

Expression of Epstein-Barr virus in patients with Hodgkin's disease: report of 64 cases from Rio de Janeiro, Brazil

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Expressão do vírus Epstein-Barr em pacientes com doença de Hodgkin: relato de 64 casos do Rio de Janeiro

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key words	abstract
Hodgkin's disease	Epstein-Barr virus (EBV) has been implicated in the pathogenesis of Hodgkin's disease (HD), and the association between EBV and HD is influenced by the patients' socioeconomic status, age and the presence of mixed cellularity (MC) histological subtype. The prevalence of EBV in HD varies widely. This study was undertaken to determine the prevalence of EBV in HD in a Brazilian population. We studied 64 cases with a monoclonal antibody to the EBV latent membrane protein-1 (LMP1). EBV was detected in 35 of the 64 cases (55%) and its presence was significantly correlated with the MC subtype (OR = 9; IC 95% = 1.66 – 66; p = 0,0015). These results support that EBV is related to HD in a Brazilian population.
Epstein-Barr virus	
LMP1	

resumo	unitermos
<i>O vírus Epstein-Barr (EBV) tem sido implicado na fisiopatogenia da doença de Hodgkin (DH) e a associação deste vírus com a DH está relacionada com as condições socioeconômicas da população estudada, com a idade e com o subtipo histológico celularidade mista (CM). A prevalência do EBV na DH é muito variável. Este estudo foi realizado com o objetivo de determinar a prevalência do EBV na DH em uma população brasileira. Foram estudados 64 casos de DH oriundos do Hospital Universitário utilizando-se o método de imunistoquímica com anticorpo monoclonal contra a proteína latente da membrana (LMP1). O vírus foi encontrado em 35 dos 64 casos estudados (55%) e sua presença correlacionou-se de maneira significativa com o subtipo histológico CM (OR = 9; IC 95% = 1,66 – 66; p = 0,0015). Estes resultados confirmam que o EBV está relacionado com a DH em uma população brasileira.</i>	Doença de Hodgkin Vírus Epstein-Barr LMP1

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The paper is based on a master dissertation by dr. Monique Morgado Loureiro, supervised by dr. Nelson Spector and dr. José Carlos Morais, entitled *Prevalência do Vírus Epstein-Barr em Pacientes com Doença de Hodgkin Acompanhados no Hospital Universitário Clementino Fraga Filho*; 2000, Postgraduate Program in Internal Medicine, Universidade Federal do Rio de Janeiro, Brazil.

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Introduction

The role of Epstein-Barr virus (EBV) in the pathogenesis of Hodgkin's disease has been intensely debated during the last three decades. There are a number of epidemiological evidences that suggest an etiologic role for EBV in Hodgkin's disease, and it is of substantial interest that the prevalence of EBV in Hodgkin's disease varies sharply worldwide. EBV was detected in 100% of patients with HD in countries such as Honduras⁽²⁾ and Peru⁽⁶⁾, but in less than 50% of HD cases in the United Kingdom⁽¹⁴⁾, the United States⁽³⁾ and Costa Rica⁽⁹⁾.

Prevalence in Brazil can be as high as 79% in children from the state of Ceará⁽¹⁾. However, geographic differences strongly influence distribution of age and of the histologic subtypes⁽¹⁰⁾.

Objective

We undertook this study in order to determine the prevalence of EBV in patients with HD from the state of Rio de Janeiro.

Methods

A full review of the pathological archive of Hospital Universitário/Universidade Federal do Rio de Janeiro since its inception in 1979 until 1996 identified 119 cases of Hodgkin's disease. Fifty-five cases had to be excluded from the analysis because the paraffin blocks were unavailable. The remainder 64 cases, forming the study group, were revised by an experienced hemopathologist and classified according to the Rye classification⁽⁸⁾.

Immunohistochemical studies for EBV latent membrane protein-1 (LMP1) were performed using the CS1-4 antibody cocktail (Dako), at a dilution of 1:50. The following clinical variables were also recorded: age, sex, clinical stage (Ann Arbor) and performance status (ECOG).

Proportions were compared with the Yates-corrected chi-square test and Fisher's test, when appropriate.

Results

Clinical characteristics and histologic features

The median age was 37 years (range, 11-84 years). In two patients, the histological classification was not

possible. The clinical and histological features are shown in Table 1.

Immunohistochemical findings

Expression of LMP1 was identified in the cytoplasm and membrane of the Hodgkin's cells and variants. Overall, EBV was identified in 35 cases (55%). The prevalence rate of EBV in the different histologic subtypes is shown in Table 2.

EBV was present in 63% of male patients but in only 38% of female patients. The EBV was identified in 61% of patients with advanced clinical stage (III and IV) and in 50% of patients with early stages.

EBV was detected in 75% of patients younger than 18 years and in 52% of older patients, but this difference was not significant.

It is noteworthy that the presence of EBV correlates significantly with the mixed cellularity subtype (OR = 9; IC 95% = 1.66 – 66; p = 0,0015), but not with sex, age and clinical stage.

Discussion

The study of the epidemiology of Hodgkin's disease has suggested the existence of different patterns of disease according to the level of industrial and socioeconomic development of the countries. Poor countries are characterized by a lower incidence of Hodgkin's disease, an absence of the incidence peak in the third decade of life, and a predominance of the mixed cellularity subtype over nodular sclerosis. In contrast, the pattern in rich countries includes an increased incidence in females, a third decade peak and a predominance of the nodular sclerosis subtype⁽⁷⁾.

The data from Brazil appear to corroborate this model. Brazil's poorest region, the northeast, still reveals a predominance of mixed cellularity cases⁽¹¹⁾, while in a series from Campinas, in Brazil's richest state of São Paulo, 78% of patients had nodular sclerosis⁽¹³⁾. In the present series, from Rio de Janeiro, 55% of the patients had nodular sclerosis.

Another salient feature in the data from Brazil is the very high prevalence of advanced disease at diagnosis. In the present series 78% had advanced disease, and in Campinas 68% had advanced disease. This is likely the result of delayed diagnosis, due to the shortcomings of the public health system. We have previously reported that the median time elapsed from the beginning of symptoms to

Table 1 Clinical and histological features of the 64 patients with Hodgkin's disease

Clinical features	Patients
Sex	
Male	43/64 (67%)
Female	21/64 (33%)
Ann Arbor stage	
Early (I-II)	10/46 (22%)
Advanced (III-IV)	36/46 (78%)
Histologic subtype	
Nodular sclerosis	35/64 (55%)
Mixed cellularity	16/64 (25%)
Lymphocyte depletion	7/64 (11%)
Lymphocyte predominance	4/64 (6%)
Unknown	2/64 (3%)
B symptoms	46/62 (74%)
PS (ECOG)	
0	47/62 (76%)
1	7/62 (11%)
2	3/62 (5%)
3	2/62 (3%)
4	3/62 (5%)
Localization	
Cervical	46/62 (74%)
Supraclavicular	27/62 (43%)
Axillary	25/62 (40%)
Retroperitoneal	25/62 (40%)
Inguinal	16/62 (26%)
Mediastinal	16/62 (26%)
Hepatomegaly	15/62 (24%)
Splenomegaly	23/62 (37%)

the diagnosis of Hodgkin's disease was five months (range, 1-36 months)⁽¹²⁾.

With regard to EBV, the overall prevalence is strongly influenced by the rate of the mixed cellularity subtype in a given population. The rates of EBV positivity are consistently high in this subtype, ranging from 88%

in the present series to 93% in Campinas and to 100% in the northeast. Such rates, not seen in the other subtypes of Hodgkin's disease, suggest that EBV might be directly implicated in the pathogenesis of this disease subtype.

The case for a role of EBV in the pathogenesis of Hodgkin's disease has become stronger as new evidence has linked EBV to the NF- κ B transcription factor. It has been demonstrated that EBV's LMP1 is a constitutively aggregated pseudo-tumor necrosis factor receptor that directly activates NF- κ B⁽⁴⁾. Moreover, it has been shown that NF- κ B activity is essential in the growth and survival of lymphoblastoid cell lines transformed by EBV, and that NF- κ B inhibition induces apoptosis of these cell lines⁽⁵⁾.

Conclusion

In this study the prevalence of de EBV in HD was 55% and it was higher in the histologic group of MC in a Brazilian population. A comprehensive panorama of the features of Hodgkin's disease in a country so diverse in its ethnicity and sociology as Brazil might help us better understand the complex interplay between biological and environmental factors that appears to lie at the very core of the pathogenesis of Hodgkin's disease.

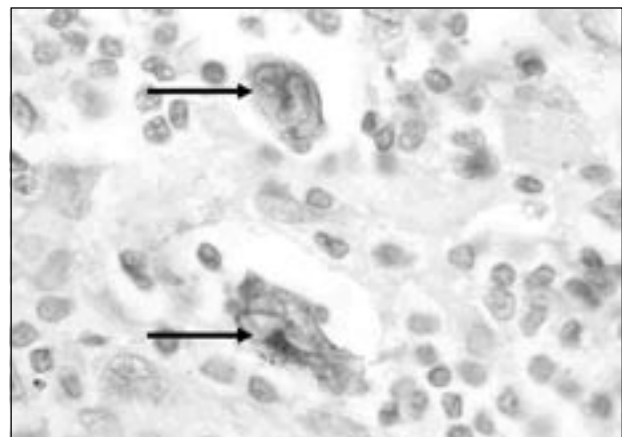


Figure – Two Reed-Sternberg cells with positive markings for EBV (arrows). The upper one has a membrane marking, and the bottom one, a membrane marking and a Golgi-placed dot

Table 2 Prevalence rate of EBV in the histologic subtypes

Histologic subtype	LMP1 positivity	Frequency
Nodular sclerosis	14/35	40%
Mixed cellularity	14/16	88%
Lymphocyte depletion	4/7	57%
Lymphocyte predominance	2/4	50%

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