**Centers for Disease Control and Prevention**
**Epidemiology Program Office**
**Case Studies in Applied Epidemiology**
**No. 871-703**

# Screening for Antibody to the Human Immunodeficiency Virus

## Student's Guide

Learning Objectives
After completing this case study, the participant should be able to:

- ☐ Define and perform calculations of sensitivity, specificity, predictive-value positive, and predictive-value negative;
- ☐ Describe the relationship between prevalence and predictive value;
- ☐ Discuss the trade-offs between sensitivity and specificity;
- ☐ List the principles of a good screening program.

This case study was developed in 1987 by Lyle Peterson, Guthrie Birkhead, and Richard Dicker.

**U.S. DEPARTMENT OF HEALTH AND HUMAN SERVICES**
**Public Health Service**

# PART I

In December 1982, a report in the *MMWR* described three persons who had developed acquired immunodeficiency syndrome (AIDS) but who had neither of the previously known risk factors for the disease: homosexual/bisexual activity with numerous partners and intravenous drug use. These three persons had previously received whole-blood transfusions. By 1983, widespread recognition of the problem of transfusion-related AIDS led to controversial recommendations that persons in known high-risk groups voluntarily defer from donating blood. In June 1984, after the discovery of the human immunodeficiency virus (HIV), five companies were licensed to produce enzyme-linked immunosorbent assay (EIA, then called ELISA) test kits for detecting HIV antibody. A Food and Drug Administration (FDA) spokesman stated that, "...getting this test out to the blood banks is our No. 1 priority...." Blood bank directors were anxiously waiting to start screening blood with the new test until March 2, 1985, the date the first test kit was approved by the FDA.

In the pre-licensure evaluation, sensitivity and specificity of the test kits were estimated using blood samples from four groups: those with AIDS by CDC criteria, those with other symptoms and signs of HIV infection, those with various autoimmune disorders and neoplastic diseases that could give a false-positive test result, and presumably healthy blood and plasma donors.

Numerous complex issues were discussed even before licensure. Among them were understanding the magnitude of the problem of false-positive test results, and determining whether test-positive blood donors should be notified.

It is now March 2, 1985. The first HIV antibody test kits will arrive in blood banks in the state in a few hours. Meeting with State Epidemiologist to discuss the appropriate use of this test are the Commissioner of Health, the medical director of the regional blood bank, and the chief of the State Drug Abuse Commission.

To help in the discussions, the State Epidemiologist turns to pre-licensure information regarding the sensitivity and specificity of test kit A. The information indicates that the sensitivity of test kit A is 95.0% (0.95) and the specificity is 98.0% (0.98). These and related measures are reviewed below.

NOTES ON SENSITIVITY AND SPECIFICITY

| | Actual antibody status | | |
|---|---|---|---|
| Test result | Present | Absent | Total |
| Positive | True positive (A) | False positive (B) | All positive tests (A+B) |
| Negative | False negative (C) | True negative (D) | All negative tests (C+D) |
| Total | All with antibody (A+C) | All without antibody (B+D) | Total (A+B+C+D) |

Sensitivity - the probability that the test result will be positive when administered to persons who actually have the antibody.

= true positives / all with antibody

Algebraically, sensitivity = A / (A+C)

Specificity - the probability that the test result will be negative when administered to persons who are actually without the antibody.

= true negatives / all without antibody

Algebraically, specificity = D / (B+D).

Predictive-value positive (PVP) - the probability that a person with a positive screening test result actually has the antibody.

= true positives / all with positive test

Algebraically, PVP = A / (A+B).

Predictive-value negative (PVN) - the probability that a person with a negative screening test result actually does not have the antibody.

= true negatives / all with negative test

Algebraically, PVN = D / (C+D).

**Question 1:** With this information, by constructing a 2-by-2 table, calculate the predictive-value positive and predictive-value negative of the EIA in a hypothetical population of 1,000,000 blood donors. Using a separate 2-by-2 table, calculate PVP and PVN for a population of 1,000 drug users. Assume that the actual prevalence of HIV antibody among blood donors is 0.04% (0.0004) and that of intravenous drug users is 10.0% (0.10).

The blood bank director wants assistance in evaluating the EIA as a test for screening donor blood in the state. In particular, she is concerned about the possibility that some antibody-positive units will be missed by the test, and she wonders about false-positive test results since she is under pressure to develop a notification procedure for EIA-positive donors.

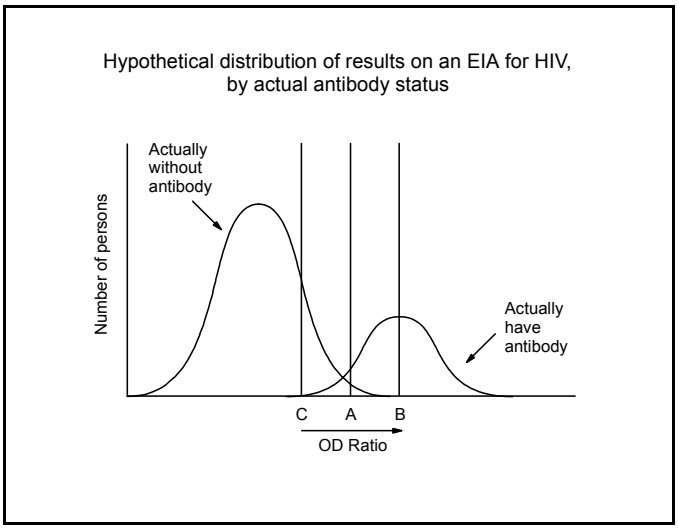
**Question 2**: Do you think that the EIA is a good screening test for the blood bank? What would you recommend to the blood bank director about notification of EIA-positive blood donors?

The chief of the State Drug Abuse Commission has noticed a dramatic increase in AIDS among clients in his intravenous-drug-abuse treatment programs. For planning purposes, he wants to do a voluntary HIV antibody seroprevalence survey of intravenous-drug-abuse clients and would like to assess the feasibility of using the test results as part of behavior-modification counseling.

**Question 3**: Do you think that the EIA performs well enough to justify informing test-positive clients in the drug abuse clinics that they are positive for HIV?

**Question 4:** If sensitivity and specificity remain constant, what is the relationship of prevalence to predictive-value positive and predictive-value negative?

EIA results are recorded as optical-density (OD) ratios. The OD ratio is the ratio of absorbance of the tested sample to the absorbance of a control sample. The greater the OD ratio, the more "positive" is the test result. The EIA, as with most other screening tests, is not perfect; there is some overlap of optical-density ratios of samples that are actually antibody positive and those that are actually antibody negative. This is illustrated in the following figure.




Establishing the cutoff value to define a positive test result from a negative one is somewhat arbitrary. Suppose that the test manufacturer intiially considered that optical density ratios greater than "A" on the above figure would be called positive.

**Question 5:** In terms of sensitivity and specificity, what happens if you raise the cutoff from "A" to "B"?

**Question 6:** In terms of sensitivity and specificity, what happens if you lower the cutoff from "A" to "C"?

**Question 7:** From what you know now, what is the relationship between sensitivity and specificity of a screening test.

**Question 8:** Where might the blood bank director and the head of drug treatment want the cutoff point to be for each program? Who would probably want a lower cutoff value?

## PART II

The blood bank director is concerned that, because of the low predictive-value positive of the EIA in the blood donor population, the blood bank personnel cannot properly inform those who are EIA positive of their actual antibody status. For this reason, he wishes to evaluate the Western blot test as a confirmatory test for HIV antibody.

The Western blot test identifies antibodies to specific proteins associated with the human immunodeficiency virus. The Western blot is the most widely used secondary test to detect HIV antibody because its specificity exceeds 99.99%; however, it is not used as a primary screening test because it is expensive and technically difficult to perform. Its sensitivity is thought to be lower than that of the EIA. Because the Western blot test is not yet generally available, the blood bank director is wondering whether the initial EIA-positive results can be confirmed by repeating the EIA and by considering persons to have the antibody only if results of both tests are positive.

The State Epidemiologist suggests that they compare the performance of the repeat EIA and the Western blot as confirmatory tests. To do this, they will use the earlier hypothetical sample of 1,000,000 blood donors. They assume that serum specimens that are initially positive by EIA are then split into two portions; a repeat EIA is performed on one portion and a Western blot on the other portion.

**Question 9:** What is the actual antibody prevalence in the population of persons whose blood samples will undergo a second test?

**Question 10:** Calculate the predictive-value positive of the two sequences of tests: EIA-EIA and EIA-Western blot. Assume that the sensitivity and specificity of the EIA are 95.0% and 98.0%, respectively. Assume that the sensitivity and specificity of the Western blot are 80.0% and 99.99%, respectively. Also assume that the tests are independent, even though they may not be (e.g., those with cross-reactive proteins are likely to cross-react each time).

**Question 11:** Why does the predictive-value positive increase so dramatically with the addition of a second test? Why is the predictive value positive higher for the EIA-WB sequence than for the EIA-EIA sequence?

It is now July 1987 and the Governor has asked the State Epidemiologist to evaluate a proposed premarital HIV-antibody-screening program. A bill to establish the program is to be presented to the state legislature tomorrow. An estimated 60,000 people will get married in the state in the next year. The proposed legislation requires that each prospective bride and groom submit a blood sample for EIA testing. Samples that test positive by EIA will undergo confirmatory Western blot testing.

The legislation describes the goal of the screening program to be to decrease inadvertent perinatal or sexual HIV transmission by determining who among those to be married are probably infected with the virus.

**Question 12:** What criteria would you consider in evaluating this proposed screening program?

The following two tables show the results of the testing, assuming that persons getting married have the same actual HIV antibody prevalence as blood donors (0.04%). In 1987, the sensitivity and specificity of the improved EIA Test Kit A available at the time were 97.0% and 99.8%, respectively. The Western blot sensitivity and specificity were 95.0% and 99.99%, respectively.

| | Actual antibody status | | | |
|---|---|---|---|---|
| Initial EIA | Present | Absent | Total | |
| Positive | 23 | 120 | 143 | (These 143 will undergo Western blot testing) |
| Negative | 1 | 59,856 | 59,857 | |
| Total | 24 | 59,976 | 60,000 | |

| Follow-up Western blot | Present | Absent | Total |
|---|---|---|---|
| Positive | 22 | 0 | 22 |
| Negative | 1 | 120 | 121 |
| Total | 23 | 120 | 143 |

With sequential tests: Sensitivity of 92%
Specificity of 100%
Predictive-value positive of 100%

**Question 13:** Compute the cost of the screening program. Assume a cost of $50.00 for every initial EIA test ($10.00 lab fee and $40.00 health-care-provider visit) and an additional $100.00 for EIA-positive persons who will need additional testing. What is the cost of the screening program in the next year? What is the cost per identified antibody-positive person?

**Question 14:** What is your final recommendation to the Governor?

THE NEW YORK TIMES NATIONAL SUNDAY, JUNE 25, 1989

# *Illinois Legislature Repeals Requirement for Prenuptial AIDS Tests*

**By ISABEL WILKERSON**
Special to The New York Times

SPRINGFIELD, ILL., June 23 - At the urging of health officials and AIDS specialists, the Illinois Legislature repealed Friday night the only law in the country requiring prenuptial testing for the AIDS virus.

The measure now goes to Gov. James R. Thompson. He has consistently declined comment on whether he will sign it, although pressure on him to do so is intense, including that of his State Health Director, Dr. Bernard Turnock.

A similar testing law in Louisiana was repealed last year, six months after it took effect.

"We made a mistake and we ought to admit it,"said Bill Marovitz, a State Senator from Chicago, urging his colleagues to overturn the testing law.

Prenuptial testing began in Illinois in January 1988 over the strong objection of both the Illinois Department of Public Health and AIDS policy experts.

They said it was an inefficient and expensive way to identify carriers of the virus and diverted already scarce resources from those most at risk.

**44 Positive Out of 221,000**

Since then, the tests, which detect the antibodies that indicate infection with the human immunodeficiency virus which cases AIDS, have turned up few cases of the disease. Of the 221,000 people who took marriage vows in Illinois since the law took effect, 44 were infected with the HIV virus, tests indicated, and health officials suspect that nearly a dozen of those results may be false. Since the testing was confidential, health officials do not know the outcome of these cases.

The tests have also led thousands of people to leave the state to get married and undetermined numbers of others to put off marriage altogether, health officials said.

Marriages in Illinois fell by nearly a quarter from 99,212 in 1987 to 77,729 in 1988, although the numbers are up slightly so far this year over 1988.

AIDS specialists hailed the repel legislation as long overdue. "It's a 'we-told-you-so' situation," said Andrew Deppe, a spokesman for the AIDS Foundation of Chicago. "Illinois has become a national laughingstock. We've had to spend our energy putting out brush fires instead of working on prevention."

But Penny Pullen, a Republican State Representative from suburban Cook County, who sponsored the prenuptial AIDS testing bill, said repeal of the law would hurt the state's efforts to curb the spread of the virus, "This is a major mistake," Ms.Pullen said. "This will send an unfortunate message to the people of Illinois and the rest of the nation that AIDS is not as serious an epidemic as it was two years ago. And that message is a lie."

**Fewer Than Predicted**

She pointed to an increase in the number of positive test results in the first half of this year as evidence that the law was working. So far this year, the tests have indicated 18 cases of the AIDS virus among 66,500 newly betrothed people, as against 8 cases among 59,000 people in the same period last year, the Illinois Department of Public Health said.

But officials of the health department said that even with that increase, the agency had found far fewer case in the 18 months of mandatory testing than the 120 cases it originally predicted would be found each year.

The agency also found that the rate of infection among engaged couple's was comparable to those of other low-risks groups. Engaged couples in Illinois and blood donors, both groups considered at very low risk, have rates of infection of about 2 per 10,000.

"The overall rate among these couples is close to the lowest rate ever recorded in this country." said Tom Schafer, a spokesman for the Illinois Department of Public Health.

While even critics say the law has been useful in raising awareness of the AIDS epidemic, state health officials said it was an expensive way to detect carriers of the virus . The test costs each person from $30 to $125, depending on whether testing is done in clinics or in a doctor's office and whether follow-up testing is required. The total cost for Illinois couples last year was $5.4 million, or about $209,00 for each case of HIV infection detected.

# Appendix 1

The following 10 principles of successful mass screening programs were proposed by Wilson and Jungner of the World Health Organization in 1968:

1. The condition being sought is an important health problem for the individual and the community.
2. There is an acceptable form of treatment for patients with recognizable disease.
3. The natural history of the condition, including its development from latent to declared disease, is adequately understood;
4. There is a recognizable latent or early symptomatic stage.
5. There is a suitable screening test or examination for detecting the disease at the latent or early symptomatic stage, and this test is acceptable to the population.
6. The facilities required for diagnosis and treatment of patients revealed by the screening program are available.
7. There is an agreed policy on whom to treat as patients.
8. Treatment at the pre-symptomatic, borderline stage of a disease favorably influences its course and prognosis.
9. The cost of the screening program (which would include the cost of diagnosis and treatment) is economically balanced in relation to possible expenditure on medical care as a whole.
10. Case-finding is a continuing process, not a "once and for all" project.

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SUMMARY OF SCREENING TEST MEASURES

| | Condition Truly Present | Condition Truly Absent | |
|---|---|---|---|
| Test positive | True Positive | False Positive | Total Testing Positive |
| Test negative | False Negative | True Negative | Total Testing Negative |
| Total | True Prevalence | 1 – Prevalence | Size of Population |

$$Sensitivity = Prob(T+ \mid D+) = \frac{TP}{TP + FN}$$

$$Specificity = Prob(T- \mid D-) = \frac{TN}{TN + FP}$$

$$Predictive\ value\ positive = Prob(D+ \mid T+) = \frac{TP}{TP + FP}$$

$$Predictive\ value\ negative = Prob(D- \mid T-) = \frac{TN}{TN + FN}$$

Bayes Theorem Formulas for PVP and PVN:

$$PVP = \frac{(Sensitivity)(Prevalence)}{(Sensitivity)(Prevalence) + (1 - Specificity)(1 - Prevalence)}$$

$$1 - PVN = \frac{(1 - Sensitivity)(Prevalence)}{(1 - Sensitivity)(Prevalence) + (Specificity)(1 - Prevalence)}$$