

Infectious Disease Serology

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Outline

Infectious Diseases and their Diagnostic Methods

- Syphilis (*Treponema pallidum*) a spirochete type of bacteria
- EBV (Epstein Barr Virus)
- HIV (Human Immunodeficiency Virus)
- Viral Hepatitis
- CMV (Cytomegalovirus)
- TOXO (*Toxoplasmosis gondii*)

Serology

- What is Serology
- General Rules in Interpreting Serology Results

IgM only – recent primary infection

IgM and IgG – recent or recurring infection

IgG only – past exposure or immunization

- Serology vs. Molecular Tests: Pros and Cons

Good for screening

Good for Early detection

Inexpensive & Fully automated

Good in immunocompromised patients

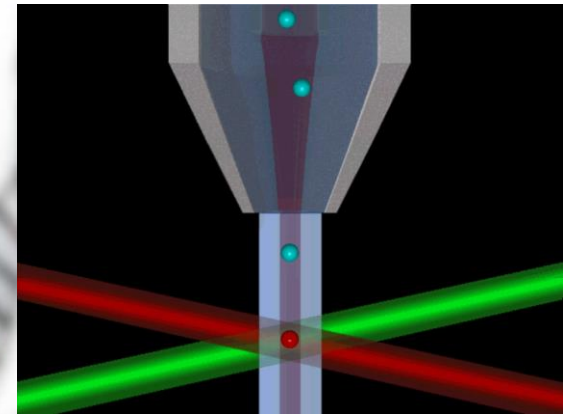
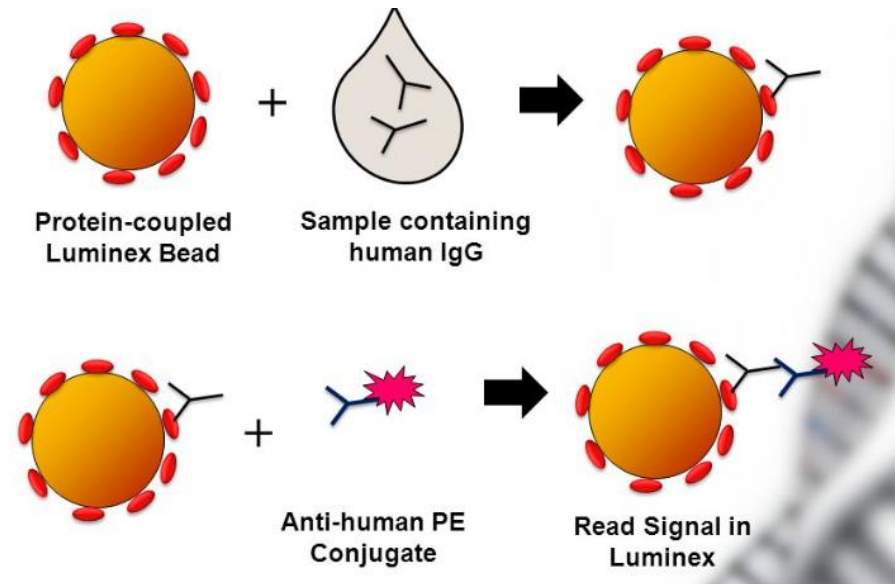
Window period

Expensive

- Serology tests helpful in diagnosis/management
 - Syphilis, EBV, HIV, Hepatitis –very useful
 - CMV, H pylori, fungal infections and HSV – may be useful

Method used for Syphilis and EBV

- Multiplex Bead Immune Assay



Syphilis

- Sexually transmitted disease
- Caused by the spirochete bacterium, *Treponema pallidum*
- The cork-screw shaped on darkfield microscopy
- Stages of syphilis
 - Primary
 - Secondary
 - Latent
 - Tertiary



Serologic testing

Treponemal: EIA, TP-PA, FTA-ABS, MHA-TP

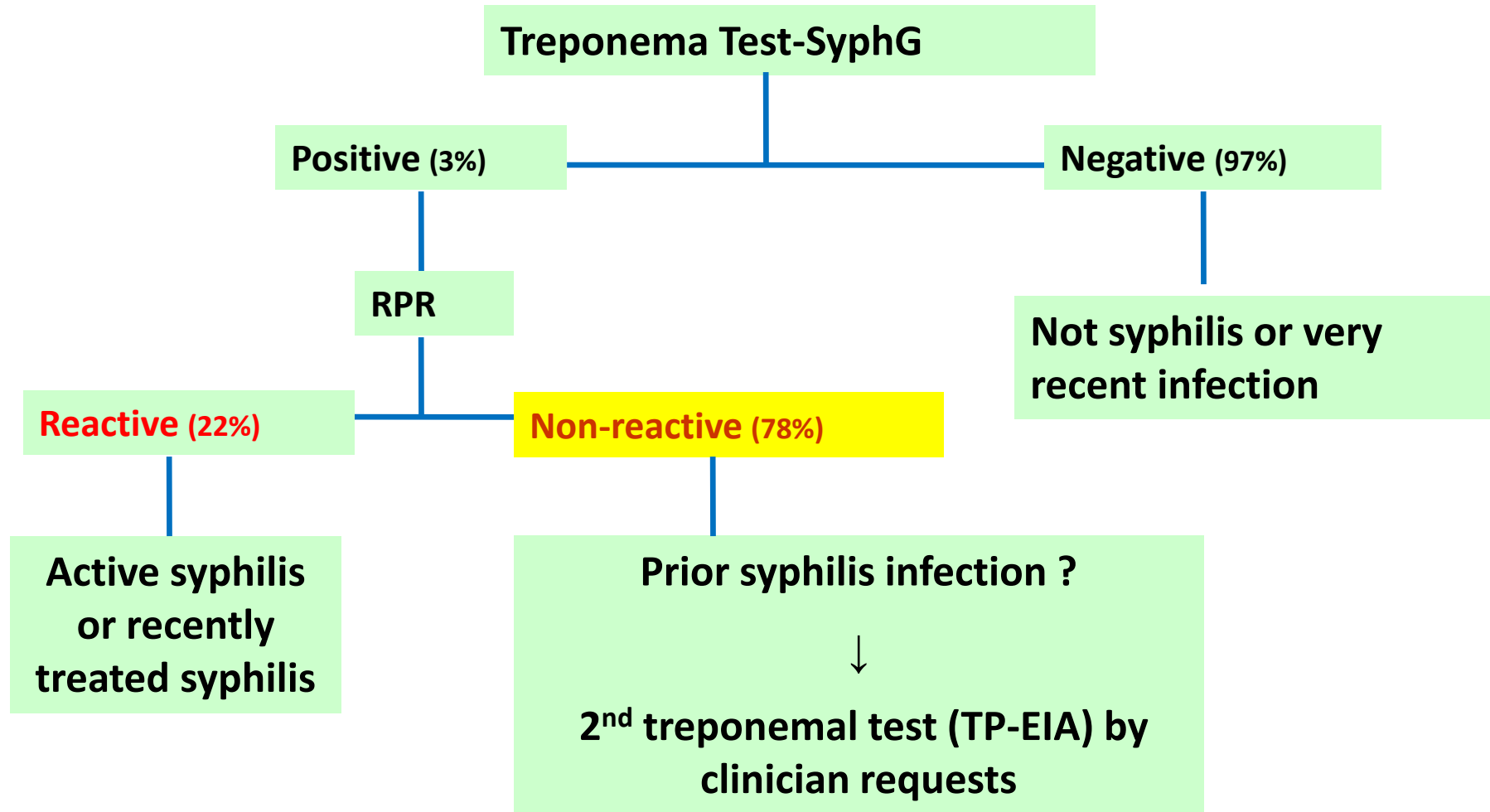
- Typically more specific than non-treponemal. Detect antibodies against specific antigens from *Treponema pallidum*

Non-treponemal: RPR, VDRL(CSF)

- Indirect method of blood test for diagnosis of infection with syphilis. Detect antibodies to nonspecific antigens that are released during cellular damage that occurred during syphilis spirochete infection.
- Both tests use cardiolipin-cholesterol-lecithin antigen to detect reaginic antibodies.

CCF Reverse Sequence Algorithm

Suitable for low disease prevalence



2nd treponemal test should be a different method from the 1st. CDC recommend TP-PA.

Epstein-Barr virus

- Infection of 90% of the world population
- Transmission by contact with salivary fluids of active viral shedders (“kissing disease”)
- Infects B/T cells, squamous cells, glandular cells (shedding virus in saliva)
- Burkitt's lymphoma,
- Nasopharyngeal carcinoma



MONOGEN BioKit Test

Agglutination methodology with latex particles coated with highly purified Paul-Bunnell antigen from bovine red cell membranes.



HIV Screening on Abbott Architect i2000

- 4th Generation Screening Assay (Antigen/Antibody Combo assay)
- Detection of **p24 antigen**
 - Increased sensitivity of testing
 - Shortened the window period
- 2nd tier HIV test (Geenius)
 - HIV 1/2 Differentiation Assay



2nd tier HIV test (Geenius)

HIV1 /2 Differentiation Assay

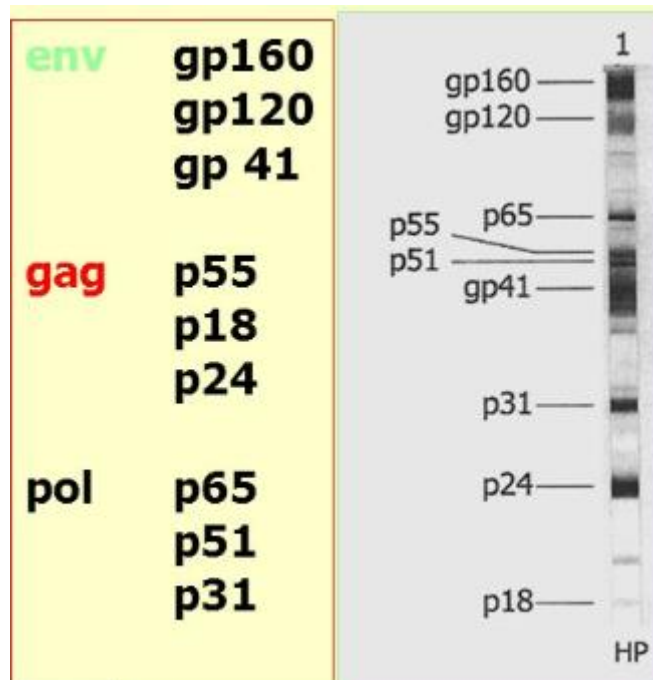


The Geenius™ HIV 1/2 Confirmatory Assay cassette contains a Control band (C) and six (6) test lines which are numbered on the cassette corresponding to the following:

Band 1:	gp36 (HIV-2, envelop peptide)	HIV-2 ENV
Band 2:	gp140 (HIV-2, envelop peptides)	HIV-2 ENV
Band 3:	p31 (HIV-1, polymerase peptide)	HIV-1 POL
Band 4:	gp160 (HIV-1, envelop recombinant protein)	HIV-1 ENV
Band 5:	p24 (HIV-1, core recombinant protein)	HIV-1 GAG
Band 6:	gp41 (Group M and O) (HIV-1, envelop peptides)	HIV-1 ENV
CTRL band:	Protein A	

BioRad HIV – 1/2 differentiation assay

Western Blot (WB) confirmation test

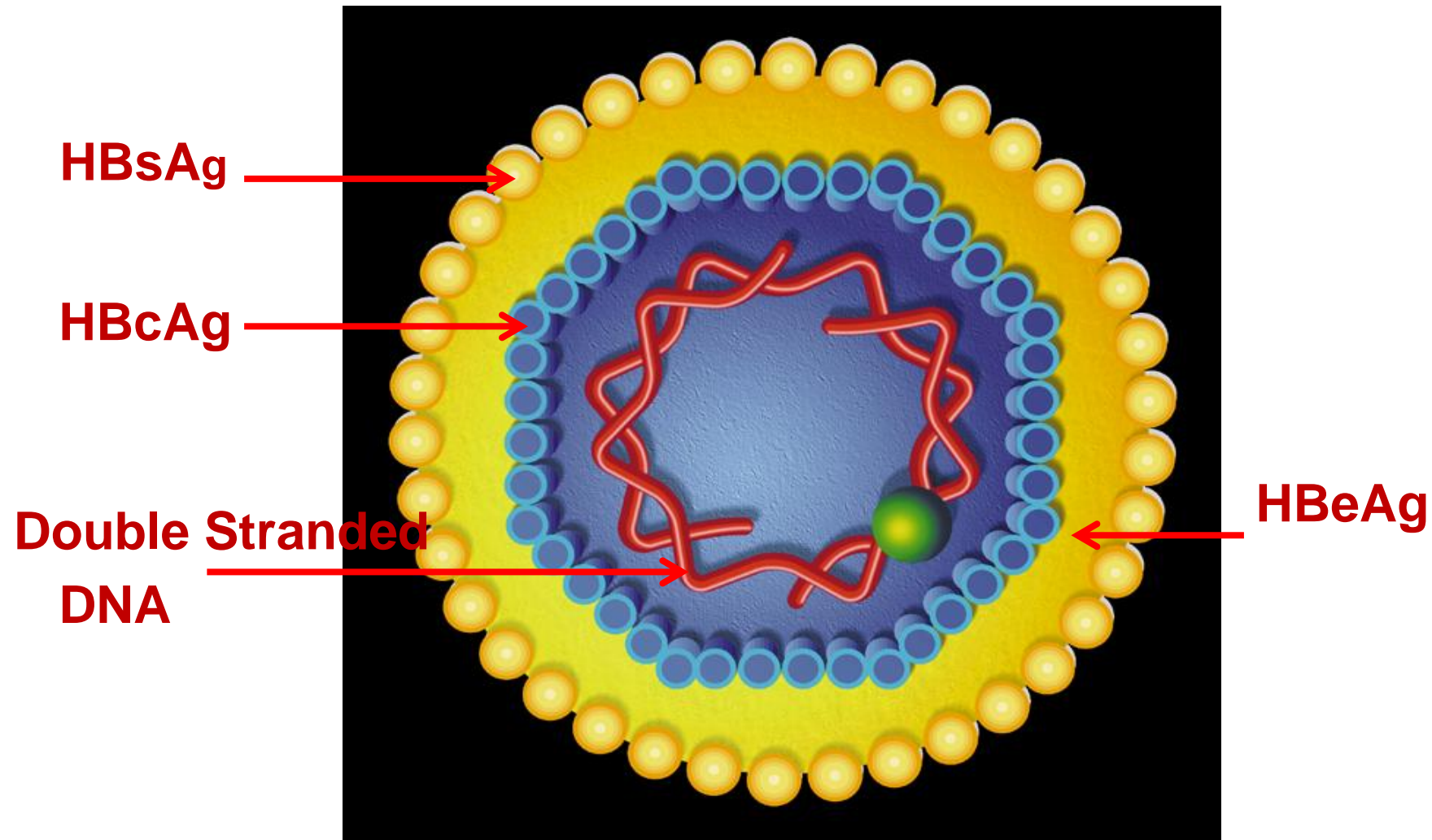


- Tests for specific **antibodies** in patient serum
- P24, gp41, and gp120/gp160

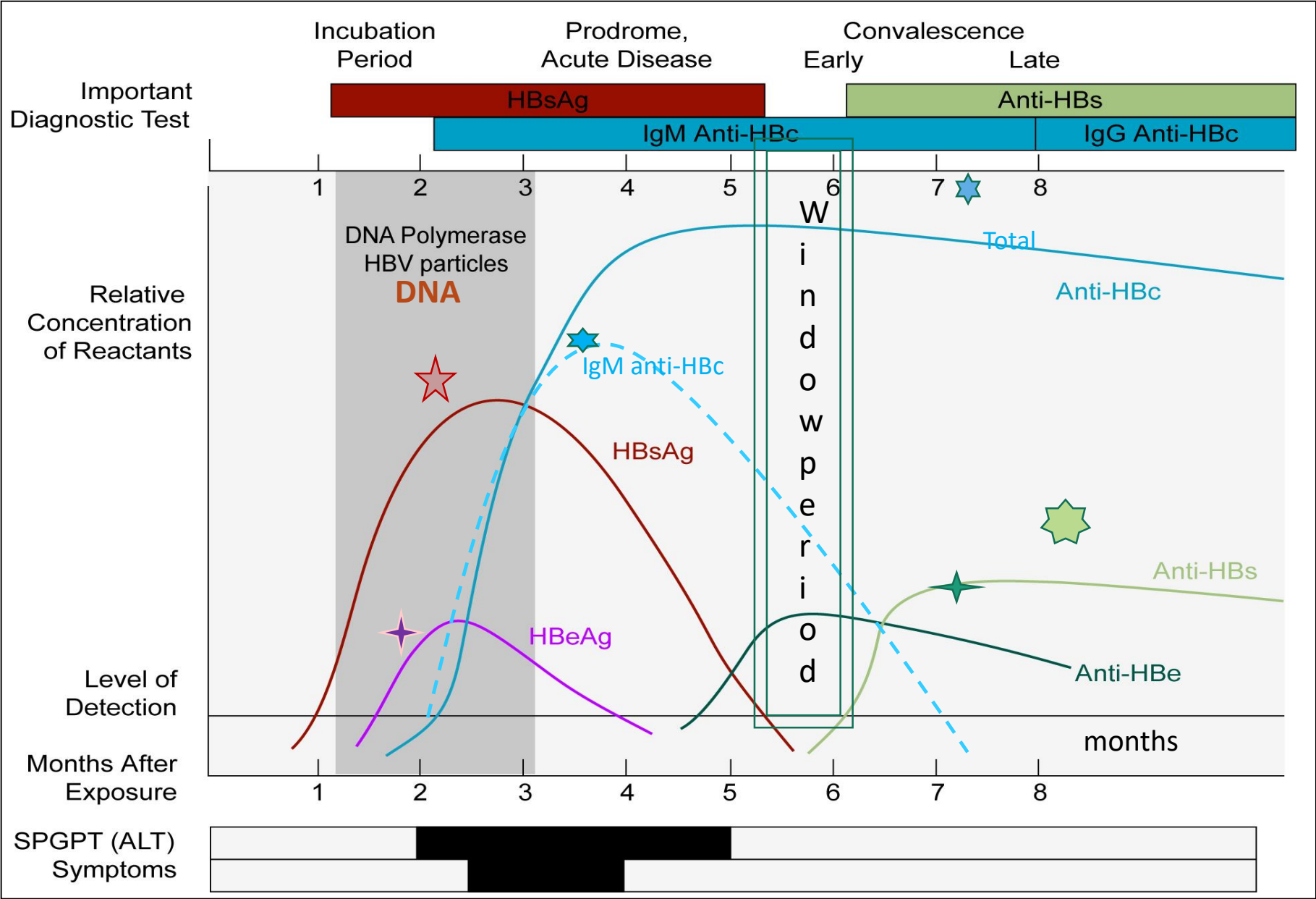
Viral Hepatitis - Overview

Virus	Transmission	Chronic Carrier	Vaccine	Disease Severity
HAV	Food	No	Yes	Not Severe, Disappears on Its Own
HBV	Blood, Sexual Intercourse, Mother-to-Child	Yes	Yes	No Severe Symptoms
HCV	Blood, Sexual Intercourse, Mother-to-Child	Yes	No	No Severe Symptoms
HDV	Blood, Sexual Intercourse, Mother-to-Child	Yes	No	Severe, High Mortality Rate
HEV	Food	No	No	Moderate

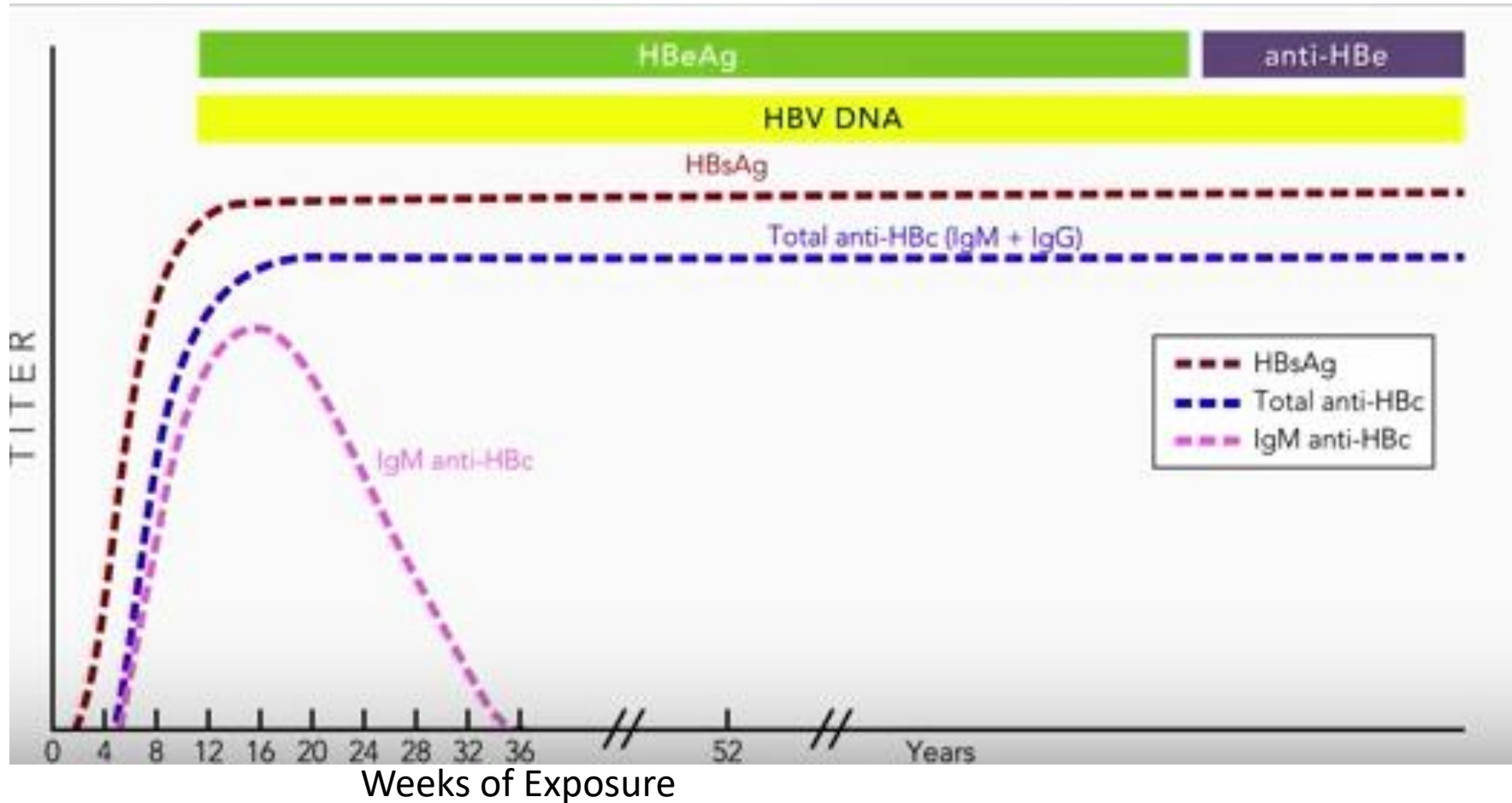
Hepatitis B Virus (Dane Particle)



Acute Hepatitis B with Recovery



Chronic Hepatitis B Serologic Course



Chronic HBV is diagnosed:

- When either HBsAg, HBe Ag or DNA is positive in at least two separate samples 6 months apart. **Or**
- Any positive result of any one of these three markers in a single sample and has a negative test for IgM ANTI-HBc.

Hepatitis B - Serologic Tests

Test	Positive Result Indication
HBs Ag	Active HBV infection (acute or chronic)
Anti-HBs Ab	Recovery/Immunity
Anti-HBc IgM Ab	Acute infection
Anti-HBc Total Ab	Current or past infection
HBe Ag	Active viral replication/contagious
Anti-HBe Ab	Convalescence/minimal replication

These tests usually utilize CIA method

CMV (Cytomegalovirus)

- 50-80 % of adults in US are infected by age 40
- Those at greatest risk of infection are fetuses and immunocompromised persons
- CMV is the most common virus transmitted to the fetus
- IgG and IgM antibody testing done on **Liaison** (chemiluminescent immunoassay)

TOXO (*Toxoplasmosis gondii*)

- Protozoan parasite
- Definitive host is house cat
- Effect on immunocompromised and organ transplant patients (TORCH)
- Congenital infection
- First –line test in endemic areas and in our Immunology lab

Test your Knowledge

- Patient X is a paramedic, 48 years of age. Laboratory work obtained during his annual physical examination reveals hyperlipidemia; CBC, glucose, BUN, and electrolytes were within normal range.
- After two months of a diet and exercise program, his cholesterol level is 256. Therefore, his physician elects to begin a lipid-lowering agent. A baseline liver profile is drawn prior to initiation of the medication. The liver profile reveals an AST of 226 Units/L and an ALT of 282 Units/L.
- In an effort to determine the cause of his elevated liver enzymes, the physician reviews Patient X's history and medications. He has been a paramedic for 25 years. He was immunized against HBV in 1988. During his career, he has experienced several exposures to blood (usually blood splashes, but also two needlesticks from IV needles). His most recent exposure was two years ago. An HIV test six months post-exposure was negative.
- The patient has never smoked. He drinks about six beers per week and rarely drinks hard liquor. He uses several herbal preparations including garlic, ginkgo, and an antioxidant preparation. The patient takes ibuprofen for pain, consuming 6 to 10 tablets (200 mg each) per month.
- Although alcohol consumption and herbal antioxidants can both cause liver inflammation, the degree of his liver inflammation is much higher than would be expected from limited use of these two factors. Therefore, the physician orders a hepatitis profile.
- The results include negative anti-HAV, negative HBsAg, positive HBsAb, and positive anti-HCV.
- What is patient's diagnosis?