Microbiology - HLS

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Viral Diseases in the Hematolymphatics

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Parvoviruses

- Members of the family Parvoviridae, are small (diameter, ~22 nm), nonenveloped, icosahedral viruses with a linear single-strand DNA genome of ~5000 nucleotides. **One of the simplest and smallest DNA viruses**
- These viruses are dependent on either rapidly dividing host cells or helper viruses for replication. **Such as Adenovirus or herpes virus , except parvovirus is independent**
- This group's only human pathogen, parvovirus B19, is the causative virus in erythema infectiosum (also known as "slapped cheek syndrome" or the "fifth disease") in children and causes aplastic crisis in anemic patients.
- The virus also contributes to joint diseases, embryopathies, and tissue rejection following renal transplants.

TABLE 31-1 Important Properties of Parvoviruses

Virion: Icosahedral, 18–26 nm in diameter, 32 capsomeres
Composition: DNA (20%), protein (80%)
Genome: Single-stranded DNA, linear, 5.6 kb, MW 1.5–2.0 million
Proteins: One major (VP2) and one minor (VP1)
Envelope: None
Replication: Nucleus, dependent on functions of dividing host cells
Outstanding characteristics: Very simple viruses Human pathogen, B19, has tropism for red blood cell progenitors One genus contains viruses that are replication-defective and require a helper virus

Parvovirus B19

- The parvoviruses are among the smallest viruses with a diameter of 19–25mm.
- They are icosahedral, nonenveloped, and their genome is in the form of single-stranded DNA (ssDNA).
- Some parvoviruses can only replicate in the presence of a helper virus (adenovirus or herpesvirus).
- Parvovirus B19, the only human pathogenic parvovirus identified to date, is capable of autonomic replication, i.e., it requires no helper virus.

Epidemiology.

- The B19 virus is widespread. Infections can occur throughout the year in all age groups and as outbreaks or as sporadic cases.
- Infections are most commonly seen as outbreaks in schools
- Droplet infection or the fecal-oral route, analogous to other parvoviruses, is suspected. Blood and blood products are infectious, so that multiple transfusion patients and drug addicts are high incidence groups. **Fomites might be a source of infection**

Pathogenesis

- Parvovirus B19 replicates in the bone marrow in erythrocyte precursor cells, which are destroyed in the process.
- In patients already suffering from anemia (sickle-cell anemia, chronic hemolytic anemia), such infections result in so-called aplastic crises in which the lack of erythrocyte resupply leads to a critical shortage.
- The virus also appears to cause spontaneous abortions in early pregnancy and fetal damage in late pregnancy (hydrops fetalis).
- In otherwise healthy persons, these infections usually run an asymptomatic course. They can, however, also cause a harmless epidemic infection in children, erythema infectiosum (“slapped-cheek syndrome” or “fifth disease”).

TABLE 31-2 Human Diseases Associated with B19 Parvovirus

Syndrome	Host or Condition	Clinical Features
Erythema infectiosum	Children (fifth disease)	Cutaneous rash
	Or Adults	Arthralgia-arthritis
Transient aplastic crisis	Underlying hemolysis	Severe acute anemia
Pure red cell aplasia	Immunodeficiencies	Chronic anemia
Hydrops fetalis	Fetus	Fatal anemia

Slap cheek rash migrates to trunk, abdomen , back and limbs

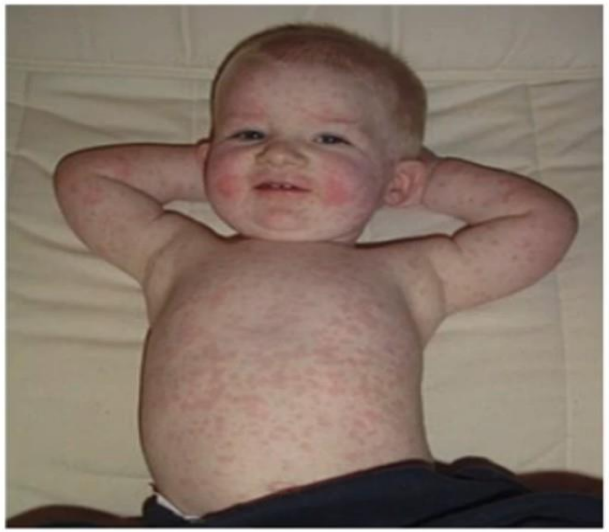
Hand Joints then ankle joints

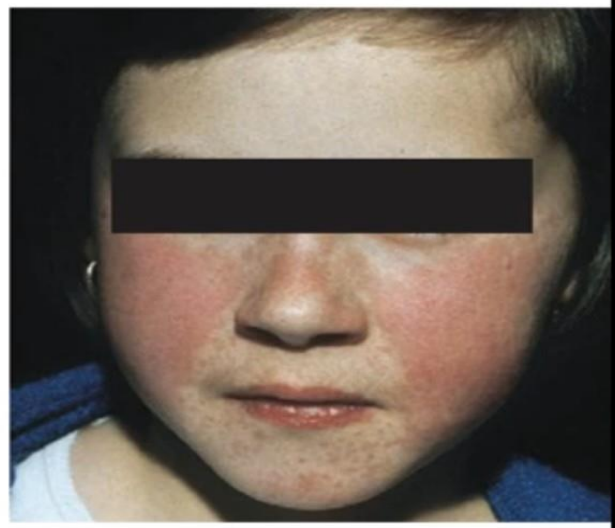
CLINICAL MANIFESTATIONS

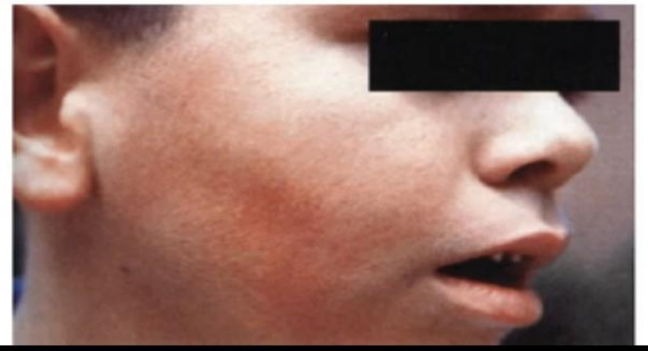
Most of B19 infections are asymptotic (a patient will show no symptoms while infecting other)

❖ Erythema Infectiosum (Fifth disease or slapped-cheek disease)

- Infection begins with a minor febrile prodrome ~7–10 days after exposure.
- the classic facial rash develops several days later; after 2–3 days, the erythematous macular rash may spread to the extremities in a lacy reticular pattern.
- Adults typically do not exhibit the "slapped-cheek" phenomenon but present with arthralgia, with or without the macular rash.

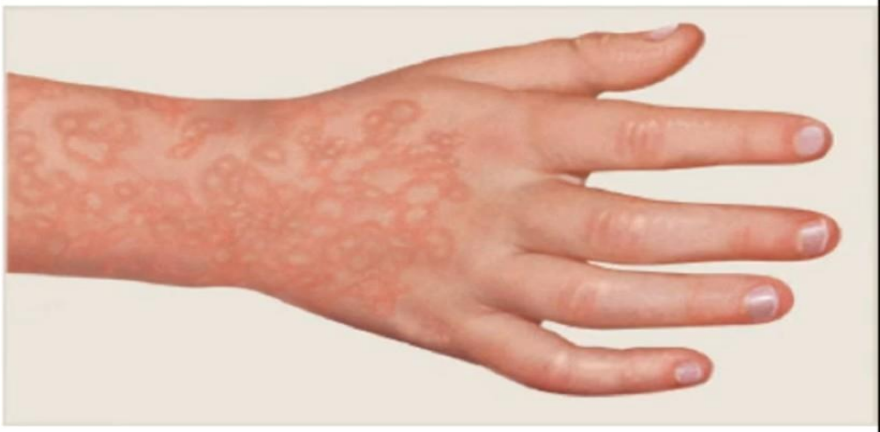






❖Polyarthropathy Syndrome

- Although uncommon among children, arthropathy occurs in ~50% of adults and is more common among women than among men.
- The distribution of the affected joints is often symmetrical, with arthralgia affecting the small joints of the hands and occasionally the ankles, knees, and wrists.
- Resolution usually occurs within a few weeks, but recurring symptoms can continue for months.



One of the differential diagnosis is idiopathic juvenile rheumatoid arthritis , but if the infected person with B19 already has rheumatoid arthritis it would be aggravating factor

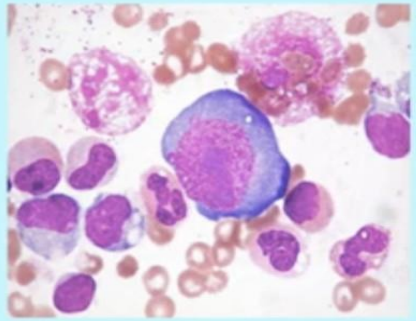
Patient already has a problem in RBCs erythropoiesis then the problem becomes worse

❖ Transient Aplastic Crisis (TAC):

Affects RBCs precursors → low HB levels → sever anemia

- In most individuals with B19V infection, asymptomatic transient reticulocytopenia occurs.
- However, in patients who depend on continual rapid production of red cells, infection can cause transient aplastic crisis. Affected individuals include those with hemolytic disorders, hemoglobinopathies, red cell enzymopathies, and autoimmune hemolytic anemias.
- Patients present with symptoms of severe anemia (sometimes life-threatening) and a low reticulocyte count, and bone marrow examination reveals an absence of erythroid precursors and characteristic giant pronormoblasts.

Giant pronormoblast is parvovirus infection



❖Pure Red-Cell Aplasia/Chronic Anemia

- Chronic B19V infection has been reported in a wide range of immunosuppressed patients, including those with congenital immunodeficiency, AIDS, lymphoproliferative disorders (especially acute lymphocytic leukemia), and transplantation.

- Patients have persistent anemia with reticulocytopenia, absent or low levels of B19V IgG,* high titers of B19V DNA in serum, and—in many cases—scattered giant pronormoblasts in bone marrow.

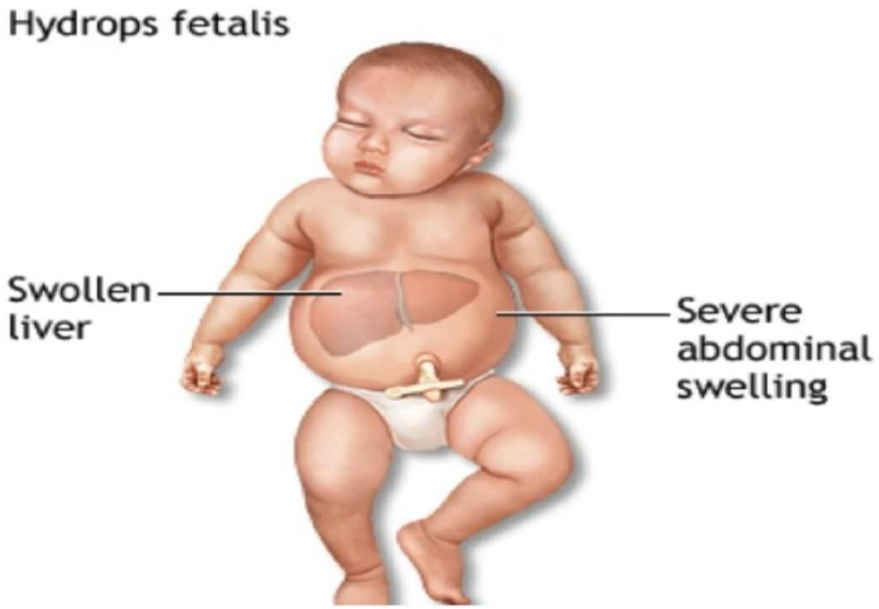
*** Because the patients are already immunosuppressed**

Hydrops Fetalis = edema affecting the baby caused by fetal anemia

❖Hydrops Fetalis

- B19 infection during pregnancy can lead to hydrops fetalis and/or fetal loss.

- The risk of transplacental fetal infection is ~30%, and the risk of fetal loss (predominantly early in the second trimester) is ~9%. Although B19V does not appear to be teratogenic



Diagnosis.

- An enzyme immunoassay reveals antibodies of the IgG and IgM classes. **Serology : western plot or ELISA**

- During the viremic phase, at the onset of clinical symptoms, the virus can also be identified in the blood by means of electron microscopy or PCR.

- In-vitro culturing of the pathogen is not standard procedure.

U can find IgG or IgM against antigens from Parvovirus BUT remember in pure cell aplasia IgG & IgM levels are low

Treatment

- Symptomatic treatment. **No specific anti viral treatment**
- TAC precipitated by B19V infection frequently necessitates symptom-based treatment with blood transfusions.

For pure red cell aplasia :

- Commercial immune globulin (IVIg) from healthy blood donors can cure or ameliorate persistent B19V infection in immunosuppressed patients.

- Administration of IVIg is not beneficial for erythema infectiosum or B19V-associated polyarthropathy. Intrauterine blood transfusion can prevent fetal loss in some cases of fetal hydrops.
- There is no vaccine against human parvovirus

Herpesviruses

HHV6 causes the sixth disease

- The viruses in this family all feature a practically identical morphology, but show little uniformity when it comes to their biology and the clinical pictures resulting from infections.

- One thing shared by all herpesviruses is the ability to reactivate after a period of latency.

Also, some information that's worth mentioning is these viruses are dsDNA Viruses, enveloped viruses and could be reactivated in a latency period. (Dormant Stage)

- The herpes simplex virus (HSV, two serotypes), The varicella-zoster virus (VZV), Cytomegalovirus (CMV), The **E**pstein-Barr virus (EBV), Human herpesvirus 6 (HHV 6) and Human herpesvirus 8 **(HHV 8).**

EBV is known to be the causative agent of infectious mononucleosis and for being responsible for multiple cancers (malignancies). & HHV is Kaposi-associated sarcoma.

Herpesviruses

- They have dsDNA genomes. Replication of the DNA and the morphogenesis of the virus particle take place in the host-cell nucleus.

- The envelope (inner nuclear membrane) is then formed when the virus penetrates the nuclear membrane.

- Common to all herpesviruses is a high level of generalized contamination (60–90% carriers) and the ability to persist in a latent state in the body over long periods.

Epstein-Barr virus (EBV)

B cells (Lymphocytes) are their main target ((Remember E**B**V + **B** cells))

AKA Kissing Disease - it transmits through saliva

- The virus is a member of the family Herpesviridae
- Is the cause of heterophile-positive infectious mononucleosis (IM), which is characterized by fever, sore throat, lymphadenopathy, and atypical lymphocytosis.
- EBV is also associated with several tumors, including nasopharyngeal and gastric carcinoma, Burkitt's lymphoma, Hodgkin's disease, and (in patients with immunodeficiencies) B cell lymphoma.
- The two types of EBV that are widely prevalent in nature are not distinguishable by conventional serologic tests.

EPIDEMIOLOGY

EBV has 2 peaks:

- EBV infections occur worldwide. These infections are most common in early childhood, with a second peak during late adolescence. By adulthood, more than 90% of individuals have been infected and have antibodies to the virus.

Risk factors:

- IM is usually a disease of young adults. In lower socioeconomic groups and in areas of the world with deficient standards of hygiene (e.g., developing regions), EBV tends to infect children at an early age, and IM is uncommon.
- In areas with higher standards of hygiene, infection with EBV is often delayed until adulthood, and IM is more prevalent.

TRANSMISSION

There are many routes of transmission, but this is the main route:

- EBV is spread by contact with oral secretions. The virus is frequently transmitted from asymptomatic adults to infants and among young adults by transfer of saliva during kissing.
- Transmission by less intimate contact is rare. EBV has been transmitted by blood transfusion and by bone marrow transplantation.
- More than 90% of asymptomatic seropositive individuals shed the virus in oropharyngeal secretions. Shedding is increased in immunocompromised patients and those with IM.

patient will show no symptoms while infecting other

PATHOGENESIS

- EBV is transmitted by salivary secretions. The virus infects the epithelium of the oropharynx and the salivary glands and is shed from these cells. While B cells may become infected after contact with epithelial cells.
- The virus then spreads through the bloodstream. The proliferation and expansion of EBV-infected B cells along with reactive T cells result in enlargement of lymphoid tissue. Polyclonal activation of B cells leads to the production of antibodies to host-cell and viral proteins.
- This virus also persists in latency, probably for the life of the patient, in (immortalized) B cells.
- If T cell immunity is compromised, EBV-infected B cells may begin to proliferate , virus-induced proliferation is but one step in a multistep process of neoplastic transformation.

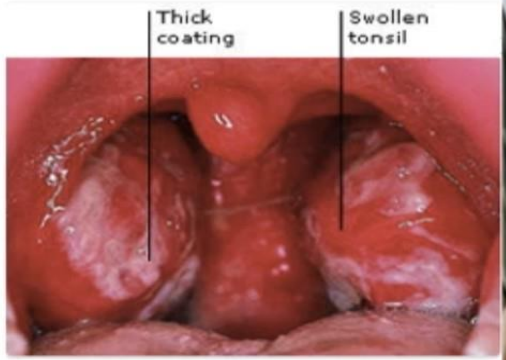
- The EBV receptor (CD21) on the surface of B cells is also the receptor for the C3d component of complement.

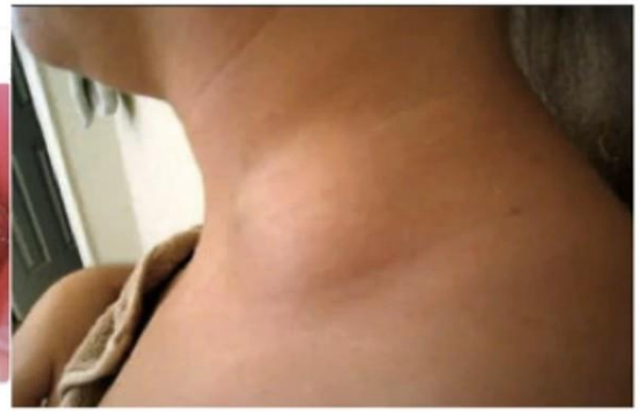
- During latent infection of B cells, only the EBV nuclear antigens (EBNAs), latent membrane proteins (LMPs), and small EBV RNAs (EBERs) are expressed in vitro. Important for serological tests

- EBV-transformed B cells secrete immunoglobulin; only a small fraction of these cells produce virus.

CLINICAL MANIFESTATIONS

In clinics, one of the differential diagnoses is -as shown in the figure- enlarged inflamed tonsils with a whitish discoloration.

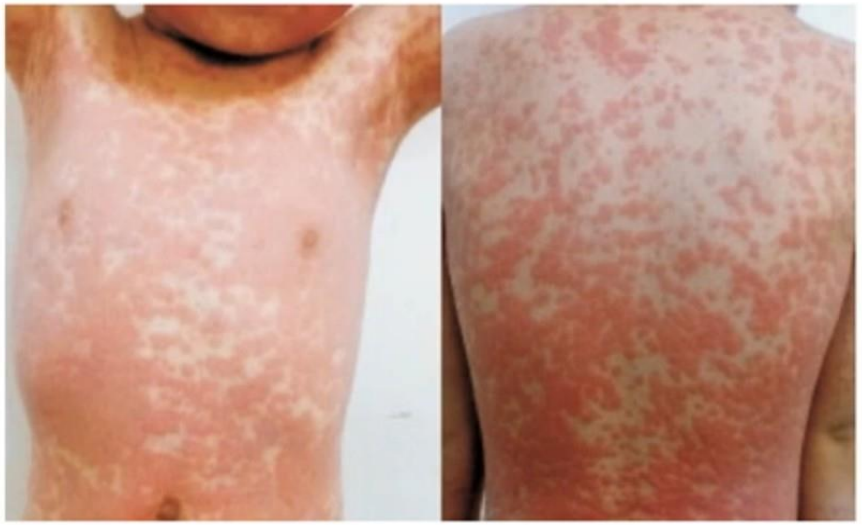




❖ Infectious mononucleosis (IM)

Again, AKA Kissing Disease

- The incubation period for IM in young adults is ~4–6 weeks. A prodrome of fatigue, malaise, and myalgia before the onset of fever, sore throat, and rash, lymphadenopathy.
- fever, fatigue, myalgia, and malaise,pharyngitis, lymphadenopathy, splenomegaly, and atypical lymphocytes
- Liver and spleen involvement and enlargement

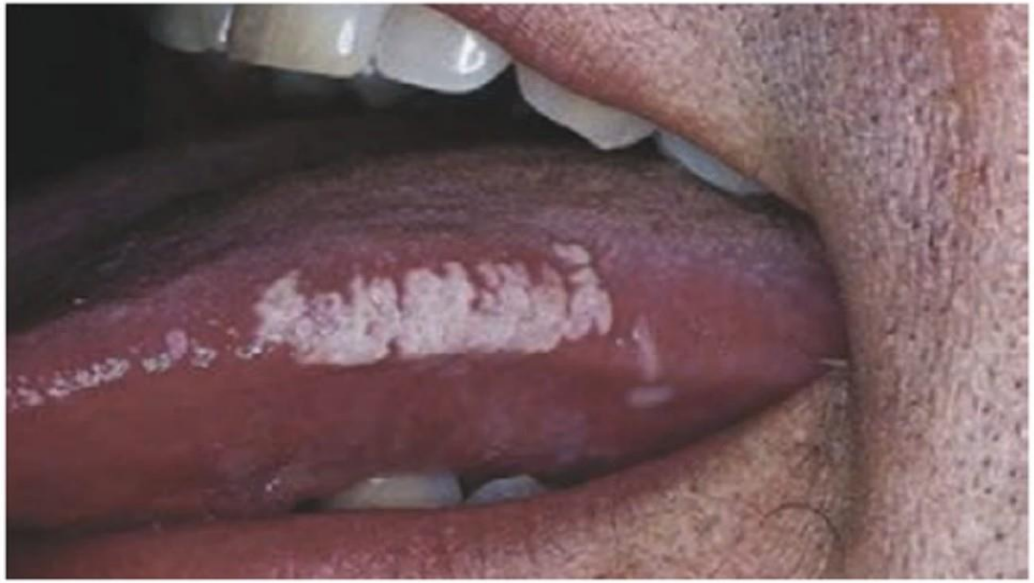


A CBC test for those patients will show increased WBC count

EBV-Associated Diseases Other Than IM

- B cell hyperplasia or poly- or monoclonal lymphoma.
- X-linked lymphoproliferative disease
- Oral hairy leukoplakia
- Burkitt's lymphoma
- Anaplastic nasopharyngeal carcinoma
- Gastric carcinoma.
- Hodgkin's disease

This figure shows



➢There are characteristic chromosome translocations that involve immunoglobulin genes and result in deregulation of expression of the c-myc proto-oncogene .

This is not the only mechanism, just one of them for illustration purposes

DIAGNOSIS

➢ Molecular Assays for Identification of Virus PCR

- Nucleic acid hybridization is the most sensitive means of detecting EBV in patient materials.

➢ Isolation of Virus

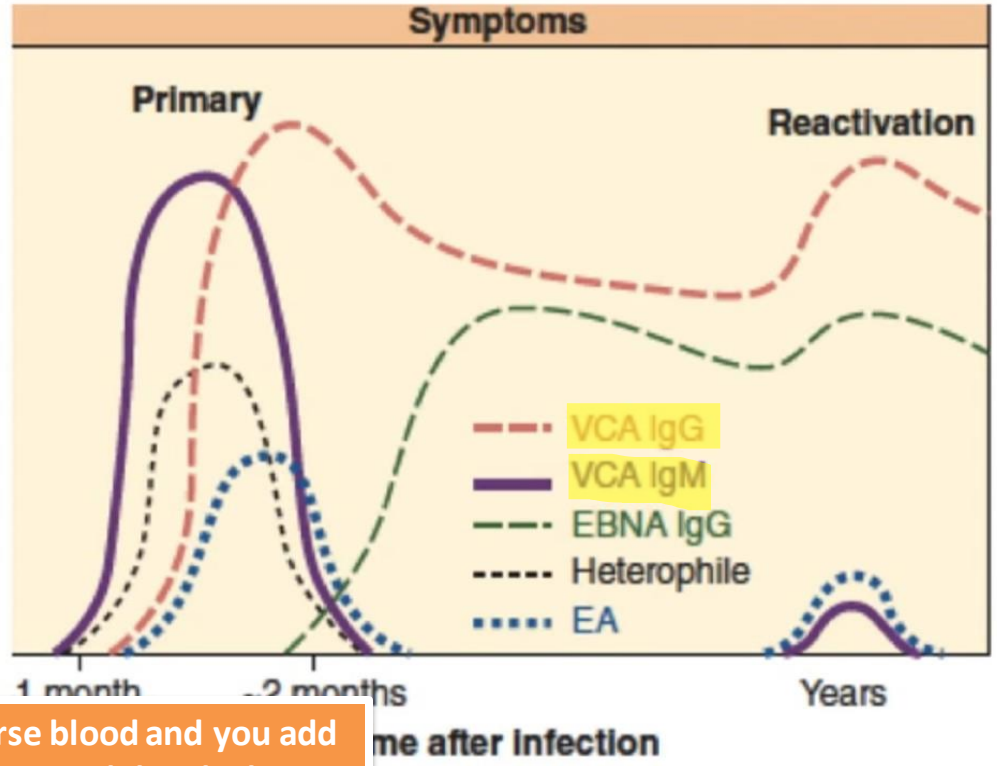
- EBV can be isolated from saliva, peripheral blood, or lymphoid tissue by immortalization of normal human lymphocytes, usually obtained from umbilical cord blood.

Doing cultures of viruses is nothing like doing cultures for bacteria and that's due to many reasons explained in the general course just know that it's done in specialized laboratories, and they're not performed on regular basis.

➢ Serology

Most commonly used

- Enzyme-linked immunosorbent assays, immunoblot assays, and indirect immunofluorescence tests using EBV-positive lymphoid cells.
- The heterophil agglutination test (Monospot)



a blood test used to determine whether you have contracted the EBV, << so you bring horse blood and you add to this sample another sample form a serum from a suspected patient if agglutination happened then he has the Virus, however this test is not approved by the FDA.

EA: Early Antigen

As you see in this figure the EBNA IgG it's raised at a late period of the exposure and it remains a positive test for a long time which causes high level of contamination in this family.

TREATMENT

- Acyclovir reduces EBV shedding from the oropharynx during the period of drug administration, but it does not affect the number of EBV-immortalized B cells

So not a curative drug

Or the immortalized cells

- Acyclovir has no effect on the symptoms of mononucleosis and is of no proved benefit in the treatment of EBV-associated lymphomas in immunocompromised patients .

- There is no EBV vaccine available.

HUMAN HERPESVIRUS 8 **Kaposi sarcoma herpes virus**

- A new herpesvirus, designated HHV-8 and also called KSHV, was first detected in 1994 in Kaposi sarcoma specimens . **After the detection of HIV in 1980s**
- KSHV is lymphotropic and is more closely related to EBV
- The KSHV genome (~165 kbp) contains numerous genes related to cellular regulatory genes involved in cell proliferation, apoptosis, and host responses (cyclin D, cytokines, chemokine receptor) that presumably contribute to viral pathogenesis.
- KSHV is the cause of Kaposi sarcomas, vascular tumors of mixed cellular composition, and is involved in the pathogenesis of body cavity-based lymphomas occurring in AIDS patients.

HIV patients suffer a vascular tumor in their body cavities of mixed cellular composition (this tumor is known as Kaposi sarcoma). It appeared that HIV patients were infected with HHV-8

TRANSMISSION

- Contact with oral secretions is likely the most common route of transmission.

Less intimate routes are considered as routes of transmission
Vertically : from the mother to her fetus through the placenta

- The virus can also be transmitted sexually, vertically, by blood, and through organ transplants. Viral DNA has also been detected in breast milk samples in Africa.

Remember this in HTLV-1, because breast milk is the main route of transmission there.

DIAGNOSIS

- Viral DNA can be detected in patient specimens using PCR assays.

- Direct virus culture is difficult and impractical.

Culture is a possible way of diagnosing HHV-8, but it needs specialized laboratories

- Serologic assays are available to measure persistent antibody to KSHV using indirect immunofluorescence, Western blot, and enzyme-linked immunosorbent assay formats.

You can either look for an antigen or an antibody directed against HHV-8

TREATMENT

Foscarnet = good therapeutics against HHV-8

- Foscarnet, famciclovir, ganciclovir, and cidofovir have activity against KSHV replication.

Human T-Lymphotropic Viruses **Targets T-cells.**

HTLV has 2 serotypes, HTLV-1 is the major serotype

- HTLV-1 has been established as the causative agent of adult T-cell leukemia-lymphomas (ATL) as well as a nervous system degenerative disorder called tropical spastic paraparesis; HTLV-1-associated myelopathy (HAM). **ATL is one of the most aggressive cancers a human can get, its 5 year survival rate is less than 5%**

- The human lymphotropic viruses have a marked affinity for mature T cells.

- The virus is distributed worldwide, with an estimated 20 million infected individuals.

TRANSMISSION

- Transmission of HTLV-1 seems to involve cell-associated virus.

- Mother-to-child transmission via breast feeding is an important mode.

  The most important route

- Blood transfusion is an effective means of transmission, as are sharing blood contaminated needles (drug abusers) and sexual intercourse.

  + Organ transplantation form affected patients

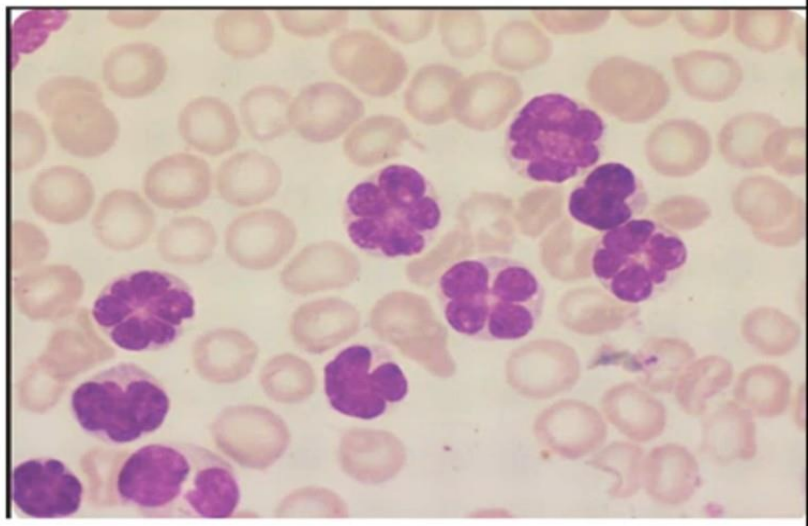
- There is a long latency period (≈30 years) before the onset of leukemia.

Incubation period is long , it takes around 20 years for a patient to know she/he has HTLV-1

Note : shedding of the cells must be very high for HTLV-1 to be transmitted as an STD

Human T-lymphotropic virus **Clinical Syndromes**

- HTLV infection is usually asymptomatic but can progress to ATLL in approximately 1 in 20 persons over a 30 years old. **The affinity for matures T-cells focuses on CD4 cells (helper T-cells)**
- ATLL caused by HTLV-1 is a neoplasia of the CD4 helper T cells that can be acute or chronic .
- The malignant cells have been termed "flower cells" because they are pleomorphic and contain lobulated nuclei. **In a flow cytometry, if you isolate these lymphocytes, you'll find a multilobulated cauliflower appearance**
- ATLL is usually fatal within a year of diagnosis, regardless of treatment



DIAGNOSIS

- Serology ELISA , Western blot **ELISA = Enzyme Linked Immunosorbent assay**

  Serology ELISA is done on the serum, we look for antibodies targeted against antigens from the infecting virus.
- Viral PCR

  This is the easiest way of diagnosis; we look for antigens

TREATMENT

- For the small number of patients who develop HTLV-1-related disease, therapies are not curative.

- No specific antiviral therapy However, the combination of interferon α and zidovudine may extend survival

Life expectancy is short because the diseases is very fatal, only 5% of patients live up to 5 years after being diagnosed with HTLV-1

PREVENTION

It's like a preventive mechanism

- Women in endemic areas should not breast-feed their children, and blood donors should be screened for serum antibodies to HTLV-1.
- As in the prevention of HIV infection, the practice of safe sex and the avoidance of needle sharing are important.

In Jordan, we screen for the following viruses :

1. **HTLV-1**
2. **HIV**
3. **Hep B**
4. **Hep C**