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Association Between Epstein-Barr Virus Infection and B-Cell Lymphoproliferative Diseases: A Case-Control Study

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Background: Epstein-Barr Virus (EBV) is a member of the Herpesviridae family that has infected more than 90% of the worlds' population. EBV is now considered etiologically associated with the endemic Burkitt's lymphoma, Hodgkin disease, and nasopharyngeal carcinoma. Recent findings show the association between EBV infection and other malignancies.

Objectives: The current study aimed to evaluate the relationship between EBV infection and B-cell lymphoproliferative disorders, including lymphoma and multiple myeloma.

Patients and Methods: In the current case-control study, a total of 43 patients with lymphoma and multiple myeloma and 46 age/sex-matched healthy people were included. After taking written consent, serum samples were taken from all subjects. The level of IgG against viral capsid antigen was measured using ELISA. Antibody titers > 5 U/mL was considered as positive. Data were analyzed using Stata 11 software.

Results: Of the 89 subjects, 53 were male and 36 females, aged 14 to 82 years. There was no significant difference between EBV seroprevalence in the patients with lymphoma and multiple myeloma, and the healthy subjects.

Conclusions: The results of the current study indicated no relationship between latent EBV infection and lymphoma or multiple myeloma. However, further studies with larger sample sizes are required.

Keywords: Patient; Lymphoma; Multiple Myeloma

1. Background

Epstein-Barr Virus (EBV) is a member of the Herpesviridae family . EBV infections occur worldwide. Most of these infections arise in early childhood, with a second peak in late teenage years. By adulthood, more than 90% of individuals have been infected and have antibodies to the virus (1). EBV, as well as the other herpes viruses, may play an important role in some hematologic malignancies due to their ability to modulate the host's immune system. Particularly, four herpes viruses including cytomegalovirus, EBV, human herpes virus -6 and -7 have the multiple mechanisms to modulate the host's immune system, allowing them to create latency in B lymphocytes (2, 3). EBV is the first virus described as carcinogen. It is now considered etiologically associated with the endemic Burkitt's lymphoma, Hodgkin disease, and nasopharyngeal carcinoma (4). Previous studies showed that EBV interferes with cellular DNA repair mechanisms and could lead to genetic changes in the infected cells (5). Years after primary EBV infection, a wide variety of B-cell-derived lymphoid malignancies, such as Burkitt's lymphoma, Hodgkin lymphoma, and nasopharyngeal carcinoma can emerge. These cancers can initiate from a clone of EBV-infected cells. The role of EBV in these late-onset malignancies is complicated (4). On the other hand, there is evidence that persistent EBV infection may reduce its pathogenicity; therefore, causal relationships between the virus and malignancies should be interpreted with care (6). Recent findings show a variety of results regarding the relationship between EBV infection and non-Hodgkin lymphoma (7, 8). To verify these relationships further studies are required.

2. Objectives

The current study aimed to investigate the relationship between EBV infection and lymphoproliferative disorders.

3. Patients and Methods

In the current case-control study, conducted in 2013, a total of 43 patients with B-cell lymphoproliferative disorders

Table 1. Correlation Between Anti-EBV Antibody and Lymphoproliferative Diseases

Groups	Number	Mean	P Value
Two groups			0.252 [a]
Controls	46	41.99 ± 34.44	
Cases (all patients)	43	33.04 ± 38.86	
Four groups			0.040 [b]
Normal	46	41.99 ± 34.44	
Lymphoma	24	43.57 ± 42.07	
Hodgkin	13	28.54 ± 23.52	
Multiple Myeloma	06	00.64 ± 01.57	

[a] t-test.
[b] ANOVA test.

Table 2. Relationship Between Anti-VCA Seropositivity and Lymphoproliferative Diseases

Groups	Anti-EBV Antibody, U/mL				
	Negative, < 5	Positive, ≥ 5	OR	95% CI	P Value
Normal	11	35	1.00		
Lymphoma	06	18	0.94	0.27, 3.64	0.920
Hodgkin	04	09	0.71	0.16, 3.79	0.616
Multiple Myeloma	06	00	-	-	-
Lymphoma/Hodgkin	10	27	0.85	0.28, 2.60	0.746
All cases	16	27	0.53	0.19, 1.46	0.172

including Hodgkin and non-Hodgkin lymphoma, and multiple myeloma referred to the Oncology Center of Hamadan, West of Iran, were enrolled in the study. In addition, 46 age/sex-matched healthy controls were also included. Subjects with the background of malignancies, chemotherapy, radiotherapy, blood transfusion in the recent year, or HIV infection were excluded. After taking written consent, serum samples were taken from all subjects and stored at -20°C until use. As a serologic marker of EBV, IgG against viral capsid antigen (VCA) was measured by ELISA using a specific kit for anti-VCA (Bioprob Srl., Milan, Italy). Antibody titers higher than 5 U/mL were considered positive. Data were analyzed using Stata11 software. The association between EBV seropositivity and each factor was estimated using logistic regression analysis with 95% confidence intervals; P value < 0.05 was considered significant. The study protocol and the informed consent were approved by the Ethics Committee of Hamadan University of Medical Sciences, Hamadan, Iran.

4. Results

Forty-three patients with lymphoproliferative disorders and 46 age/sex-matched control subjects participated in the study. Mean anti-VCA antibody titers in the patients and controls are shown in Table 1. There was no statistically significant difference between the mean anti-EBV titers in the case and control groups. Positive titers of anti-VCA were detected in 27 (62.7%) of the case and 35 (76%) of the control groups. As shown in Table 2, no significant relationship was observed between seropositivity of anti-VCA and lymphoprolferative diseases. In addition, there was no relationship between the types of diseases including lymphoma, Hodgkin, multiple myeloma, and anti-VCA positivity.

5. Discussion

The present study found no relationship between the seropositivity of EBV and lymphoma, Hodgkin, or multiple myeloma. The current study results were in contrast with the previous reports regarding the relationship between seropositivity of EBV antibodies and the risk of hematologic malignancies. High level of anti-EBV antibodies are detected in patients with non-Hodgkin's lymphoma, Hodgkin, and nasopharyngeal carcinoma (9). Few studies determined the role of EVB in multiple myeloma. A recent case-control study demonstrated this relationship by detecting EBV-DNA in bone marrow cells of the patients with multiple myeloma (10). On the other hand, there is supporting evidence regarding the increased risk of lymphoma in patients with EBV infection and underlying severe immunodeficiency. Some types of primary immunodeficiency disorders are predisposing factors for the development of EBV-associated hematological diseases (11). Moreover, reactivation of EBV infection in the patients with bone marrow transplant results

in fatal lymphoproliferative diseases (12, 13). These observations indicate potential oncogenicity of EBV. Several studies showed the relationship between lymphoma and EBV. In a case-control study by Mueller et al. seropositivity of anti-VCA IgG was associated with the increased risk of non-Hodgkin's lymphoma (14). In a prospective study, Lehtinen et al. reported the relationship between the elevated levels of antibodies to EBV-early antigen (EA) and Epstein-Barr nuclear antigen (EBNA) and the increased risk of lymphoma/leukemia (15). Furthermore, increased risk of lymphoma was shown in patients with anti-EA IgG positive, in a prospective study by Rothman et al. (16). Hardell et al. (17) reported a significantly increased level of IgG anti-EA and no significant elevation of anti-VCA IgG in patients with lymphoma. In a study by Gonzalez et al. (18), no relationship was found between anti –EBNA seropositivity and lymphoma, Hodgkin, or multiple myeloma. In a recent case-control study, Bertrand et al. (7) reported no relationship between serum levels of anti-VCA, anti-EA, or anti-EBNA and the risk of non-Hodgkin lymphoma. Although the results of the current study were compatible with some of above-mentioned ones, it is difficult to compare the results because of the different employed methods. Moreover, one of the limitations of the present study was the small sample size which limited of the comparison of the variables in subgroups. Another limitation of the current study was the seropositivity of EBV which is considered as only a marker of latent infection, and does not necessarily mean the reactivation of latent EBV infection. However, studies based on viral markers suggest that EBV viremia may be related with lymphoma and Hodgkin (19-21). In conclusion, according to the obtained results, the relationship between EBV infection and B-cell lymphoproliferative diseases is still controversial. To evaluate the role of EBV in lymphoproliferative disorders further studies with larger population and detection of viral DNA are necessary.

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References

1. Cohen JI. Epstein-Barr Virus Infections, Including Infectious Mononucleosis. In: Longo DL, Fauci AS, Kapser DL, Hauser SL, Jameson JL, Loscalzo J editors. *Harrison's principles of internal medicine*.. New York: McGraw-Hill; 2012. pp. 1467–70.
2. Steininger C, Rassenti LZ, Vanura K, Eigenberger K, Jager U, Kipps TJ, et al. Relative seroprevalence of human herpes viruses in patients with chronic lymphocytic leukaemia. *Eur J Clin Invest.* 2009;**39**(6):497–506.
3. Grywalska E, Markowicz J, Grabarczyk P, Pasiarski M, Rolinski J. Epstein-Barr virus-associated lymphoproliferative disorders. *Postepy Hig Med Dosw (Online).* 2013;**67**:481–90.
4. Thompson MP, Kurzrock R. Epstein-Barr virus and cancer. *Clin Cancer Res.* 2004;**10**(3):803–21.
5. Mahjour SB, Ghaffarpasand F, Fattahi MJ, Ghaderi A, Fotouhi Ghiam A, Karimi M. Seroprevalence of human herpes simplex, hepatitis B and epstein-barr viruses in children with acute lymphoblastic leukemia in southern iran. *Pathol Oncol Res.* 2010;**16**(4):579–82.
6. Thorley-Lawson DA, Gross A. Persistence of the Epstein-Barr virus and the origins of associated lymphomas. *N Engl J Med.* 2004;**350**(13):1328–37.
7. Bertrand KA, Birmann BM, Chang ET, Spiegelman D, Aster JC, Zhang SM, et al. A prospective study of Epstein-Barr virus antibodies and risk of non-Hodgkin lymphoma. *Blood.* 2010;**116**(18):3547–53.
8. Teras LR, Rollison DE, Pawlita M, Michel A, Brozy J, de Sanjose S, et al. Epstein-Barr virus and risk of non-Hodgkin lymphoma in the cancer prevention study-II and a meta-analysis of serologic studies. *Int J Cancer.* 2015;**136**(1):108–16.
9. Kutok JL, Wang F. Spectrum of Epstein-Barr virus-associated diseases. *Annu Rev Pathol.* 2006;**1**:375–404.
10. Sadeghian MH, Ayatollahi H, Keramati MR, Memar B, Jamedar SA, Avval MM, et al. The association of Epstein-Barr virus infection with multiple myeloma. *Indian J Pathol Microbiol.* 2011;**54**(4):720–4.
11. Parvaneh N, Filipovich AH, Borkhardt A. Primary immunodeficiencies predisposed to Epstein-Barr virus-driven haematological diseases. *Br J Haematol.* 2013;**162**(5):573–86.
12. Chen DB, Song QJ, Chen YX, Chen YH, Shen DH. Clinicopathologic spectrum and EBV status of post-transplant lymphoproliferative disorders after allogeneic hematopoietic stem cell transplantation. *Int J Hematol.* 2013;**97**(1):117–24.
13. Furuya A, Ishida M, Hodohara K, Yoshii M, Okuno H, Horinouchi A, et al. Epstein-Barr virus-related post-transplant lymphoproliferative disorder occurring after bone marrow transplantation for aplastic anemia in Down's syndrome. *Int J Clin Exp Pathol.* 2014;**7**(1):438–42.
14. Mueller N, Mohar A, Evans A, Harris NL, Comstock GW, Jellum E, et al. Epstein-Barr virus antibody patterns preceding the diagnosis of non-Hodgkin's lymphoma. *Int J Cancer.* 1991;**49**(3):387–93.
15. Lehtinen T, Lumio J, Dillner J, Hakama M, Knekt P, Lehtinen M, et al. Increased risk of malignant lymphoma indicated by elevated Epstein-Barr virus antibodies--a prospective study. *Cancer Causes Control.* 1993;**4**(3):187–93.
16. Rothman N, Cantor KP, Blair A, Bush D, Brock JW, Helzlsouer K, et al. A nested case-control study of non-Hodgkin lymphoma and serum organochlorine residues. *Lancet.* 1997;**350**(9073):240–4.
17. Hardell K, Carlberg M, Hardell L, Bjornfoth H, Ericson Jogsten I, Eriksson M, et al. Concentrations of organohalogen compounds and titres of antibodies to Epstein-Barr virus antigens and the risk for non-Hodgkin lymphoma. *Oncol Rep.* 2009;**21**(6):1567–76.
18. Berrington de Gonzalez A, Urban MI, Sitas F, Blackburn N, Hale M, Patel M, et al. Antibodies against six human herpesviruses in relation to seven cancers in black South Africans: a case control study. *Infect Agent Cancer.* 2006;**1**:2.
19. Gandhi MK, Lambley E, Burrows J, Dua U, Elliott S, Shaw PJ, et al. Plasma Epstein-Barr virus (EBV) DNA is a biomarker for EBV-positive Hodgkin's lymphoma. *Clin Cancer Res.* 2006;**12**(2):460–4.
20. Kabyemera R, Masalu N, Rambau P, Kamugisha E, Kidenya B, De Rossi A, et al. Relationship between non-Hodgkin's lymphoma and blood levels of Epstein-Barr virus in children in north-western Tanzania: a case control study. *BMC Pediatr.* 2013;**13**:4.
21. Morishima S, Nakamura S, Yamamoto K, Miyauchi H, Kagami Y, Kinoshita T, et al. Increased T-cell responses to Epstein-Barr virus with high viral load in patients with Epstein-Barr virus-positive diffuse large B-cell lymphoma. *Leuk Lymphoma.* 2014:1–7.