Human herpes virus-8 (HHV-8) is linked to four lymphoproliferative diseases: primary effusion lymphoma, HHV-8 positive multicentric Castleman disease (MCD), HHV-8 positive diffuse large B cell lymphoma and HHV-8 positive germinotropic lymphoproliferative disorder (GLPD). The diagnosis of HHV-8 associated lymphoproliferative diseases is quite challenging because each entity is rare and has a wide morphological spectrum. Our aim is to emphasize the overlapping histopathological features of MCD and GLPD as well as to underline the importance of clinicopathological correlation in case these two entities cannot be distinguished by pathological examination.

We present here a case of an 82-year-old male patient who was examined for weight loss and multiple lymphadenopathy. Histopathological examination of the axillary lymph node revealed lymphoid follicle structures of varying shapes and sizes, including some atrophic germinal centers. Plasmablast-like cells were notable in some of these areas. HHV-8 and Epstein Barr Virus (EBV) positivity were noted in some of these cells and in a small number of cells in the mantle zone. Based on these findings; a diagnosis of “HHV-8 and EBV positive lymphoproliferative disease” was established.

Since HHV-8 positive MCD and GLPD have similar histopathological features, it may not be possible to distinguish these two entities by histopathological examination only. At this point, the importance of clinicopathological correlation becomes more evident, especially in the determination of the treatment protocol to be applied to the patient.

Keywords: Castleman disease, Germinotropic lymphoproliferative disorder, EBV, HHV-8
Abdominopelvic ultrasound showed hepatosplenomegaly while PET revealed cervical, supraclavicular, axillary, mediastinal-hilar, intraabdominal, bilateral inguinal and femoral multifocal lymphadenopathy in addition to bilateral pleural effusion. The largest lymph node was in the right axillary with a size of 30x18 mm and SUVmax of 3.3.

On evaluation of the resected right axillary lymph node specimen measuring 23x13x7 mm, serial sections were gray-white colored and a nodular appearance was remarkable. In the sections of the total processed lymph node, the normal structure was partially preserved and lymphoid follicle structures (CD20 and PAX5 positive) of varying shapes and sizes, including some atrophic germinal centers (CD21 and CD23 positive, Bcl-2 negative) were observed (Figure 1). In some of the germinal central structures, it was seen that lymphoid cells were decreased and hyalinized. Plasmablast-like cells were notable in some of these areas (Figure 2). HHV-8 and EBV positivity was noted by in situ hybridization (EBER) in some of these cells and in a small number of cells in the mantle zone (Figure 3,4). In some follicle structures, a concentric arrangement in the mantle zone areas and vascular structure penetrating into the germinal center were noteworthy (Figure 5). Occasionally, interfollicular areas were enlarged. In these areas, mostly CD3 positive T lymphocytes as well as CD38 positive plasma cells, some of which formed large aggregates, and marked vascular proliferation in the endothelium were seen (Figure 6). Although plasma cells and plasmablast-like cells were predominantly lambda positive, some of them were positive with lambda and some with kappa.

Finally the case was reported as “HHV-8 and EBV Positive Lymphoproliferative Disease” instead of giving a definite diagnosis. Two cycles of Rituximab one month apart were administered to the patient.

**DISCUSSION**

Kaposi sarcoma associated herpesvirus (KSHV), also known as HHV-8, is a lymphotropic virus and associated with 4 lymphoproliferative diseases: primary effusion lymphoma (PEL), HHV-8 positive multicentric Castleman disease (MCD), HHV-8-positive diffuse large B-cell lymphoma and rarely germinotropic lymphoproliferative disorder (GLPD) (2).

PEL presents as serous effusion in body cavities (peritoneal, pleural and pericardial) or solid tumour without effusion (“solid” PEL) and occurs in immunodeficient patients with
HIV infection. Infected patients have systemic symptoms and prognosis is poor (5). That patients have HHV-8 and EBV positive immunoblasts with plasmacytoid cytoplasm and pleomorphic nuclei. PEL differs from GLPD in the absence of cytoplasmic immunoglobulin expression (6).

Castleman disease (CD) describes 4 diseases: unicentric CD, HHV-8 associated MCD, POEMS associated MCD, idiopathic -who are negative for HHV-8 and HIV-MCD (7,8). MCD is characterized by enlarged lymph nodes in multiple regions and spleen involvement. It is a systemic disease and involves hepatomegaly, splenomegaly, constitutional symptoms and cytopenias (9). HHV-8 associated MCD occurs in most commonly HIV positive patients but HIV negative patients have also been reported (10). Histopathology is prominent, includes hyperplastic/atrophic germinal centers and hyervascularization; plasmablasts generally located in mantle zones (11,12).

GLPD is a rare HHV-8 associated lymphoproliferative disorder, first described in 3 cases in 2002 by Du et al. and followed by 15 more case reports (12) (Table I). It presents as localized lymphadenopathy and on histopathological examination it is characterized by an infiltration of germinal centers by plasmablastic cells, which are coinfected by HHV-8 and EBV. Migration of neoplastic B-lymphocytes into germinal centers may be the origin of plasmablasts in GLPD. The presence of the atypical plasma cells in the mantle zone and interfollicular area supports this theory. In addition to plasmablastic cells, residual follicle centers can be seen. There are sometimes atrophic follicles similar to MCD. GLPD responds well to chemotherapy and radiotherapy.

In keeping with these features, the possibilities of “HHV-8 Positive Multicentric Castleman Disease” and “HHV-8 Positive Germinotropic Lymphoproliferative Disorder” were considered in the differential diagnosis of our case.

Although MCD and GLPD are two distinct diseases, similar/overlapping histopathological features can be seen in these two entities (Table II).

As MCD progresses with systemic involvement, the multiple lymph node involvement and systemic symptoms in our patient primarily directed us to a diagnosis of MCD. Although GLPD usually presents as localized and sometimes multifocal lymphadenopathy (12), a few cases with symptoms such as mild splenomegaly and systemic symptoms have been reported (10,11).

Since GLPD is mostly seen in HIV-negative immunocompetent patients, we may consider the HIV negativity in favor of GLPD in our patient. However there is also a 58-year-old HIV-positive patient who was diagnosed with GLPD in the literature (11). In addition, an HIV-negative HHV-8 positive subgroup of MCD, which occurs mostly in immunosuppressive patients, has also been identified (10). Therefore, the HIV status of the patient is not a reliable criterion in distinguishing these two diseases.

Some features described in microscopic findings (plasmablast-like cells, atrophic germinal centers, decreased lymphoid cells, hyalinization etc.) overlap with both entities but the presence of a concentric arrangement in the mantle zone strengthens the diagnosis of MCD.

Another important point according to all published GLPD cases in the literature is that HHV-8 and EBV co-infection is one of the most significant criteria that differentiates GLPD from MCD (11-16). However, in an article published by Nobel et al. in 2019, EBV positivity was detected in two of two HHV-8 positive MCD patients included in the study (17). This newly defined condition, the presence of EBV positivity in MCD, will cause serious difficulties in distinguishing these two diseases, as in our case (18-19).
Table I: Clinicopathological features of patients diagnosed with germinotropic lymphoproliferative disorder

| Case | Age/Sex | Clinical Presentation | HIV | Ig heavy/light chain expression | Treatment and Prognosis |
|------|---------|-----------------------|-----|---------------------------------|------------------------|
| 1    | 41y/M (12) | Axillary and cervical lymph node enlargement for 6 years | - | Lambda cIgM, cIgD | CHOP, Complete remission |
| 2    | 61y/M (12) | Submandibular and inguinal lymph node enlargement for 4 years | - | Lambda cIgA | Excision and radiotherapy, Complete remission |
| 3    | 63y/F (12) | Paresthesia, Left leg swelling, Paraortic lymph node enlargement | NI | Kappa | NI |
| 4    | 60y/M (12) | Localized cervical lymphadenopathy | - | Kappa cIgM | Excision, No evidence of relapse |
| 5    | 65y/M (16) | Right cervical lymph node enlargement | - | Kappa cIgM | Without therapy, alive 7 years |
| 6    | 75y/M (20) | Mass in the neck, Cervical lymph node enlargement, Cystic lymph node in left submandibular area | - | Kappa | CHOP, 19 months disease free |
| 7    | 49y/F (15) | Right jugulo-cervical nodal mass | - | Lambda | Excision and radiotherapy, Complete remission |
| 8    | 84y/F (11) | Multifocal lymphadenopathy | - | None | CHOP, Complete remission |
| 9    | 58y/M (11) | Localized right axillary mass for 10 years, Mild splenomegaly | + | None | Resection, One year later developed DLBCL, died due to his disease subsequent |
| 10   | 72y/F (21) | Palpable left cervical lymph node | - | Lambda | Without therapy, No evidence of relapse |
| 11   | 63y/F (1) | Autoimmune hemolytic anemia, Prominent mesenteric lymphadenopathy | - | Lambda | Without therapy, 8 months later HHV8 + EBV + lymphoma |
| 12   | 53y/M (22) | Swelling of cervical nodes | - | μ | NI |
| 13   | 86y/M (14) | Localized cervical lymphadenopathy | - | Kappa | Without therapy, No evidence of relapse |
| 14   | 52y/M (14) | Inguinal lymph node enlargement for 3 years | - | None | CHOP |
| 15   | 47y/M (14) | Generalized lymphadenopathy, B Symptoms | + | None | CHOP |
| 16   | 27y/M (14) | Generalized lymphadenopathy, B Symptoms | + | Kappa | Rituximab |
| 17   | 30y/M (14) | Generalized lymphadenopathy, B Symptoms | + | Kappa | R-DA-EPOCH |
| 18   | 42y/M (14) | Generalized lymphadenopathy, B Symptoms | + | Lambda | R-DA-EPOCH |

NI: No information, DLBCL: Diffuse Large B Cell Lymphoma, CHOP: Rituximab, cyclophosphamide, doxorubicin, vincristine, prednisone, EPOCH: Etoposide, prednisone, vincristine, cyclophosphamide, doxorubicin, R-DA-EPOCH: Rituximab, vincristine, adriamycin, cyclophosphamide, methylprednisolone.
Table II: Comparison of the clinical and pathological features of HHV-8 positive MCD and GLPD

|                     | HHV-8 Positive MCD                                                                 | HHV-8 Positive GLPD                                                                 |
|---------------------|-----------------------------------------------------------------------------------|-----------------------------------------------------------------------------------|
| Clinical Presentation | Mostly in HIV positive immunodeficient patients                                   | Predominantly in HIV negative immunocompetent patients                             |
|                     | Generalized lymphadenopathy, splenomegaly, constitutional symptoms                | Often localized lymphadenopathy                                                    |
|                     |                                                                                  | Sometimes multifocal lymph node involvement and rarely systemic symptoms           |
| Prognosis           | Poor prognosis                                                                    | Usually favorable response to chemotherapy and radiotherapy                        |
| Microscopic Findings | Abnormal follicle structures                                                       | Residual follicle centers can be seen                                              |
|                     | Plasmablasts generally located in the mantle zone but they may intrude into germinal centers | Plasmablasts partially/completely invade germinal centers                          |
|                     | Atrophic or hyperplastic germinal centers                                          | Sometimes atrophic follicles similar to MCD                                        |
|                     | Prominent vascular proliferation                                                  |                                                                                  |
|                     | Concentric onion skin-like layering                                               |                                                                                  |
|                     | Plasma cell hyperplasia in interfollicular area                                    |                                                                                  |
| EBER                | Positive/Negative                                                                  | Always positive                                                                     |
| HIV                 | Usually positive, rarely negative                                                  | Predominantly negative, rarely positive                                            |
| Cytoplasmic Ig Heavy Chain | Elevated, only IgM                                                                      | Elevated, any heavy chain                                                                     |
| Ig Light Chain      | Monotypic lambda +                                                                 | Monotypic kappa or lambda +                                                         |
| Clonality (Ig gene rearrangements) | Polyclonal                                                                          | Polyclonal/Oligoclonal                                                               |
| Mutated Ig Genes    | Absent                                                                            | Present                                                                           |
| Cell of Origin      | A naive B cell                                                                     | A germinal center B cell                                                            |

Due to the reasons described above and the morphologically similar features, it is very difficult to distinguish between the two entities only by histopathological examination. At this point, the importance of clinicopathological correlation becomes more evident, especially in the determination of the treatment protocol applied to the patient. The physical examination and laboratory findings should also be evaluated in detail and carefully.

Conflict of Interest
The authors declare no conflict of interest.

Authorship Contributions
Concept: GB, GG, Design: GB, GG, Data collection or processing: GB, GG, MAÖ, SÖ, Analysis or Interpretation: GB, GG, MAÖ, SÖ, Literature search: GB, GG, Writing: GB, GG, Approval: SÖ.

REFERENCES
1. Courville EL, Sohani AR, Hasserjian RP, Zukerberg LR, Harris NL, Ferry JA. Diverse clinicopathologic features in human herpesvirus 8-associated lymphomas lead to diagnostic problems. Am J Clin Pathol. 2014;142:816-29.
2. Guerrero C, Jain T, Kelemen K. HHV-8-associated lymphoproliferative disorders and pathogenesis in an HIV-positive patient. Case Rep Hematol. 2019;2019:4536157.
3. Chen CH, Liu HC, Hung TT, Liu TP. Possible roles of Epstein-Barr virus in Castleman disease. J Cardiothorac Surg. 2009;4:31.
4. Taris M, de Mascarel A, Riols M, Delwail V, Milpied N, Dubus P, Parrens M. KHSV/EBV associated germinotropic lymphoproliferative disorder: A rare entity, case report and review of the literature. Ann Pathol. 2014;34:373-7.
5. Nador RG, CasARMant E, Chadburn A, Dawson DB, Ansari MQ, Said J, Knowles DM. Primary effusion lymphoma: A distinct clinicopathologic entity associated with the Kaposi’s sarcoma-associated herpes virus. Blood. 1996;88:645-56.
6. Dupin N, Diss TL, Kellam P, Tulliez Micheline, Du MQ, Sicard D, Weiss RA, Isaacsom PG, Bosoff C. HHV-8 is associated with a plasmablastic variant of Castleman disease that is linked to HHV-8-positive plasmablastic lymphoma. Blood. 2000;95:1406-12.

7. van Rhee F, Greenway A, Stone K. Treatment of Idiopathic Castleman Disease. Hematol Oncol Clin North Am. 2018;32:89-106.

8. Dispensieri A, Fajgenbaum DC. Overview of castleman disease. Blood. 2020;135:1535-64.

9. Waterston A, Bower M. Fifty years of multicentric Castleman’s disease. Acta Oncol. 2004;43:698-704.

10. Wang H, Pittaluga S, Jaffe ES. Multicentric Castleman disease: Where are we now? Semin Diagn Pathol. 2016;33:294-306.

11. Bhavsar T, Lee JC, Perner Y, Raffeld M, Xi L, Pittaluga S, Jaffe ES. KSHV- and EBV-associated germinotropic lymphoproliferative disorder: New findings and review of the literature. Am J Surg Pathol. 2018;41:795-800.

12. Du MQ, Diss TC, Liu H, Ye H, Hamoudi RA, Cabeçadas J, Dong HY, Harris NL, Chan JKC, Rees JW, Dogan A, Isaacsom PG. KSHV- and EBV-associated germinotropic lymphoproliferative disorder. Blood. 2002;100:3415-8.

13. Swerdlow SH, Campo E, Harris N, Jaffe ES, Pileri SA, Stein H, Thiele J. World Health Organization Histological Classification of Tumors of Haematopoietic and Lymphoid Tissues. 4th ed. IARC Press; 2017.

14. Gonzalez-Farre B, Martinez D, Lopez-Guerra M, Xipell M, Monclus E, Rovira J, Garcia F, Lopez-Guillermo A, Colomo L, Campo E, Martinez A. HHV8-related lymphoid proliferations: A broad spectrum of lesions from reactive lymphoid hyperplasia to overt lymphoma. Mod Pathol. 2017;30:745-60.

15. Oh J, Yoon H, Shin DK, Jang MJ, Kim G, Chong SY, Oh D. A case of successful management of HHV-8 +, EBV + germinotropic lymphoproliferative disorder (GLD). Int J Hematol. 2012;95:107-11.

16. Antonio D, Amedeo B, Maria A, Angel PM, Oscar N. KSHV- and EBV-positive germinotropic lymphoproliferative disorder: A rare lymphoproliferative disease of HIV patient with plasmablastic morphology, indolent course and favourable response to therapy. Leuk Lymphoma. 2007;48:1444-7.

17. Nabel CS, Sameroff S, Shilling D, Alapat D, Ruth JR, Kawano M, Sato Y, Stone K, Spetalen S, Valdivieso F, Feldman MD, Chadburn A, Fossà A, van Rhee F, Lipkin WI, Fajgenbaum DC. Virome capture sequencing does not identify active viral infection in unicentric and idiopathic multicentric Castleman disease. PLoS ONE. 2018;14:1-13.

18. Zanelli M, Zizzo M, Bisagni A, Froio E, Marco LD, Valli R, Filosa A, Luminari S, Martino G, Massaro F, Fratoni S, Ascani S. Germinotropic lymphoproliferative disorder: A systematic review. Ann Hematol. 2020;99:2243-53.

19. Nakaya Y, Ishii N, Kasamatsu Y, Shimizu K, Tatsumi N, Tsutsumi M, Yoshida M, Yoshimura T, Hayashi Y, Nakao T, Inoue T, Yamane T. Human herpesvirus 8-positive multicentric Castleman disease with germinotropic plasmablastic aggregates: Overlapping spectrum of human herpesvirus 8-associated lymphoproliferative disorder. Pathol Int. 2020;70:574-80.

20. D’Antonio A, Addesso M, Memoli D, Liguori P, Cuomo R, Boscaino A, Nappi O. Lymph node-based disease and HHV-8/KSHV infection in HIV seronegative patients: Report of three new cases of a heterogeneous group of diseases. Int J Hematol. 2011;93:795-801.

21. Zanelli M, Fraternali Orcioni G, Zizzo M, Marco LD, Martino G, Cerrone G, Cabras AD, Ascani S. HHV-8- and EBV-positive germinotropic lymphoproliferative disorder. Ann Hematol. 2019;98:2439-41.

22. Papoudou-Bai A, Hatzimichael E, Kyriazopoulou L, Briasonis E, Kanavaros P. Rare variants in the spectrum of human herpesvirus 8/Epstein-Barr virus-copositive lymphoproliferations. Hum Pathol. 2015;46:1566-71.