Virologic Diagnosis, Viral Monitoring, and Treatment of Epstein-Barr Virus Infectious Mononucleosis

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Epstein-Barr virus (EBV) is the cause of infectious mononucleosis and is associated with severe infections in immunocompromised patients. EBV is also causally linked with several human malignancies. The heterophile antibody test and EBV-specific antibody tests remain the principal means of diagnosis of initial infection in otherwise healthy patients. Enzyme-linked immunosorbent assays have replaced the traditional immunofluorescence assays for EBV-specific antibodies. Several newer molecular diagnostic tests have become available that facilitate accurate monitoring of infection. The role of these tests for patients with uncomplicated infectious mononucleosis is limited, although these tests are being increasingly used to monitor the state and level of EBV replication for severe infections and among immunocompromised patients. Antiviral therapy has a limited, short-term effect on oropharyngeal shedding but has proven ineffective for the clinical manifestations of infectious mononucleosis. Patients with selected complications frequently benefit from short-term corticosteroid therapy.

Introduction

Epstein-Barr virus (EBV) is a ubiquitous member of the Herpesviridae family and the principal cause of infectious mononucleosis, which is associated with a characteristic triad of fever, pharyngitis, and generalized lymphadenopathy. EBV is distributed worldwide and infects up to 95% of the world's adult population. An important relationship exists between age of primary EBV-infected patient and incidence of symptomatic disease. In developing countries and in socioeconomically disadvantaged populations of industrialized countries, primary infection usually occurs during early childhood and usually is asymptomatic or indistinguishable from other childhood viral infections. Up to 95% to 100% of children in these populations are seropositive by age 2 to 4 years. In industrialized countries with more affluent populations, early childhood infection is still most common, but approximately one third to one half of cases of infection occur during adolescence and early adulthood. Primary EBV infection in adolescents and adults is manifest in approximately 50% of cases as infectious mononucleosis, a clinical syndrome characterized by systemic somatic complaints, primarily of fever, fatigue and malaise, lymphadenopathy, and sore throat. This symptom pattern is recognized clinically as the clinical syndrome of infectious mononucleosis. Infectious mononucleosis in higher socioeconomic groups in industrialized countries is most commonly observed among individuals aged 15 to 24 years, with a peak age-specific incidence of approximately 18 years for men and 16 years for women, despite the higher incidence of asymptomatic EBV infection among infants and young children.

After primary infection, EBV establishes lifelong infection in the host and is maintained as episomes in resting B lymphocytes [1]. Latently infected lymphocytes may enter active replication, especially in the presence of host immunosuppression, but it is replication in the oral cavity, including the oral epithelium, that results in shedding of transmissible virus and spread of infection. Recent studies using more sensitive methods of detection such as polymerase chain reaction (PCR) show that the excretion of EBV in saliva is much greater than conventionally thought. EBV is detected by PCR from the oral cavity in 90% of healthy adults [2]. Almost all previous studies have used EBV culture, which is less sensitive, especially for identification of EBV type 2, which constitutes 14% of oral EBV isolates [2].

In patients with HIV infection and AIDS, uncontrolled EBV replication in the epithelium along the lateral margins of the tongue may develop into oral hairy leukoplakia [3], and in the lung is the apparent cause of lymphocytic interstitial pneumonitis, which has been observed principally among children [4].

Severe and even fatal lymphoproliferative disease occurs rarely among persons who appear to be immunologically incapable of limiting the replication and dissemination of EBV-infected B cells [5]. Patients with the X-linked lymphoproliferative syndrome have a specific genetic immunologic defect that permits overwhelming and frequently fatal primary EBV infection [6].

Epstein-Barr virus infection also has been linked with several human tumors, including African Burkitt's lymphoma, nasopharyngeal carcinoma, Hodgkin's disease, and some T-cell lymphomas. In immunocompromised patients and organ transplant recipients, EBV is associated with leiomyosarcomas, post-transplant lymphoproliferative disease, and B-cell lymphomas, especially in the central nervous system.

Diagnosis

There are several methods used for virologic diagnosis and monitoring of EBV infection (Table 1). Heterophile antibody is a relatively nonspecific but simple and very useful diagnostic test for primary EBV infection. EBV-specific antibodies provide very good specificity, with sensitivity from good to high that is dependent on the methodology and the test that is used. EBV serologic test results are often misinterpreted. The newer molecular diagnostic tests provide the highest levels of specificity and have sensitivity from very high to the highest of all available tests, depending on the specific methodology used.

The conventional diagnostic approach for acute infectious mononucleosis has been serologic testing for the antibody response, including heterophile antibody and EBV-specific antibodies to selected viral proteins. Serologic tests remain the mainstay of diagnosis of infectious mononucleosis because of their widespread availability, specificity, and sensitivity. The traditional but time-consuming immunofluorescence assays have been supplanted in most laboratories by enzyme-linked immunosorbent assay (ELISA)–based tests. Serologic tests are useful for diagnosis of acute infection if performed in the first 2 to 3 months of infection, and distinguish acute from remote infection, but cannot time the onset of infection after the first 3 to 8 months. In a few circumstances, serologic tests can be used to monitor disease progression over time, such as for nasopharyngeal carcinoma that is associated with characteristically high titers of immunoglobulin A (IgA) viral capsid antigen (VCA) and IgA early antigen (EA).

There are several additional tests, including newer molecular diagnostic tests, that can be used for diagnosis of EBV infection. These tests are used infrequently to diagnose infectious mononucleosis in otherwise healthy persons but are very useful in severe infections or for EBV infection in immunocompromised patients. They also are useful to delineate the state of EBV infection, as latent or replicative, and the burden of EBV infection because they can be quantitated. Immunohistochemistry of tissues identifies EBV protein expression and distinguishes latent from active, replicative EBV infection. Culture of EBV from blood or saliva identifies infectious virions and can be further semiquantitated using the virocyte assay. Culture usually requires 3 to 6 weeks and is impractical for routine diagnostic use. Electron microscopy identifies whole virions in tissues and fluids but is impractical for routine diagnostic use. Southern blot analysis of the EBV genome terminal repeats determines the clonality of EBV infection and distinguishes latent and replicative states. Newer molecular methods for detection of EBV nucleic acid include PCR for DNA amplification, which also can be quantitated using real-time PCR to monitor viral burden and disease progression. In situ hybridization identifies EBV in tissues and in specific cell types within lesions.

Antibody testing

Several distinct EBV-associated antigen systems and their corresponding antibodies have been characterized (Table 2) and developed into important diagnostic tests [7,8]. The EBV antigen systems are classified by the phase of the viral replicative cycle during which they are expressed. Only a few viral genes are transcribed during latent infection and include: six EBV nuclear antigens (EBNA 1, 2, 3A, 3B, 3C, and LP [leader protein]), of which EBNA 1 is the principal constituent; three latent membrane proteins (LMP 1, 2A, and 2B); and two short EBV-encoded RNAs (EBER 1 and 2), which are not translated. LMP 1 and EBNA 2, in combination with the major histocompatibility complex antigens, constitute the lymphocyte-detected membrane antigen, a cell surface antigen recognized by cytotoxic T cells. The EAs are produced during the initial stages of viral lytic replication before viral DNA synthesis. The EAs include two morphologic components, diffuse (EA-D) and restricted (EA-R), which are distinguished by the basis of their distribution within the cells and by their differential denaturation by fixation procedures and proteolytic enzymes. EA-D and EA-R each comprise two different EBV proteins. The late antigens are produced after viral DNA synthesis and include the VCAs, the structural proteins of the capsid. Membrane antigens (MAs) are structural polypeptides of the virus expressed before and after viral DNA synthesis on the cell surface and also form part of the viral envelope.

Heterophile antibody

Heterophile antibody associated with acute infectious mononucleosis agglutinates sheep and horse erythrocytes, among others, and is adsorbed by beef erythrocytes but not guinea pig kidney cells [9•]. This heterophile antibody response usually peaks during the second and third weeks of infectious mononucleosis and is frequently detectable for several months after resolution of clinical symptoms (Fig. 1).

The rapid slide test is the most widely used method to detect heterophile antibody. Slide tests that use horse erythrocytes are more sensitive and detect heterophile antibody in 90% of cases of EBV-associated infectious mononucleosis in adults, with a false-positive rate of less than 10%. This method detects a heterophile response in ≤ 50% of cases in children aged younger than 4 years, although a greater percentage may have detectable heterophile antibody by the immune adherence hemagglutination procedure [8]. Heterophile antibody test kits using sheep erythrocytes or those that lack a guinea pig kidney or beef absorption step (unless the method incorporates native and enzyme-treated erythrocytes for agglutination) should not be used.

Table 1. Tests for diagnosis and monitoring of EBV infection

Test	Target	Specimen	Applications
Serology			
Heterophile antibody	Nonspecific antibody	Serum	Diagnosis of acute infection, distinguish acute from remote infection, serologic monitoring of disease and response to treatment (Table 2)
VCA, EA, and EBNA antibody	EBV-specific antibody	Serum	
Virus culture			
Transformation	Replicating virus	Blood or oropharyngeal secretions	Detect presence and quantitate infectious EBV
Virocyte assay (semiquantitative)	Virus-containing cells	Blood or oropharyngeal secretions	
Electron microscopy	Whole virus	Biopsies, tissues, cultured cells from tumors, and smears of suspension cells (cultured cells or peripheral blood cells)	Detect presence of a member of the herpesvirus family (large, enveloped viruses)
Direct antigen detection (immunohistochemistry)	EBV latent antigens (*eg*, EBNA, LMP); EBV replicative antigens (*eg*, EA, VCA) in lesions of oral hairy leukoplakia	Biopsies, tissues, or touch preparations	Detect EBV protein expression in tissues, including localization to specific cells; distinguish latent from replicative EBV infection based on virus protein expression
Southern blot analysis for clonality and lytic replication	EBV genome terminal repeats	Biopsies or tissues	Detect EBV DNA; distinguish monoclonal, oligoclonal, or polyclonal EBV infection; demonstrate the presence of linear EBV genomes as evidence of active viral replication
Nucleic acid detection	EBV genomic DNA	Peripheral blood, oropharyngeal, secretions, cervical secretions, and fresh or frozen biopsy	Detect presence of EBV; quantitate EBV (using real-time PCR); viral load monitoring of disease and response to treatment (using real-time PCR)
Detection (and lower limits)			
Southern blot (10^5 EBV genomes)			
Dot blot (10^4 EBV genomes)			
PCR (10^{1-3} EBV genomes)			
Real-time PCR (10^{2-7} EBV genomes)			
Quantitation			
PCR			
Real-time PCR			
In situ hybridization	EBER	Biopsies and tissues	Detect presence of EBV in tissues, including localization to specific cells

EA—early antigen; EBNA—Epstein-Barr virus–determined nuclear antigens; EBV—Epstein-Barr virus; LMP—latent membrane protein; PCR—polymerase chain reaction; VCA—viral capsid antigen.

Epstein-Barr virus–specific serologic tests

The EBNA, EA, and VCA antigen systems are the most useful for clinical diagnostic purposes and are determined principally by ELISA tests in most clinical laboratories, or by indirect immunofluorescent assay (IFA) in research laboratories [9•]. IFA methods are more specific but more time consuming and technically demanding, and remain the standard for evaluation of new tests. There is high concordance between commercial ELISA kits, although comparative testing of ELISA to IFA shows a sensitivity of ELISA of only approximately 71% for VCA immunoglobulin G (IgG), 95% for VCA IgM, and 99% for anti-EBNA, and with an overall specificity of 99% [10•].

Serum neutralizing antibodies to EBV can be measured using a neutralization assay based on interference of lymphocyte transformation [11]. This is technically demanding and requires several weeks for cultivation and transformation, and therefore is not used for clinical diagnostic testing. Neutralizing antibodies correlate well with anti-VCA and anti-MA titers. Determination of complement-fixing antibodies against antigens extracted from EBV-positive lymphoblastoid cell lines is rarely performed.

Table 2. Correlation of clinical status and serologic responses to EBV infection*

Clinical status	Heterophile antibodies (qualitative test)	EBV-specific antibody				
		IgM VCA	IgG VCA	EA-D	EA-R	EBNA
Negative reaction	-	< 1:8†	< 1:10†	< 1:10†	< 1:10†	< 1:2.5†
Susceptible	-	-	-	-	-‡	-
Acute primary infection (infectious mononucleosis)	+	1:32 to 1:256	1:160 to 1:640	1:40 to 1:160	-‡	- to 1:2.5
Recent primary infection (infectious mononucleosis)	+/-	- to 1:32	1:320 to 1:1280	1:40 to 1:160	-‡	1:5 to 1:10
Remote infection	-	-	1:40 to 1:160	-§	- to 1:40	1:10 to 1:40
Reactivation (immunosuppressed or immunocompromised)	-	-	1:320 to 1:1280	-§	1:80 to 1:320	- to 1:160
Burkitt's lymphoma	-	-	1:320 to 1:1280	-§	1:80 to 1:320	1:10 to 1:80
Nasopharyngeal carcinoma	-	-	1:320 to 1:1280	1:40 to 1:160	-¶	1:20 to 1:160

*The data were obtained from many studies. Individual responses outside the characteristic range may occur.
†Or the lowest test dilution.
‡In young children and adults with asymptomatic seroconversion, the anti-early antigen response may be mainly to the EA-R component.
§Some patients will have the anti-early antigen response mainly to the EA-D component.
¶Some patients will have the anti-early antigen response mainly to the EA-R component.
EA-D—diffuse staining component of early antigen; EA-R—cytoplasmic-restricted component of early antigen; EBNA—Epstein-Barr virus–determined nuclear antigens; EBV—Epstein-Barr virus; IgG—immunoglobulin G; IgM—immunoglobulin M; VCA—viral capsid antigen.
(*From* Jenson and Ench [9•]; with permission.)

The serologic profile of EBV-specific antibodies following infectious mononucleosis has been well-documented (Fig. 1). The range of antibody responses occasionally complicates interpretation of an individual antibody profile (Table 2). The acute phase of infectious mononucleosis is characterized by rapid IgM and IgG antibody responses to VCA, and in most cases, a lower IgG response to the EA complex. The IgM response to VCA is transient, lasting approximately 1 to 3 months. The IgG response to this antigen usually peaks during the acute illness, decreases slightly over the next few weeks to months, and then persists at a relatively stable level for life. Anti-EA antibodies usually are present for several months but may persist for several years after resolution of the acute infection. Anti-EBNA antibodies typically appear much later, gradually emerging after 2 to 4 months, and occasionally longer, after the onset of illness, and lasting for life. Some immunocompromised patients demonstrate diminishing or undetectable levels of anti-EBNA antibodies [5].

The determination of IgM antibody to VCA is the most valuable serologic procedure to diagnose acute EBV infection. Properly performed, the IgM VCA antibody test is quite reliable and specific, and can be detected in almost all patients for 4 to 12 weeks after clinical onset of infectious mononucleosis. Rheumatoid factor may cause a false-positive IgM reaction, and therefore it is essential to routinely use methodology such as an IgG-inactivating reagent (GullSORB; Meridian Diagnostics, Cincinnati, OH) that removes rheumatoid factor. An adsorption step to remove serum IgG and thus eliminate the false-positive reaction associated with rheumatoid factor also reduces the incubation time needed for a satisfactory reaction between VCA and the test serum. A less acceptable alternative is to test each IgM VCA–positive serum undiluted for the presence of rheumatoid factor.

It is prudent to augment the IgM VCA test with at least one other antibody determination, such as IgG VCA or EBNA. Very high titers of IgG VCA alone do not necessarily indicate acute infection. Most patients with infectious mononucleosis develop a transient antibody response to the EA-D component, although in some adult patients and especially in young children, this response may be directed mainly against the EA-R component. The testing of a second, or convalescent phase, serum 4 to 6 weeks after clinical onset is of minimal assistance for diagnosing a preceding acute EBV infection because less than 10% of patients have a significant antibody titer increase during this interval.

A past, currently quiescent, EBV infection is characterized by the concurrent presence of moderate but stable antibody titers to IgG VCA and EBNA and the absence of IgM VCA. Antibodies to EA often are absent, but if present, are usually at a low level and are directed predominantly to the EA-R component. High titers of IgG VCA and EA, considered to indicate enhanced EBV replication or reactivation, have been noted in patients with Burkitt's lymphoma, nasopharyngeal carcinoma, or immunosuppressed or immunodeficient states. The

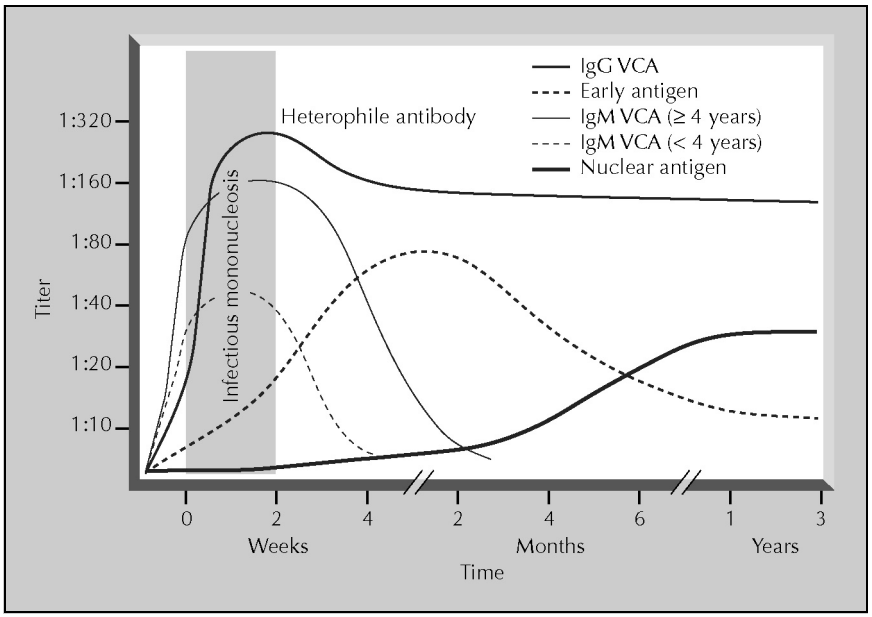

Figure 1. Schematic representation of the development of antibodies to various Epstein-Barr virus antigens in patients with infectious mononucleosis. The titers are geometric mean values expressed as reciprocals of the serum dilution. The immunoglobulin M (IgM) response to viral capsid antigen (VCA) is less in children aged younger than 4 years. IgG—immunoglobulin G. (*Adapted from* Jenson and Ench [9•]; with permission.)

antibody response to EA is directed principally to the EA-R component in patients with Burkitt's lymphoma and immunosuppressed or immunodeficient patients and to EA-D in patients with nasopharyngeal carcinoma. High levels of IgA VCA and IgA EA also are found in patients with nasopharyngeal carcinoma, including the early asymptomatic stages. In these patients, tumor activity and response to cancer therapy may be monitored by serial IgA VCA and IgA EA determinations. Serologic testing for IgA antibody responses to VCA and EA-D may be useful in diagnosing and monitoring nasopharyngeal carcinoma.

Virus detection

The transformation assay can semiquantitatively measure the level of infectious EBV by measuring induction of lymphoproliferation of human umbilical cord lymphocytes, or activation of B cells to immunoglobulin production. Transforming EBV can be isolated from oral secretions and peripheral lymphocytes, especially during acute infectious mononucleosis and then intermittently for life.

The number of virus-containing cells (virocytes) in a clinical specimen can be quantitated by the virocyte assay [12]. The concentration of virocytes is calculated from the dilutions of cells showing signs of transformation.

Electron microscopy can be used to identify viruses with a morphologic appearance of herpesviruses, which are large, enveloped viruses. This finding indicates active viral replication with production of intact viruses. However, electron microscopy is impractical for routine clinical use.

Direct antigen detection

Immunohistochemistry is used to detect EBV antigens directly in cryostat sections from biopsies, tissues, cultured cells from tumors, and smears of suspension cells (cultured cells or peripheral blood cells) or touch preparations. Latent antigens (*eg*, EBNA, LMP) are routinely detected in EBV-associated lesions, but it is unusual to detect replicative antigens (*eg*, EA, VCA) in tissues, with the exception of lesions of oral hairy leukoplakia, which typically demonstrate dense concentrations of VCA and EA. Testing for minor EBV regulatory (*eg*, BLZF1) or structural antigens using newer monoclonal antibodies that are available is not necessary for the routine diagnostic evaluation of EBV infection but is useful in research of the nuances of viral replication or reactivation of latent EBV.

Nucleic acid detection

Nucleic acid hybridization is a very sensitive and specific method for detection of nucleic acid in clinical specimens and has distinct advantages over serologic methods. Direct detection of EBV DNA is more reliable than serologic testing for immunosuppressed or immunocompromised patients who may not exhibit a complete humoral response, and in patients who have received blood or immune globulin products, which confound serologic diagnosis.

Dot blotting (also known as spot or slot blotting), Southern blotting, PCR, and in situ hybridization for EBV DNA have been applied to peripheral blood cells, oropharyngeal secretions, cervical secretions, and fresh or frozen biopsy specimens, principally from lymphoproliferative lesions (Table 1). These techniques vary in sensitivity and specificity for identification of EBV DNA, and therefore, the results of these tests for the establishment of the causal role of EBV for different syndromes and lesions must be interpreted within the limitations of each test. The approximate lower limits of sensitivity for EBV detection are 10^5 EBV genomes for Southern blotting, 10^4 EBV genomes for dot blotting, 10^{1-3} EBV genomes for conventional PCR, and a range between 10^2 to 10^7 EBV genomes for real-time or quantitative PCR. The specificity of dot blotting, Southern blotting, and especially PCR for localizing EBV within specific cells of tissue specimens is confounded by the

inherent simultaneous detection of EBV genomes present in circulating lymphocytes that may be present in the tissues. In situ hybridization is the most specific of these four molecular biological methods because it permits direct evaluation of individual cells [13••].

Dot blotting and Southern blotting

Numerous radionuclide probes derived from sequences throughout the EBV genome have been successfully used in dot blotting and Southern blotting to demonstrate EBV in clinical specimens. Certain sequences, such as the large internal repeat (IR_1), have theoretical advantages for increased sensitivity because of the presence of multiple copies in each EBV genome. The sensitivity and specificity may vary for each nucleic acid probe, and therefore, the limits must be established experimentally using appropriate controls [14]. Dot blotting uses total intracellular DNA and may be semiquantitated using serial dilutions. Southern blotting uses total intracellular DNA digested with one or more restriction endonucleases followed by separation of restriction fragments by gel electrophoresis. Southern blotting is very specific because restriction fragments can be distinguished from nonspecific hybridization. The precise pattern of hybridization is dependent on the restriction endonuclease used to digest the sample DNA and the EBV probe used in the Southern blotting. Genomic variation, including large deletions, among different EBV isolates frequently results in restriction fragment length polymorphisms that may result in false-negative test results or may confound interpretation [15].

Southern blot analysis also can be used to determine the clonality of EBV infection based on comparison of the numbers of terminal repeats at the ends of each EBV genome [16•]. The terminal repeat structure of the infecting virion is passed to progeny cells during cell division. Analysis of the pattern confirms the presence of EBV, distinguishes monoclonal from oligoclonal or polyclonal EBV infection, and identifies active viral replication by the presence of linear EBV genomes (in addition to episomal genomes of latent infection). Most EBV-associated tumors demonstrate monoclonal EBV infection, whereas oral hairy leukoplakia represents high levels of virus replication and reinfection, resulting in polyclonal infections within the lesion [17].

Polymerase chain reaction

Polymerase chain reaction is perhaps the most sensitive method available for detection of EBV DNA, and several assay methods have been developed to determine the EBV burden in serum, plasma, and peripheral blood lymphocytes [2,18–23,24••]. Real-time PCR, using fluorescent TaqMan methodology (Roche Diagnostics, Basel, Switzerland), provides better quantitation with excellent sensitivity, specificity, and reproducibility across a large dynamic range [18,25]. Certain regions of the EBV genome are highly conserved and may be preferable regions for Southern blot and PCR analyses, whereas diversity in other areas of the genome among EBV isolates has been used as a tool for studying the molecular epidemiology of EBV infection. Differences in restriction endonuclease sites by Southern blot or PCR analysis in a clinical EBV isolate must be interpreted in the context of expected EBV genomic diversity. The sensitivity and specificity may vary for each set of PCR primers, and therefore the limits must be established experimentally using appropriate controls.

Polymerase chain reaction is much more sensitive than serologic testing early in the course of infection and has been used to confirm EBV infection in EBV-seronegative infants with infectious mononucleosis–like symptoms [25,26]. The magnitude of EBV burden measured by PCR also has been shown to correlate with the severity of acute illness [20,21,27], and serial PCR measurements have been used to define the risk for post-transplant lymphoproliferative disease [28,29•].

In situ hybridization

The EBER transcripts, EBER 1 and 2, are expressed in excess of 10^6 copies per cell in latently infected lymphocytes, and because of their abundance, are the best target for in situ hybridization [13••,30]. These RNA polymerase III transcripts of unknown function are approximately 170 nucleotides in length and have minimal homology with mammalian RNAs. EBER riboprobes may be labeled with ^3H, ^{35}S, digoxigenin, biotin, or fluorescein for detection. In situ hybridization using nonisotopic EBER probes is more sensitive for detection of virus than PCR is for detection of viral DNA, provides cellular localization of EBV that demonstrates infected cells even when they are only a minor subpopulation, and can be applied to routinely processed paraffin-embedded tissue sections.

Treatment

Antiviral treatment

There have been five randomized controlled trials of intravenous acyclovir [31,32] or oral acyclovir [33–35] treatment among 339 otherwise healthy persons with infectious mononucleosis. Despite in vitro virologic activity of acyclovir, these studies showed no statistically significant benefit or clinical effectiveness of acyclovir treatment, individually or by meta-analysis [36••]. There is significant reduction in the rate of oropharyngeal EBV shedding at the end of therapy, but no difference at 3 weeks. There is no evidence that antiviral therapy hastens resolution of clinical symptoms or reduces the risk for development of complications of infectious mononucleosis.

The experience of treatment of oral hairy leukoplakia in patients with AIDS provides further insights into the antiviral treatment of EBV infections. Although EBV is found in the oral cavity with or without oral hairy leukoplakia [37], and hairy leukoplakia does not routinely require treatment, studies have shown that the advanced lesions usually respond to short-term oral acyclovir [38,39], desciclovir

[40], and valacyclovir [41], with restoration of the normal epithelial architecture within 2 to 4 weeks. In most cases, EBV replication subsides and the hairy leukoplakia lesion resolves. The virologic response to treatment is initially persistent, nonproductive EBV infection of the oral mucosa with limited expression of replicative EBV genes. Productive EBV replication often recurs after discontinuation of antiviral therapy, and recurrence is usually within 1 to 4 months after discontinuing treatment. Occasionally, treatment fails and EBV replication persists.

Thus, antiviral treatment decreases viral shedding among immunocompetent and immunocompromised patients, but the clinical benefits are only discernible among immunocompromised patients. In all patients, EBV infection persists and oral shedding recurs after discontinuation of antiviral therapy. One important distinction is that hairy leukoplakia in patients with AIDS is characterized by infection with multiple EBV strains [42] with potential for intrastrain recombination [15,43]. When treating coinfection with multiple genotypes and impaired immunity, each of which facilitates molecular evolution, the addition of acyclovir or valacyclovir provides pressure for selection of resistant EBV strains.

Corticosteroids

The use of corticosteroids to treat complications of infectious mononucleosis is based on anecdotal experience but has not been evaluated by randomized, controlled trials. Corticosteroids are unnecessary in mild, uncomplicated cases of infectious mononucleosis and should not be routinely administered to all patients. Possible indications include incipient upper airway obstruction, autoimmune hemolytic anemia or neutropenia, thrombocytopenia with hemorrhage, and meningoencephalitis and other neurologic complications. Intravenous dexamethasone (0.25 mg/kg every 6 hours), or methylprednisolone (1 mg/kg every 6 hours), or oral prednisone (40–60 mg daily) administered for 1 to 3 days have been used with similar results of dramatic subjective improvement within 24 hours and objective improvement within 72 hours [44–47]. Corticosteroid therapy may hasten resolution of complications in some but not all patients with infectious mononucleosis [48]. Reluctance to use corticosteroids is based on the unknown long-term effects of using an immunomodulator for a virus that establishes intracellular latency and for which the normal immune response is apparently quite effective in preventing progression and subsequent development of EBV-associated malignancies.

Conclusions

Epstein-Barr virus infection remains a ubiquitous right of passage, whether causing clinically nondescript illness or classic infectious mononucleosis. Until an effective vaccine is developed, lifelong infection with EBV remains an element of the "normal" adult state of "health." Serologic diagnosis is still the principal means of diagnosis of primary EBV infection, but with some exceptions, is of limited value in monitoring the state of EBV infection and associated disease. Newer molecular diagnostic tests provide important insights into the state and level of activity of EBV and will continue to be used more widely, especially among patients with severe infection or in immunocompromised patients. Antiviral treatment of infectious mononucleosis in otherwise healthy patients has proven ineffective and is not recommended. Short-term corticosteroid therapy is useful for patients severely affected with selected complications.

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