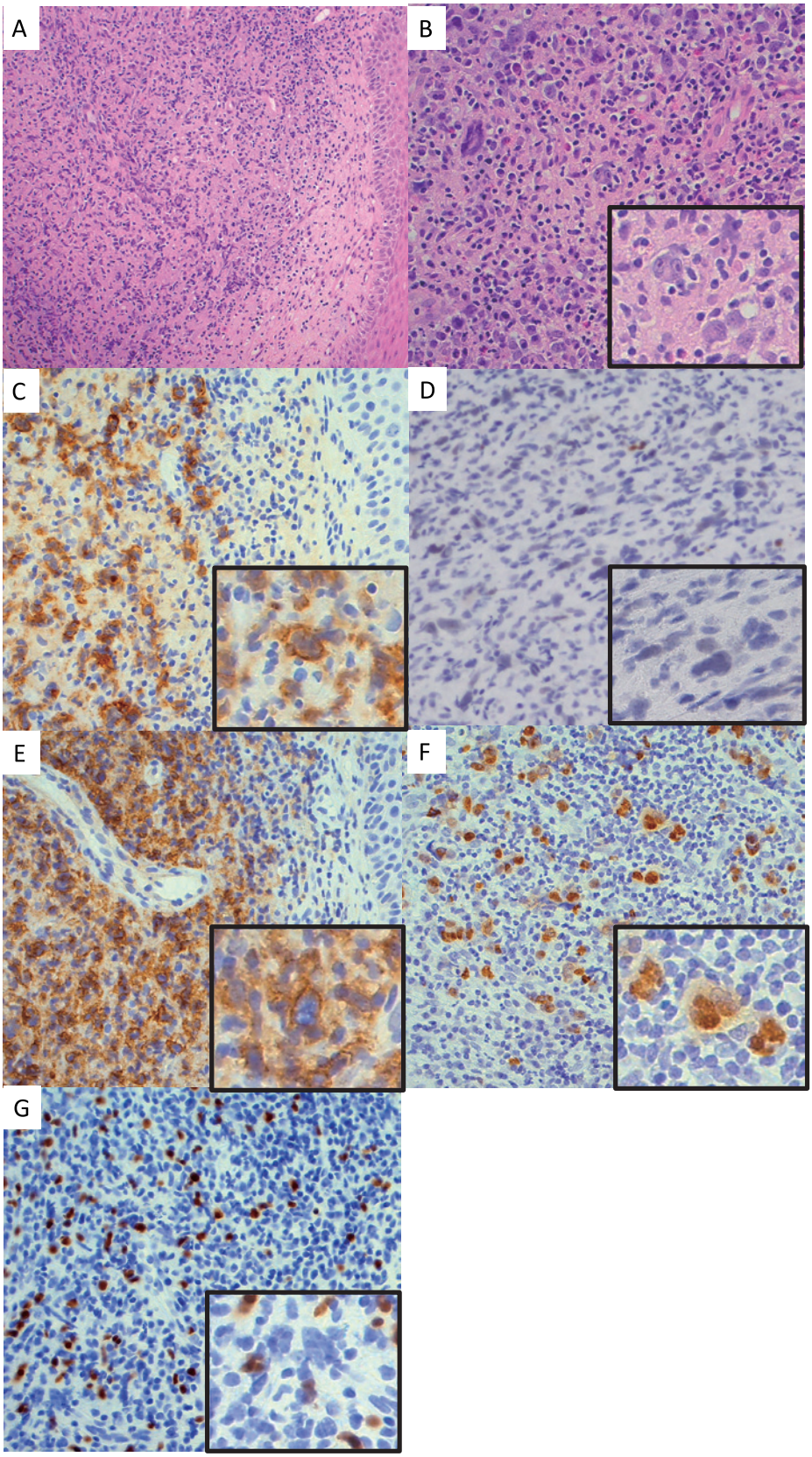

Fig. 2. Lymphocyte-depleted classic Hodgkin lymphoma that affected the cervix uteri (Case #1). Hematoxylin-eosin stained biopsy specimens show (***A***) pleomorphic large cells and (***B***) an abundance of Hodgkin and Reed-Sternberg cells. On immunohistochemistry, these biopsied tumor cells are strongly positive for (***C***) CD30, but negative for (***D***) PAX5. Tumor cells are positive for (***E***) PD-L1 (clone SP142), and (***F***) EBER. (***G***) FoxP3+ T-lymphocytes are scattered in the background.

Bremerhaven, Germany). Little or no gene amplification was observed in the H-RS cells in the lesion.

Case 2

A biopsy specimen of the bone marrow exhibited a large tumor cell-rich lesion with fibrosis (Fig. 3A). The histopathological morphology was somewhat similar to an anaplastic variant of adult T-cell leukemia/lymphoma (ATLL). Based on immunohistology, these candidate H-RS cells expressed CD30 (Fig. 3B), BOB1, and fascin (Fig. 3C), but not CD15, CD20 (Fig. 3D), PAX5 (Fig. 3E), OCT2, MUM1, or BCL6, similar to Case 1. PD-L1 expression (clone SP142) was detected in 30% of the tumor cells (Fig. 3F), and EBER *in situ* hybridization analysis confirmed that they were not associated with EBV. The background cells exhibited a scattered distribution of a limited number of CD3+ small lymphocytes (Fig. 3G), but these cells were negative for FoxP3. We did not perform PCR analysis due to the paucity of unstained sections.

DISCUSSION

We described two unique cases of LD-CHL with primary extranodal disease in addition to lymph node swelling. Case 1 presented as a large mass that involved the uterus. The patient died of the disease within 1 month of the diagnosis. Case 2 presented as a large bone marrow lesion rich in tumor cells in the context of HTLV-1. This case posed the diagnostic problem of differentiating between LD-CHL and ATLL because the incidence of bone marrow involvement in CHL is relatively low at 5-10%.[5] This patient achieved clinical remission by prednisolone therapy and had a relatively longer clinical course (15 months) than the patient in Case 1.

The combined morphological and phenotypical features of the tumor cells in both cases were consistent with those documented for prototypic CHL, including EBV positivity and neoplastic PD-L1 expression on H-RS cells. Our preferred diagnosis was LD-CHL for both cases, with primary extranodal disease in the immunosuppressed host.[1,4] These cases shed light on the abnormal biological behavior of B-cell neoplasms in such settings; i.e., the combination of neoplastic PD-L1 expression on tumor cells and host immunodeficiency due to immune senescence and HTLV-1 infection.

LD-CHL is defined as a diffuse form of CHL with abundant H-RS cells and/or a reduced number of other cell populations typical in CHL. LD-CHL is often mistaken for other entities such as aggressive forms of other B-cell lymphomas.[1,4] EBV is relatively frequent in LD-CHL, as observed in Case 1. In general, EBV-positivity should prompt the suspicion of EBV-positive diffuse large B-cell lymphoma (DLBCL), not otherwise specified, particularly in patients with bulky extranodal disease.[6-8] DLBCL is similar to immunodeficiency-associated B-cell lymphoproliferative disorders, and it often displays a broad range of CHL-like morphologic features, which poses the diagnostic problem of differentiating DLBCL from CHL.[6,9-14] We recently reported in a short series study that PD-L1 positivity is significantly lower in EBV+ tumors and non-malignant large B cells than in EBV+ CHL. Indeed, neoplastic PD-L1 expression was consistently detected in EBV+ CHL.[8,15] In another series, Kiyasu *et al*. noted neoplastic PD-L1 expression in 22/114 (19%) cases of EBV+ DLBCL.[16] Furthermore, Ishikawa *et al*. examined primary gastric DLBCL in patients with no prior history of lymphoma and found no (0%) patients with tumor cells that harbored EBV or expressed PD-L1.[17] In a later study, among 10 cases of primary intestinal EBV+ DLBCL, tumor cell PD-L1 expression was found in two (20%) cases. Moreover, 8 of the 10 cases had preceding iatrogenic immunodeficiency.[18] This suggested that PD-L1 immunostaining is particularly useful in the diagnosis of abnormal cases, such as CHL with primary extranodal disease, which exhibit morphological features that are observed in both EBV+ DLBCL and CHL. However, Chen *et al*. described frequent PD-L1 expression in EBV+ DLBCL samples from patients that were immunocompromised, aged, or had posttransplant LPD (100%, 100%, and 60%, respectively).[19] Nicolae *et al*. also reported a high frequency of PD-L1 expression (77%) in nodal EBV+ large B-cell lymphoma in young patients.[20] On the other hand, Takahara *et al*. recently documented that neoplastic PD-L1 positivity was detected on a minority (>5%) of the tumor cells in 6 (11%) of their EBV+ DLBCL patients aged >45 years old.[21] These discrepancies among studies may be explained by differences in ethnicity, cut-off values, and anti-PD-L1 antibodies, and should be clarified in the future.

In Case 2, LD-CHL developed primarily in the bone marrow of a patient that carried HTLV-1. This nodal disease presentation was distinct from that of Hodgkin-like ATLL.[22,23] Although HTLV-1 is not associated with the lymphomagenesis of B-cell neoplasms, HTLV-1 can theoretically cause T-cell dysfunction, which can compromise host immunity. This condition may play a role in the pathogenesis of LD-CHL with a primary extranodal disease in the absence of EBV.[24,25] Of note, detecting neoplastic PD-L1 expression in large cells by PD-L1 immunostaining was helpful in reaching a definite diagnosis of LD-CHL in the bone marrow, as previously emphasized by Xing *et al*.[5]

A primary extranodal presentation is rare in CHL.[1] Of note, CHL associated with human immunodeficiency virus (HIV) also exhibited abnormalities compared with CHL in an immunocompetent population. In the presence of HIV, CHL exhibited unusually aggressive clinical behavior with advanced extranodal disease. Moreover, patients with HIV had a high incidence of LD-CHL and an unusually large proportion of H-RS cells.[26-28] In previous studies, these features were not investigated well in patients with CHL lacking HIV infection.

In summary, the current study highlighted the abnormal biological behavior of immune-escape-related lymphoid neoplasms that developed in patients with immune senescence and/or HTLV-1 infection. Our stud emphasizes the utility of the PD-L1 immunostaining (clone SP142) for diagnosing LD-CHL. This disease affects older, fragile individuals, and

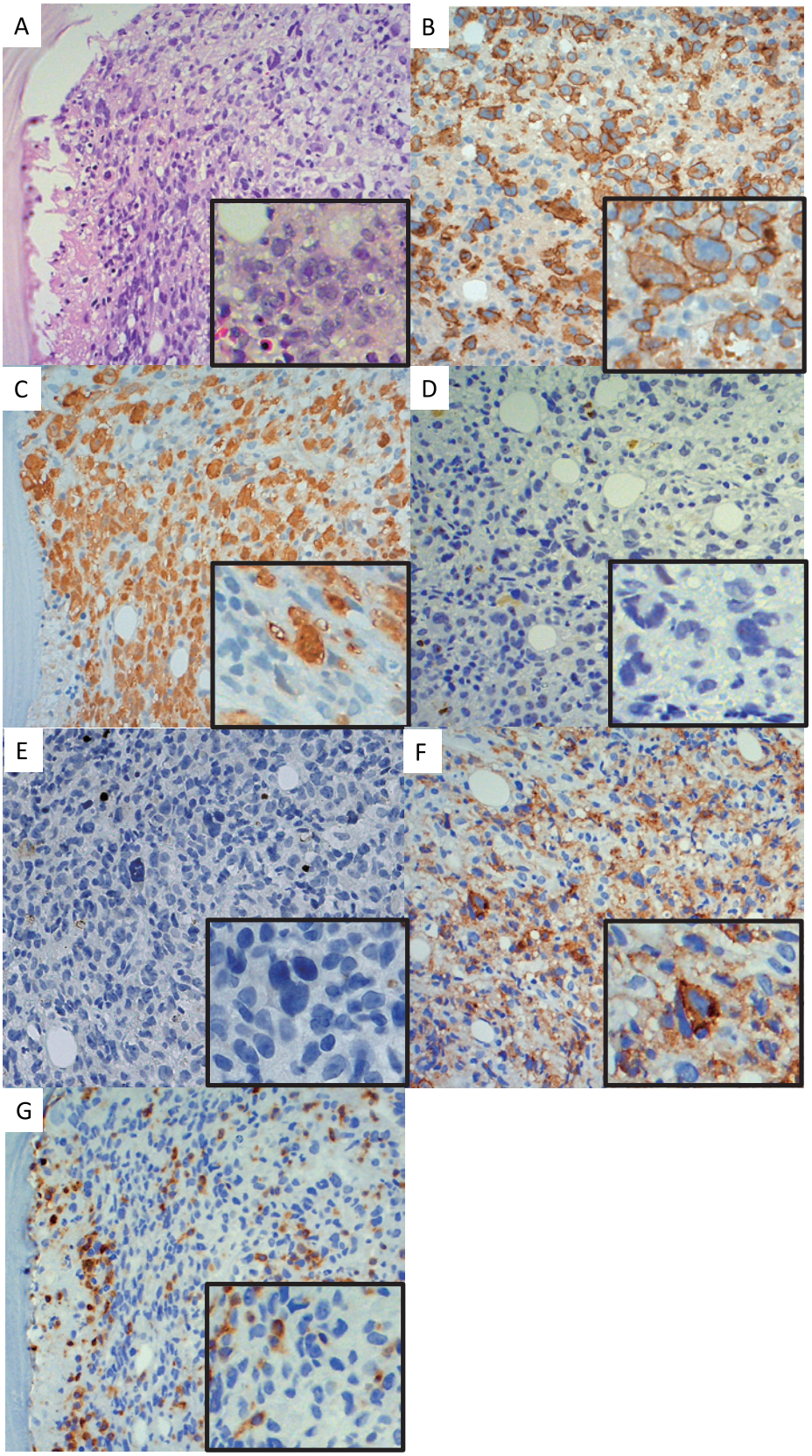

Fig. 3. Lymphocyte-depleted classic Hodgkin lymphoma in the bone marrow of a 76year-old patient that carried HTLV-1 (Case #2). Hematoxylin-eosin stained biopsy specimen shows (***A***) an abundance of large cells. On immunohistochemistry, these biopsied tumor cells are strongly positive for (***B***) CD30 and (***C***) fascin, but not (***D***) CD20 or (***E***) PAX5. (***F***) A subset of tumor cells showed PD-L1 (clone SP142) expression in the cytoplasm. (***G***) A small number of CD3+ small lymphocytes is scattered in the background.

its incidence is increasing in parallel with the aging society in Japan. Future studies are expected to clarify the abnormal biological behavior of LD-CHL developing in the fragile elderly.

ACKNOWLEDGMENTS

The authors would like to thank Y. Katayama, Y. Inagaki, K. Matsuba, and K. Kito for their technical assistance.

CONFLICTS OF INTEREST

The authors have no significant relationships with or financial interests in any commercial companies pertaining to this article.

REFERENCES

1 Jaffe ES, Stein H, Swerdlow SH. Classic Hodgkin lymphoma. In : Swerdlow SH, Campo E, Harris NL, *et al*. (eds) : WHO Classification of Tumours of Haematopoietic and Lymphoid Tissues. Lyon, IARC. 2017; pp. 425-442.
2 Roemer MGM, Advani RH, Ligon AH, *et al*. PD-L1 and PD-L2 genetic alterations define classical Hodgkin lymphoma and predict outcome. J Clin Oncol. 2016; 34 : 2690-2697.
3 De Goycoechea D, Stalder G, Martins F, Duchosal MA. Immune checkpoint inhibition in classical Hodgkin lymphoma: from early achievements towards new perspectives. J Oncol. 2019; 2019 : 9513701.
4 Wang HW, Balakrishna JP, Pittaluga S, Jaffe ES. Diagnosis of Hodgkin lymphoma in the modern era. Br J Haematol. 2019; 184 : 45-59.
5 Xing W, Mai N, Dresser K, Chen BJ. PD-L1 immunohistochemistry highlights bone marrow involvement by classic Hodgkin lymphoma in staging biopsies: implications for diagnosis and tumor microenvironment alterations. Appl Immunohistochem Mol Morphol. 2019; 27 : 356-363.
6 Oliveira CC, Paschoalini RB, Domingues MAC. Fas-ligand and caspase-3 positivity in three cases of histiocytic sarcoma: a different etiopathogenic pathway? Autops Case Rep. 2018; 8 : e2018001.
7 Nakamura S, Jaffe ES, Swerdlow SH. EBV positive diffuse large B-cell lymphoma, not otherwise specified. In : Swerdlow SH, Campo E, Harris NL, *et al*. (eds) : WHO Classification of Tumours of Haematopoietic and Lymphoid Tissues. Lyon, IARC. 2017; pp. 304-306.
8 Sakakibara A, Kohno K, Ishikawa E, *et al*. Age-related EBV-associated B-cell lymphoproliferative disorders and other EBV + lymphoproliferative diseases: new insights into immune escape and immunodeficiency through staining with anti-PD-L1 antibody clone SP142. Pathol Int. 2020; 70 : 481-492.
9 Oyama T, Ichimura K, Suzuki R, *et al*. Senile EBV+ B-cell lymphoproliferative disorders: a clinicopathologic study of 22 patients. Am J Surg Pathol. 2003; 27 : 16-26.
10 Oyama T, Yamamoto K, Asano N, *et al*. Age-related EBV-associated B-cell lymphoproliferative disorders constitute a distinct clinicopathologic group: a study of 96 patients. Clin Cancer Res. 2007; 13 : 5124-5132.
11 Shimoyama Y, Yamamoto K, Asano N, *et al*. Age-related Epstein-Barr virus-associated B-cell lymphoproliferative disorders: special references to lymphomas surrounding this newly recognized clinicopathologic disease. Cancer Sci. 2008; 99 : 1085-1091.
12 Asano N, Yamamoto K, Tamaru J, *et al*. Age-related Epstein-Barr virus (EBV)–associated B-cell lymphoproliferative disorders: comparison with EBV-positive classic Hodgkin lymphoma in elderly patients. Blood. 2009; 113 : 2629-2636.
13 Shimoyama Y, Asano N, Kojima M, *et al*. Age-related EBV-associated B-cell lymphoproliferative disorders: diagnostic approach to a newly recognized clinicopathological entity. Pathol Int. 2009; 59 : 835-843.
14 Dojcinov SD, Venkataraman G, Pittaluga S, *et al*. Age-related EBV-associated lymphoproliferative disorders in the Western population: a spectrum of reactive lymphoid hyperplasia and lymphoma. Blood. 2011; 117 : 4726-4735.
15 Sakakibara A, Kohno K, Eladl AE, *et al*. Immunohistochemical assessment of the diagnostic utility of PD-L1: a preliminary analysis of anti-PD-L1 antibody (SP142) for lymphoproliferative diseases with tumour and non-malignant Hodgkin-Reed-Sternberg (HRS)-like cells. Histopathology. 2018; 72 : 1156-1163.
16 Kiyasu J, Miyoshi H, Hirata A, *et al*. Expression of programmed cell death ligand 1 is associated with poor overall survival in patients with diffuse large B-cell lymphoma. Blood. 2015; 126 : 2193-2201.
17 Ishikawa E, Tanaka T, Shimada K, *et al*. A prognostic model, including the EBV status of tumor cells, for primary gastric diffuse large B-cell lymphoma in the rituximab era. Cancer Med. 2018; 7 : 3510-3520.
18 Ishikawa E, Kato S, Shimada K, *et al*. Clinicopathological analysis of primary intestinal diffuse large B-cell lymphoma: prognostic evaluation of CD5, PD-L1, and Epstein-Barr virus on tumor cells. Cancer Med. 2018; 7 : 6051-6063.
19 Chen BJ, Chapuy B, Ouyang J, *et al*. PD-L1 expression is characteristic of a subset of aggressive B-cell lymphomas and virus-associated malignancies. Clin Cancer Res. 2013; 19 : 3462-3473.
20 Nicolae A, Pittaluga S, Abdullah S, *et al*. EBV-positive large B-cell lymphomas in young patients: a nodal lymphoma with evidence for a tolerogenic immune environment. Blood. 2015; 126 : 863-872.
21 Takahara T, Satou A, Ishikawa E, *et al*. Clinicopathological analysis of neoplastic PD-L1-positive EBV^{+} diffuse large B cell lymphoma, not otherwise specified, in a Japanese cohort. Virchows Arch. 2021; 478 : 541-552.
22 Ohshima K, Suzumiya J, Kato A, Tashiro K, Kikuchi M. Clonal HTLV-I-infected CD4^{+} T-lymphocytes and non-clonal non-HTLV-I-infected giant cells in incipient ATLL with Hodgkin-like histologic features. Int J Cancer. 1997; 72 : 592-598.
23 Ohshima K, Jaffe ES, Yoshino T, Siebert R. Adult T-cell leukaemia/lymphoma. In : Swerdlow SH, Campo E, Harris NL, *et al*. (eds) : WHO Classification of Tumours of Haematopoietic and Lymphoid Tissues. Lyon, IARC. 2017; pp. 363-367.
24 Suefuji H, Ohshima K, Hayabuchi N, Nakamura K, Kikuchi M.

HTLV-1 carriers with B-cell lymphoma of localized stage head and neck: prognosis, clinical and immunopathological features. Br J Haematol. 2003; 123 : 606-612.

25 Beltran BE, Quiñones P, Morales D, *et al*. Diffuse large B-cell lymphoma in human T-lymphotropic virus type 1 carriers. Leuk Res Treatment. 2012; 2012 : 262363.

26 Spina M, Carbone A, Gloghini A, *et al*. Hodgkin's disease in patients with HIV infection. Adv Hematol. 2011; 2011 : 402682.

27 Ding J, Kumar AJ, Wilding E, Lee ST, Law JY. Multi-organ failure as the initial presentation of lymphocyte-depleted Hodgkin lymphoma in two patients with human immunodeficiency virus. Acta Haematol. 2020; 143 : 184-188.

28 Said J, Cesarman E, Rosenwald A, Harris NL. Lymphomas associated HIV infection. In : Swerdlow SH, Campo E, Harris NL, *et al*. (eds) : WHO Classification of Tumours of Haematopoietic and Lymphoid Tissues. Lyon, IARC. 2017; pp. 449-452.