EDITORIAL COMMENTARY

Determining HIV Incidence in Populations: Moving in the Right Direction

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(See the major article by Laeyendecker et al, on pages 232–9 and the major article by Eshleman et al, on pages 223–31.)

Incidence, the number of new infections in a population over time, is the most important epidemiologic parameter for human immunodeficiency virus (HIV), as it is for other infectious diseases [1–3]. Accurate determinations of HIV incidence are essential to characterize and monitor the epidemic, to identify populations at highest risk, and to assess the effectiveness of interventions. HIV incidence is also used as the end point in community randomized prevention trials. The *AIDS 2012* International AIDS Conference in Washington, D.C., generated renewed enthusiasm for using the new treatment and prevention tools in our armamentarium to reverse the epidemic; we need to be able to measure HIV incidence to assess progress. However, in the fourth decade of the HIV/AIDS pandemic, we continue to be constrained by the inadequate tools available to efficiently and accurately determine HIV incidence.

Traditional methods of HIV incidence determination are often complex and expensive or imprecise and unreliable [1–3]. The direct measurement of HIV incidence through the prospective follow up of cohorts of HIV-negative persons is logistically complicated and expensive and often unrepresentative of the larger population; the interventions involved in prospective cohort studies result in changes in behavior and HIV incidence. Use of mathematical models to estimate incidence on the basis of HIV prevalence, AIDS cases, and deaths has a role, but input parameters are often not available, and estimates usually lack precision [3, 4].

Given these limitations, since 1998, substantial efforts have focused on the identification of specimens from HIV-seropositive persons who were recently infected (eg, within the past 6 months) through the use of laboratory assays in a method known as either a serologic testing algorithm for recent HIV seroconversion (STARHS) or a recent infection testing algorithm (RITA) [1, 2, 5–8]. These assays are based on the maturation of the antibody response to HIV infection, in terms of quantity, proportion, avidity, isotype or specificity [7, 8]. With the ability to accurately determine which HIV-positive persons were recently infected and the duration of the recent period, HIV incidence can be estimated from a cross-sectional survey. However, the development and evaluation of HIV incidence assays that are based on detection of recent infection has been fraught with pitfalls. Initially, investigators used modified commercial enzyme immunoassays (EIAs) that were made less sensitive or "detuned" [2]. These assays were based in HIV type 1 (HIV-1) subtype B, which is prevalent in North America and Europe, and were found to have different performance characteristics for other HIV-1 subtypes, limiting their global usefulness. Furthermore, the assays first used for HIV incidence determination were taken off the market and became unavailable [1, 8]. The perceived market for HIV incidence assays, which are used mainly as tools for surveillance and research, has not driven a great deal of interest from industry [2, 9].

To address the problem of HIV-1 subtype differences, scientists at the US Centers for Disease Control and Prevention (CDC) developed the BED capture EIA (BED-CEIA), which is based on multiple HIV-1 subtypes (B, E [CRF01_AE], and D) [6]. The BED-CEIA's performance is based on the proportion of immunoglobulin G that is anti-HIV. The assay was licensed and made commercially available for surveillance purposes [2, 9]. While the assay performed better for some other subtypes [10], within a few years of use in research studies and surveillance surveys, it became clear that the BED-CEIA misclassified a substantial proportion of specimens from persons with long-standing infection as recent infection, resulting in overestimations of HIV incidence [1, 2]. In 2005, UNAIDS advised

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that the BED-CEIA not be used for routine surveillance applications [1, 2]. Subsequent studies indicated that the misclassification of long-standing infection as recent was associated with low anti-HIV antibody levels in persons receiving antiretroviral therapy, with advanced HIV disease, with naturally low HIV levels, or because of variability in the immune response [1, 2, 8]. The measurement and use of false-recent rates was introduced as a correction factor for this problem [1, 2, 11–13]. However, BED-CEIA false-recent rates have been found to vary widely in various settings, necessitating local determination of such rates [1, 2]. Despite these challenges, the CDC continues to use the BED-CEIA as part of the system for HIV incidence estimation in the United States by accounting for the limitations of the assay in a complex HIV case-based surveillance system [14–16]. Until recently, the CDC-developed BED-CEIA was the only commercially available HIV incidence assay and has been, by far, the most widely used globally [1, 2, 9]. Unfortunately, the performance limitations described above and the 2005 UNAIDS statement advising caution regarding its use have resulted in confusion among public health scientists charged with determining HIV incidence in populations. The World Health Organization Working Group on HIV Incidence Assays (http://www.who.int/diagnostics_laboratory/links/hiv_incidence_assay/en/index.html) has provided some guidance [17], but lack of a sufficiently accurate assay remains a serious limitation.

In the past several years, a variety of assays have been developed and evaluated; however, rigorous evaluations require a large collection of geographically and HIV-1–subtype diverse specimens from persons with both early and long-standing HIV infection [1, 8]. Such specimens are difficult to collect and assemble and are not widely available. The Bill & Melinda Gates Foundation is supporting the Consortium for the Evaluation and Performance of HIV Incidence Assays (http://incidence-estimation.com/page/cephia) as part of a collaborative effort to address these challenges and clarify the critical path to HIV incidence assay development [18].

In this issue of the *Journal*, Laeyendecker and colleagues present data on the evaluation of a new tool, a multiassay algorithm for HIV incidence estimation [19]. This algorithm employs the BED-CEIA, an antibody avidity assay, the HIV RNA level, and the $CD4^+$ T-cell count. HIV RNA levels and $CD4^+$ T-cell counts were used to exclude persons with long-standing HIV infection who were elite controllers of HIV infection or had advanced HIV disease, respectively, and might have low HIV antibody levels. To evaluate the performance of this algorithm, the study team assembled a collection of 1782 samples from 709 individuals in the United States with known duration of HIV infection. Available data indicate that these specimens were almost exclusively from persons infected with HIV-1 subtype B [19]. The analysis demonstrated excellent ability of this algorithm to identify samples from recently infected persons (with a postserconversion window period of 141 days) as indicative of recent infection. Only 0.4% of 1474 samples from persons infected for >1 year were misclassified as indicative of recent infection. The team went on to compare the annual incidence of HIV seroconversion, measured prospectively in the HIV Network for Prevention Trials study 001 cohort (1.04% [95% confidence interval [CI], .70%–1.55%), with annual incidence estimated from cross-sectional specimens tested with the multiassay algorithm and found a very similar rate (0.97% [95% CI, .51%–1.71%]).

These findings on the performance of the multiassay algorithm are encouraging. However, a similar rigorous analysis will be required to further assess this algorithm for different HIV-1 subtypes from other populations in other geographic locations, including those with a more complicated HIV molecular epidemiology involving multiple subtypes and recombinant HIV-1 strains. Furthermore, the requirement of a $CD4^+$ T-cell count is logistically challenging because it cannot be measured in stored specimens, and the use of HIV RNA assays further increases the cost.

Also in this issue of the *Journal*, Eshleman and colleagues compare 3 methods of HIV incidence determination, including the multiassay algorithm described above [19], in a cohort study of women in the United States [20].The HIV Prevention Trials Network 064 study was designed to assess HIV incidence in women with HIV risk characteristics in high-risk areas. The investigators enrolled 2067 HIV-seronegative women, monitored them for up to 12 months, and measured an annual HIV incidence of 0.24% (95% CI, .07%–.62%), based on the seroconversion of 4 women. This rate was similar to that determined from use of the multiassay algorithm [19] on specimens from the 32 women found to be HIV seropositive, with no prior HIV diagnosis, at enrollment (0.25% [95% CI, .03%–.93%]). This similarity is encouraging and provides additional evidence for the accuracy of this algorithm, even in this relatively low-incidence cohort. However, the number of events was small in this study, and further validation of the algorithm is needed. The investigators also estimated HIV incidence by detecting acute HIV infection in women found to be HIV seronegative at enrollment and by assuming a 14-day window period of viremia or antigenemia prior to seroconversion. They found 2 women with acute infection, yielding an annual incidence estimate 10-fold higher than that yielded by the other 2 methods (2.52% [95% CI, .17%–9.33%]). This method was first described in 1995 [21], and although it is conceptually solid, it has been found to yield unreliable incidence estimates because of the small numbers of events found, owing to the narrow window before HIV seroconversion [1, 2].

New approaches to HIV incidence assays are emerging. Recently, CDC scientists have developed the limiting antigen avidity assay [22]. Data from laboratory and field surveys are encouraging [22–24], and the technology has been transferred to at least 1 company for commercial manufacturing and marketing [23]. However, before recommendations can be made for routine use in surveillance surveys, this assay must be rigorously evaluated in a variety of populations and global settings and externally reviewed, and a stable supply of high-quality assay kits must be ensured.

The type of rigorous evaluation of HIV incidence assays conducted by Laeyendecker et al [19] and Eshleman et al [20] is needed to advance the field. On another front, progress is being made in assessments of viral diversity as a marker for duration of HIV infection [25–27]. Advances in genetic characterization methods may make this feasible in the not-too-distant future. Substantial progress has been made in refining our tools for HIV incidence determination, but focused, collaborative efforts will be required to yield the simple, inexpensive assays needed to allow us to monitor the global response to the HIV/AIDS pandemic.

Note

Potential conflicts of interest. Author certifies no potential conflicts of interest.

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References

1. Mastro TD, Kim AA, Hallett T, et al. Estimating HIV incidence in populations using tests for recent infection: issues, challenges and the way forward. J HIV AIDS Surveill Epidemiol **2010**; 2:1–14.
2. Busch MP, Pilcher CD, Mastro TD, et al. Beyond detuning: 10 years of progress and new challenges in the development and application of assays for HIV incidence estimation. AIDS **2010**; 24:2763–71.
3. Brookmeyer R. Measuring the HIV/AIDS epidemic: approaches and challenges. Epidemiol Rev. **2010**; 32:26–37.
4. Hallett TB, Stover J, Mishra V, Ghys PD, Gregson S, Boerma T. Estimates of HIV incidence from household-based prevalence surveys. AIDS **2010**; 24:147–52.
5. Janssen RS, Satten GA, Stramer SL, et al. New testing strategy to detect early HIV-1 infection for use in incidence estimates and for clinical and prevention purposes. JAMA **1998**; 280:42–8.
6. Parekh BS, Kennedy MS, Dobbs T, et al. Quantitative detection of increasing HIV type 1 antibodies after seroconversion: a simple assay for detecting recent HIV infection and estimating incidence. AIDS Res Hum Retroviruses **2002**; 18:295–307.
7. Le Vu S, Pillonel J, Semaille C, et al. Principles and uses of HIV incidence estimation from recent infection testing—a review. Euro Surveill **2008**; Sept 4;13(36).
8. Murphy G, Parry JV. Assays for the detection of recent infections with human immunodeficiency virus type 1. Euro Surveill **2008**; 2008 Sept 4;13(36).
9. Family Health International, Bill and Melinda Gates Foundation. Assays to estimate HIV incidence and detect acute HIV infection: global landscape and market assessments. Research Triangle Park, NC: Family Health International, **2009**.
10. Guy R, Gold J, Calleja JM, et al. Accuracy of serological assays for detection of recent infection with HIV and estimation of population incidence: a systematic review. Lancet Infect Dis **2009**; 9:747–59.
11. Hargrove JW, Humphrey JH, Mutasa K, et al. Improved HIV-1 incidence estimates using the BED capture enzyme immunoassay. AIDS **2008**; 22:511–8.
12. Brookmeyer R. Should biomarker estimates of HIV incidence be adjusted? AIDS **2009**; 23:485–91.
13. Brookmeyer R. On the statistical accuracy of biomarker assays for HIV incidence. J Acquir Immune Defic Syndr **2010**; 54:406–14.
14. Hall HI, Song R, Rhodes P, et al. Estimation of HIV incidence in the United States. JAMA **2008**; 300:520–9.
15. Prejean J, Song R, Hernandez A, et al. Estimated HIV incidence in the United States, 2006–2009. PLoS One **2011**; 6:e17502.
16. Parekh BS, Hanson DL, Hargrove J, et al. Determination of mean recency period for estimation of HIV type 1 incidence with the BED-capture EIA in persons infected with diverse subtypes. AIDS Res Hum Retroviruses **2011**; 27:265–73.
17. World Health Organization (WHO). When and how to use assays for recent infection to estimate HIV incidence at a population level. Geneva: WHO, **2011**.
18. Incidence Assay Critical Path Working Group. More and better information to tackle HIV epidemics: towards improved HIV incidence assays. PLoS Med **2011**; 8: e1001045.
19. Laeyendecker O, Brookmeyer R, Cousins M, et al. HIV incidence determination in the United States: a multi-assay approach. J Infect Dis **2012**. In press.
20. Eshleman SH, Hughes J, Laeyendecker O, et al. Comparison of three methods for assessing HIV incidence in a cohort study of women in the United States: HIV Prevention Trials Network 064 Study. J Infect Dis **2012**. In press.
21. Brookmeyer R, Quinn TC. Estimation of current human immunodeficiency virus incidence rates from a cross-sectional survey using early diagnostic tests. Am J Epidemiol **1995**; 141:166–72.
22. Duong YT, Qiu M, De AK, et al. Detection of recent HIV-1 infection using a new limiting-antigen avidity assay: potential for HIV-1 incidence estimates and avidity maturation studies. PLoS One **2012**; 7: e33328.
23. Parekh B, Duong YT, Mavenger Y, et al., editors. Performance of ne LAg-Avidity EIA to measure HIV-1 incidence in a cross-sectional population: Swaziland HIV Incidence Measurement Survey (SHIMS). Presented at: XIX International AIDS Conference, Washington, DC, 22–27 July 2012. Abstract no. LBPE27.
24. Reed JB, Justman J, Bicego G, et al. editors. Estimating national HIV incidence from directly observed seroconversions in the Swaziland HIV Incidence Measurement Survey (SHIMS) longitudinal cohort. Presented at: XIX International AIDS Conference, Washington, DC, 22–27 July 2012. Abstract no. FRLBX02.
25. Cousins MM, Laeyendecker O, Beauchamp G, et al. Use of a high resolution melting (HRM) assay to compare gag, pol, and env diversity in adults with different stages of HIV infection. PLoS One **2011**; 6: e27211.
26. Cousins MM, Ou SS, Wawer MJ, et al. Comparison of a high-resolution melting assay to next-generation sequencing for analysis of HIV diversity. J Clin Microbiol **2012**; 50:3054–9.
27. Ragonnet-Cronin M, Aris-Brosou S, Joanisse I, et al. Genetic diversity as a marker for timing infection in HIV-infected patients: evaluation of a 6-month window and comparison with BED. J Infect Dis **2012**; 206: 756–64.