



## HIV Diagnosis

Frank Maldarelli, MD  
Bethesda, MD

07/20/2026



## Disclosures of Financial Relationships with Relevant Commercial Interests

- None

### Question #1

- A 26-year-old otherwise healthy gay white man has his first HIV test as part of a new health plan
- The fourth-generation test is antibody reactive and antigen non-reactive
- A supplemental third generation HIV-1/2 ELISA is non-reactive, and an HIV RNA test does not detect HIV RNA

#### What is the most likely explanation for these results?

- A. This person HIV-infected and is an elite controller
- B. This person is HIV-infected but is in the window period for HIV infection
- C. This person is infected with an HIV variant that is not detected by the supplemental test
- D. This person is not HIV-infected

### Question #1

- A 26-year-old otherwise healthy gay white man has his first HIV test as part of a new health plan
- The fourth-generation test is antibody reactive and antigen non-reactive
- A supplemental third generation HIV-1/2 ELISA is non-reactive, and an HIV RNA test does not detect HIV RNA

#### What is the most likely explanation for these results?

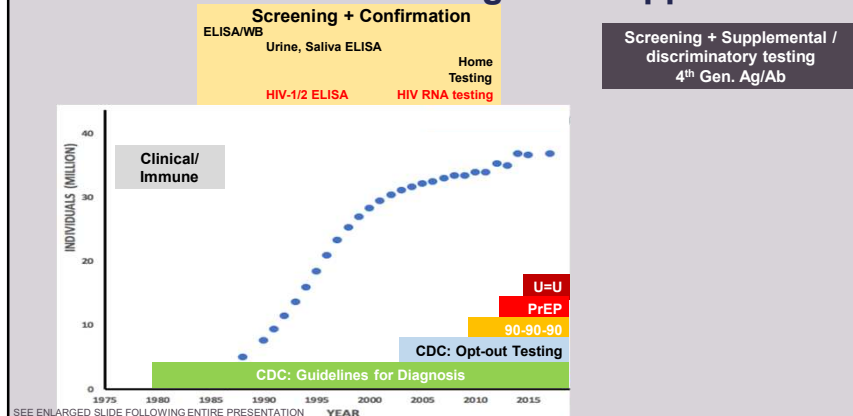
- A. This person HIV-infected and is an elite controller
- B. This person is HIV-infected but is in the window period for HIV infection
- C. This person is infected with an HIV variant that is not detected by the supplemental test
- D. This person is not HIV-infected\***

## HIV Diagnosis:

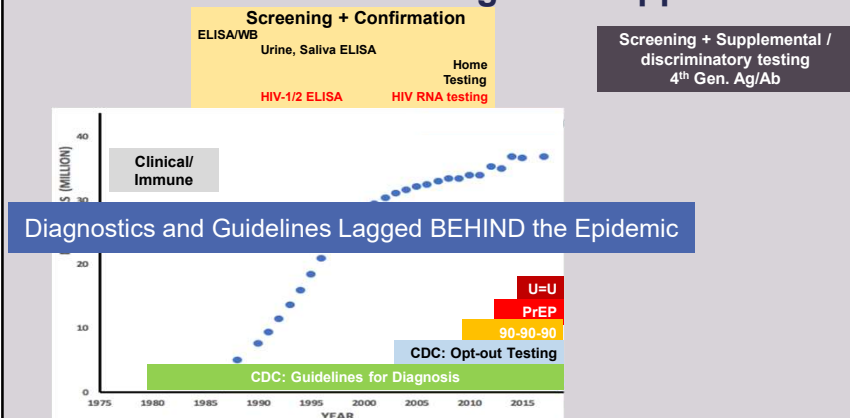
New Modalities and New Terminology Old Limitations Persist

- HIV Diagnosis
  - History
  - Physical
  - Laboratory testing
- Two Step Diagnostic Approach
- No Laboratory Test is Perfect
- False positive results require resolution
- PrEP places new stress on testing performance

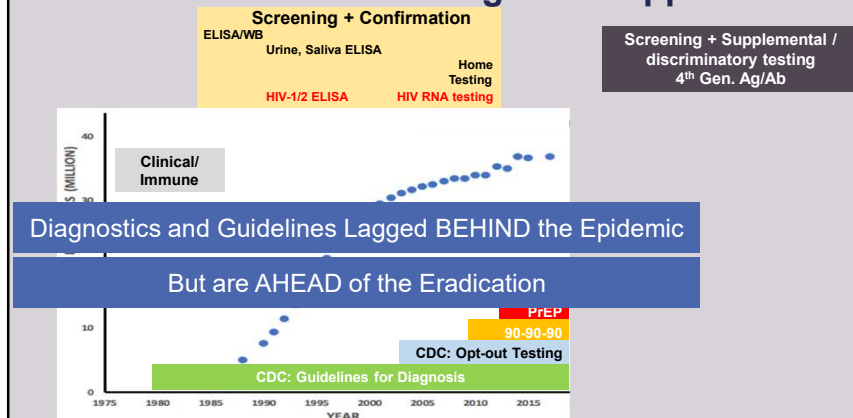
## Evolution of the HIV Diagnostic Approach



## Evolution of the HIV Diagnostic Approach



## Evolution of the HIV Diagnostic Approach



## Question #2

- 27-year-old female commercial sex worker working in Washington DC visits your clinic and requests PrEP
- She shows you her home HIV test, which she took yesterday, and which is non-reactive
- She has normal laboratory results and a negative pregnancy test

**Which of the following is most appropriate next step?**

- A. She can immediately initiate PrEP with tenofovir-FTC with no additional testing
- B. She requires additional testing with fourth generation Ag/Ab HIV test to determine whether she is infected with a non-B subtype of HIV-1 that is not detected by the home HIV test
- C. She requires additional testing with fourth generation HIV test to determine whether she has early HIV infection not detected by the home HIV test
- D. She should not initiate PrEP because PrEP does not work well in women

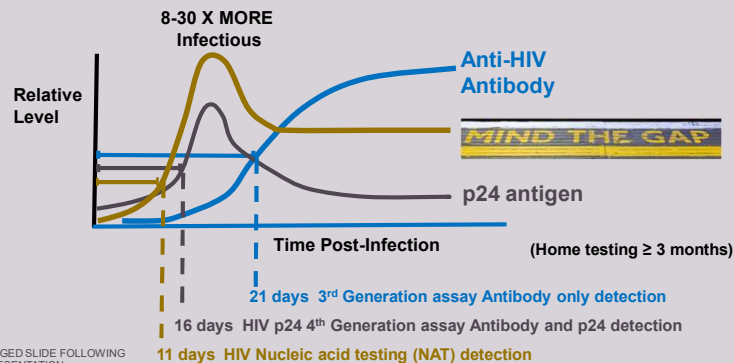
## Question #2

- 27-year-old female commercial sex worker working in Washington DC visits your clinic and requests PrEP
- She shows you her home HIV test, which she took yesterday, and which is non-reactive
- She has normal laboratory results and a negative pregnancy test

**Which of the following is most appropriate next step?**

- A. She can immediately initiate PrEP with tenofovir-FTC with no additional testing
- B. She requires additional testing with fourth generation Ag/Ab HIV test to determine whether she is infected with a non-B subtype of HIV-1 that is not detected by the home HIV test
- C. She requires additional testing with fourth generation HIV test to determine whether she has early HIV infection not detected by the home HIV test\***
- D. She should not initiate PrEP because PrEP does not work well in women

## HIV Detection: There is Always a Window Period



## Detecting HIV Infection TWO STEPS

- Screening - Highest Sensitivity
  - 4th gen ELISA for HIV antibody + p24 antigen detection
  - Qualitative HIV RNA
- Supplemental/Discriminatory - Highest Specificity
  - GEENIUS
    - Confirms HIV-1 or HIV-2

## Diagnosis of Early HIV Infection

### History, Physical, Laboratory Testing

- Most sensitive Modalities
  - 4<sup>th</sup> Generation
  - HIV RNA: APTIMA
- Less Sensitive Modalities
  - Oral or urine testing
  - Home testing (3-month window)
  - GEENIUS is LESS sensitive for EARLY infection compared with 4<sup>th</sup> gen testing
- FOLLOW UP and REPEAT testing
- Antiretroviral therapy may blunt serologic immune response from maturing

## Evaluation for HIV Infection During PrEP

- Every three months
- Includes detailed history and physical examination
- Ag/Ab (4<sup>th</sup> generation) testing preferred
- Viral RNA
  - Qualitative assay – FDA approved
  - Quantitative assay
    - >3000 copies/ml plasma cutoff
- DELAYED antibody emergence POSSIBLE in individuals infected during PrEP with extended-release cabotegravir

## Question #3

- You are following a couple who have had a planned pregnancy
- The man is HIV positive and 100% adherent with first line therapy with Tenofovir+3TC+Dolutegravir
- The woman has had monthly fourth generation HIV testing, which has been non-reactive throughout the first two trimesters
- On the most recent visit the man has an HIV RNA was <20 c/ml, but the woman has shows HIV antigen negative and HIV antibody positive

### What is the most appropriate next step?

- Obtain the HIV viral RNA test to find out how high the viral load is, and begin antiretroviral therapy immediately
- Consider laboratory error, repeat the same 4<sup>th</sup> generation test
- Perform supplemental testing with third generation discriminatory testing
- Reassure the couple that the woman is not infected and the test is just a false positive

## Question #3

- You are following a couple who have had a planned pregnancy
- The man is HIV positive and 100% adherent with first line therapy with Tenofovir+3TC+Dolutegravir
- The woman has had monthly fourth generation HIV testing, which has been non-reactive throughout the first two trimesters
- On the most recent visit the man has an HIV RNA was <20 c/ml, but the woman has shows HIV antigen negative and HIV antibody positive

### What is the most appropriate next step?

- Obtain the HIV viral RNA test to find out how high the viral load is, and begin antiretroviral therapy immediately
- Consider laboratory error, repeat the same 4<sup>th</sup> generation test
- C. Perform supplemental testing with third generation discriminatory testing\***
- Reassure the couple that the woman is not infected and the test is just a false positive

## HIV Serologic Testing Pregnancy

- False positive results with antibody testing are possible in pregnancy
- May be specific for individual's tests and persist during pregnancy
- Testing with viral RNA testing can resolve most issues
  - Qualitative tests (e.g., APTIMA) ARE FDA-APPROVED for testing
    - Expensive and generally longer turn around
  - Quantitative testing are NOT FDA-APPROVED for diagnosis
    - Rapid turnaround but low-level results are possible
- Rapid screening reactive during labor in previously untested
  - Initiate therapy
  - Do not wait for supplemental results

## Question #4

- A 65-year-old American male has had unprotected sex with men for many years
- The HIV-1/2 ELISA is reactive, and supplemental testing is positive for HIV-1
- Viral RNA level is <50 copies/ml and CD4 count is 700 cells/ $\mu$ l.
- He has never been on antiretroviral therapy and has no history of travel outside the US

### Which of the following is most likely?

- A. The patient is in the window period of HIV-1 infection
- B. The patient is chronically infected with HIV-1 and has a viral load too low to be detected because he is a long term non progressor
- C. The patient is not infected with HIV-1 or -2, all tests are false positive
- D. The patient is infected with non-B subtype of HIV-1

## Question #4

- A 65-year-old American male has had unprotected sex with men for many years
- The HIV-1/2 ELISA is reactive, and supplemental testing is positive for HIV-1
- Viral RNA level is <50 copies/ml and CD4 count is 700 cells/ $\mu$ l.
- He has never been on antiretroviral therapy and has no history of travel outside the US

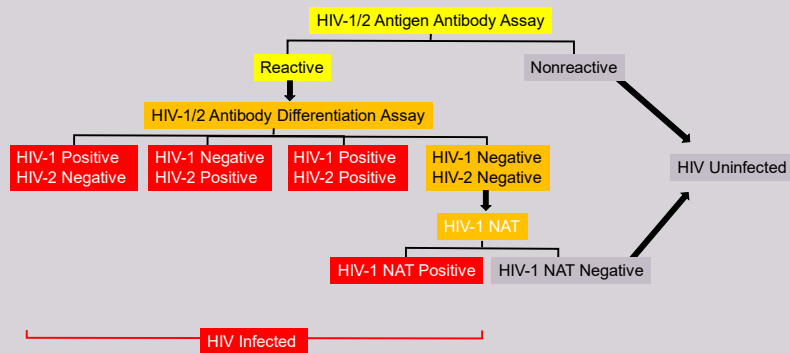
### Which of the following is most likely?

- A. The patient is in the window period of HIV-1 infection
- B. The patient is chronically infected with HIV-1 and has a viral load too low to be detected because he is a long term non progressor\***
- C. The patient is not infected with HIV-1 or -2, all tests are false positive
- D. The patient is infected with non-B subtype of HIV-1

## HIV-1 Long Term Non-Progressors

- Represents authentic HIV infection
- ELISA REACTIVE
- SUPPLEMENTAL POSITIVE
- HIV RNA may not be detectable
- Slow disease progression
- Associated with specific HLA subtypes

## HIV Diagnostic Algorithm



SEE ENLARGED SLIDE FOLLOWING ENTIRE PRESENTATION

## Question #5

- A 42-year-old woman has a reactive 4<sup>th</sup> generation test for HIV infection
- She is 7 months pregnant and had COVID-19 infection one month ago despite vaccination with Moderna COVID vaccine four months prior to testing
- She had a nonreactive 4<sup>th</sup> generation screen 7 months ago at the beginning of her pregnancy, she denies any HIV exposures
- Subsequent qualitative HIV RNA testing is negative

### What is the most likely explanation for these results?

- False positive 4<sup>th</sup> generation test for HIV infection due to pregnancy
- False positive 4<sup>th</sup> generation test for HIV infection due to COVID vaccination
- False positive 4<sup>th</sup> generation test for HIV infection due to COVID infection
- False negative HIV RNA testing in the setting of recent HIV infection

## Question #5

- A 42-year-old woman has a reactive 4<sup>th</sup> generation test for HIV infection
- She is 7 months pregnant and had COVID-19 infection one month ago despite vaccination with Moderna COVID vaccine four months prior to testing
- She had a nonreactive 4<sup>th</sup> generation screen 7 months ago at the beginning of her pregnancy, she denies any HIV exposures
- Subsequent qualitative HIV RNA testing is negative

### What is the most likely explanation for these results?

- False positive 4<sup>th</sup> generation test for HIV infection due to pregnancy
- False positive 4<sup>th</sup> generation test for HIV infection due to COVID vaccination
- False positive 4<sup>th</sup> generation test for HIV infection due to COVID infection\***
- False negative HIV RNA testing in the setting of recent HIV infection

## MANY HIV Screening False Positives

### Infections

- COVID
- African trypanosomiasis
- Babesiosis
- Schistosomiasis
- Malaria
- Dengue
- Mycobacterial
- Hepatitis A/C
- Hematologic
  - Polyclonal B cell responses
  - Heterophile antibodies
  - Workers in pork processing plant
  - B cell lymphoma
  - Autoimmune hemolytic anemia
  - Angioblastic T cell lymphoma
  - Hodgkin lymphoma

### Rheumatologic Diseases

- Lupus
- Sjogren
- Rheumatoid
- Latrogenic
  - Vaccines
  - Rabies
  - T-dap
  - Hepatitis B
  - SARS-CoV-2
  - HIV vaccine trials
  - Lentiviral therapy (e.g., CAR-T)

SEE ENLARGED SLIDE FOLLOWING ENTIRE PRESENTATION

## HIV Testing

- Opt-out testing is Recommended by IDSA and CDC
  - Patients are informed that an HIV test will be conducted unless they explicitly decline to be tested
  - Written consent in this setting is incorporated into intake
  - Counseling is available
- Opt-in: NOT Recommended by IDSA and CDC
  - Patients need to initiate the request for HIV infection
- Requirements for testing: FIVE C's:
  - Counseling
  - Consent
  - Confidentiality
  - Correct test results
  - Connection to prevention care and treatment

## Pearls for Board Exam

### HIV Testing is Comprehensive

- Non-B Subtypes are all detectable
- HIV-2 has an approved diagnosis
- Long term Non-Progressor
  - ELISA reactive / Supplemental Positive

### Resources:

<https://www.cdc.gov/hiv/guidelines/testing.html>  
[Fmaldarelli3@gmail.com](mailto:Fmaldarelli3@gmail.com)

Reference slides follow

### No Test is Perfect

- 4<sup>th</sup> Gen less sensitive
  - Acute
  - PEP/PrEP
  - Early Antiretroviral therapy
- False Positives
  - Pregnancy
- Mind the gap
  - Long gap for Home testing

**Board exam isn't perfect either...so don't overthink it**

