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HIV Type 1 Incidence Estimates by Detection of Recent Infection from a Cross-Sectional Sampling of Injection Drug Users in Bangkok: Use of the IgG Capture BED Enzyme Immunoassay

DALE J. HU,[1] SUPHAK VANICHSENI,[2] PHILIP A. MOCK,[3] NANCY L. YOUNG,[3] TRUDY DOBBS,[1] ROBERT H. BYERS, JR.,[1] KACHIT CHOOPANYA,[2] FRITS VAN GRIENSVEN,[1,3] DWIP KITAYAPORN,[3,4] J. STEVEN McDOUGAL,[1] JORDAN W. TAPPERO,[1,3] TIMOTHY D. MASTRO,[1,3] and BHARAT S. PAREKH[1]

ABSTRACT

Development of serologic tests to detect recent HIV-1 infection has generated worldwide interest in applying this approach to estimate incidence. We previously devised an IgG-capture BED-EIA (or BED-CEIA) that detects increasing levels of anti-HIV IgG following seroconversion to identify recent infection and to estimate incidence among persons infected with diverse HIV-1 subtypes worldwide. Injection drug users (IDUs; n = 1969) were screened in 1996 for participation in a prospective cohort study. Serum specimens from 594 IDUs were HIV-1 seropositive (30.2%) and were tested with the BED-CEIA. The proportion of recent infections and estimated incidence by different epidemiological risk factors were compared with incidence data measured from the prospective cohort. Of 594 HIV-1-seropositive specimens, 113 (19%) were identified as recent infections. Overall, the estimated annual incidence among persons screened was 17.3%/year (95% CI, 12.8–24.2%/year) compared with 9.0%/year (95% CI, 6.7–11.9%/year) measured from the prospective cohort during the same time period. Estimated incidence was higher among younger aged and unemployed IDUs as well as among those who injected more frequently, confirming previously reported risk factors from this prospective cohort. As persons screened from a cross-sectional sampling probably have higher risk for HIV than selected uninfected individuals who choose to participate and receive risk reduction counseling in a longitudinal cohort study, use of this or other serologic testing strategies to identify populations with high incidence (such as for HIV vaccine trials) may overestimate incidence measured from prospective cohorts.

INTRODUCTION

ESTIMATION OF INCIDENCE is a key component of monitoring the HIV-1 epidemic in various populations around the world. In addition, identification of persons with early HIV-1 infection has become increasingly important both to focus prevention efforts and to improve opportunities for early HIV therapy and prevention of opportunistic infections.[1] However, identification of recently infected persons (generally within 6 months of infection) and accurate estimation of incidence are difficult and have traditionally relied on the prospective testing and longitudinal follow-up of people at risk.[2–5] Aside from the logistic challenges in conducting prospective studies, estimates of incidence based on cohorts are biased for a number of reasons.[6]

A number of methods have been proposed to estimate HIV incidence data from cross-sectional surveys.[6–11] In 1998, Janssen and colleagues described a simple and practical serologic testing algorithm to identify recent HIV-1 infection and to provide population incidence estimates using cross-sectional

[1]Division of HIV/AIDS Prevention and Division of AIDS, STD, TB Laboratory Research, Centers for Disease Control and Prevention, Atlanta, Georgia 30333.
[2]Bangkok Metropolitan Administration, Bangkok, 10600 Thailand.
[3]Thailand MOPH–U.S. CDC Collaboration, Bangkok, 11000 Thailand.
[4]Mahidol University, Bangkok, 10400 Thailand.

specimens.[11] Validation of this strategy among blood donors and other population groups in North America demonstrated that estimated incidence compared favorably with observed incidence.[11,12] While there was widespread interest in applying the Serologic Testing Algorithm for Recent HIV Seroconversion (STARHS) approach to detect recent infections and to estimate HIV-1 incidence in various populations around the world, subsequent evaluation showed that the initial assay had different performance characteristics for HIV-1 subtypes other than subtype B, the predominant subtype in North America and Western Europe.[13] This finding prompted the development of a new assay, the BED-CEIA (HIV-1 subtypes B, E, and D, IgG-capture enzyme immunoassay), which was shown to have similar sensitivity to multiple HIV-1 subtypes.[14] The objectives of our study were to use the BED-CEIA to detect recent HIV-1 infections among HIV-1-seropositive persons screened from a large cross-sectional sample for enrollment in a prospective cohort and to estimate incidence. We then compared this estimate with measured incidence from previously published analyses of the same prospective cohort study.[5,15]

MATERIALS AND METHODS

To distinguish recent from established HIV-1 infection and to overcome the problems associated with testing different subtypes, we previously devised a simple enzyme immunoassay (BED-CEIA), using a branched synthetic peptide that incorporates immunodominant gp41 sequences form subtypes B and E (also known as circulating recombinant form CRF01_AE) and subtype D.[14] Consensus sequences form subtypes B, E, and D were found to be sufficiently divergent and representative of the major subtypes of HIV-1 prevalent in various areas of the world. This assay, whose performance characteristics have been described previously, detects increasing levels of anti-HIV IgG during early infection and was used to test longitudinal specimens obtained from known incident infections in the United States (subtype B) and in Thailand (subtypes B and E).[14] Our analysis indicated that an optimal normalized optical density cutoff of 1.0 and a seroconversion period of 160 days offered the best combination of sensitivity and specificity for distinguishing between incident and long-term infections. Separate analyses of subtype B and E infections yielded the same optimal threshold and similar seroconversion periods. Subsequent validation of the assay with specimens from Africa (subtypes A, C, and D) demonstrated that this assay was useful for detecting recent HIV-1 infection and for estimating incidence from a range of different HIV-1 subtypes.[14]

The HIV-1 epidemic among injection drug users (IDUs) in Bangkok has been well documented since the rapid rise in HIV-1 prevalence in the late 1980s.[16,17] As part of an HIV vaccine preparatory cohort, among 1969 IDUs screened in 1996, who were not known to be HIV-1 infected,[5,18] serum specimens from 594 IDUs who tested HIV-1 seropositive during the period April 1996 through December 1996 were then tested with the BED-CEIA. The proportion of recent infections and estimated BED incidence stratified by different epidemiological risk factors were compared with incidence data measured from IDUs monitored in the prospective cohort.[5,15] Estimated BED incidence was determined to be equal to $(R)/(R + N)$ times $(365/W)$, where R is the number of recent infections as determined by BED-CEIA, N is the total number of HIV-seronegative subjects, and W is the seroconversion interval (window) of the assay, which was previously determined to be 160 days.[11,14] The 95% confidence intervals (CIs) for estimated BED incidence were calculated using the Bonferroni method.[11]

RESULTS

Of the 1969 persons screened for HIV-1 in 1996, 594 persons were seropositive, yielding an overall prevalence of 30.2%. Of the seropositive specimens, 113 were classified as recent infections by reactivity on the BED-CEIA and overall, estimated BED incidence among persons screened was 17.3%/year (95% CI, 12.8–24.2%/year) compared with 9.0%/year (95% CI, 6.7–11.9%/year) measured from the prospective cohort during the same time period from April 1996 through December 1996.[15]

Analysis of the numbers of recent infections and estimated BED incidence associated with different factors (Table 1) showed that the estimated incidence was higher among younger aged and unemployed IDUs as well as among those who injected more frequently. Although the estimated BED incidence at screening was higher than the incidence measured from the prospective cohort, the relative patterns of incidence within the different strata were consistent with previously reported factors associated with higher observed incidence in the prospective cohort.[5] Characteristics associated with higher incidence in both the cross-sectional screening sample and the prospective cohort included being younger, unmarried, unemployed, and reporting higher frequencies of drug injection and incarceration.[5] In addition, the estimated BED incidence among persons who were screened in the cross-sectional sample increased from the second quarter through the fourth quarter of 1996, which was consistent with the period of peak incidence observed in late 1996 among persons in the prospective cohort.[15]

DISCUSSION

Our study is the first application of the BED-CEIA to a cross-sectional sample for estimating incidence and compares the estimated BED incidence with incidence measured from the subset of persons who consented to subsequent follow-up in a prospective cohort. It is important to note that our study is not a validation of the BED-CEIA itself, but rather an illustration of how incidence estimates can differ between two methodologies applied to the same target population.

Although the estimated BED incidences appeared to be high, incidence of this magnitude is certainly plausible in this high-risk population. For example, in the early phases of the IDU epidemic in Bangkok in the late 1980s, annual HIV-1 incidence was estimated to range from 20% to almost 60% and still remained above 10% during the 1990s.[17] In addition, the high estimated BED incidence in late 1996 was consistent with a documented period of high incidence when temporal trends in incidence observed from the prospective cohort showed a peak of 16.5/100 person-years (95% CI, 11.7–22.5/100 person-years) during the fourth quarter of 1996.[15]

TABLE 1. FACTORS ASSOCIATED WITH RECENT INFECTION AND ESTIMATED INCIDENCE AMONG INJECTION DRUG USERS IN BANGKOK, THAILAND

		HIV-1 seropositive[a]		*Recent infections*[b]			*Estimated annual incidence*	
Factor	*Total screened*	*Total HIV+*	*HIV prev.*	*# recent*	*% recent*	p *value*	*Point estimate*	*95% C*
Sex						0.368		
Male	1815	560	30.9%	109	19.5%		18.2%	(13.4–25.4)
Female	154	34	22.1%	4	11.8%		7.4%	(1.5–23.4)
Age group						0.001		
16–24	591	126	21.3%	48	38.1%		21.3%	(13.8–33.1)
25–34	753	258	34.3%	36	14.0%		15.5%	(9.5–25.1)
35+	623	210	33.7%	29	13.8%		15.0%	(8.7–25.4)
Marital status						0.458		
Married	699	180	25.8%	29	16.1%		12.1%	(7.0–20.4)
Not married	1270	414	32.6%	84	20.3%		20.4%	(14.5–29.3)
Education						0.211		
Primary or less	830	299	36.0%	51	17.1%		20.0%	(13.1–30.7)
Some secondary or more	1138	294	25.8%	62	21.1%		15.6%	(10.6–23.3)
Employment						0.038		
Employed	1372	406	29.6%	68	16.7%		15.0%	(10.3–22.1)
Unemployed	597	188	31.5%	45	23.9%		22.6%	(14.5–35.4)
Methadone						0.32		
45 day detox	1733	535	30.9%	104	19.4%		18.2%	(13.3–25.5)
Maintenance	193	51	26.4%	7	13.7%		10.7%	(3.4–27.0)
Injection (last 6 mo)						0.072		
Daily	1708	529	31.0%	106	20.0%		18.8%	(13.8–26.3)
Less than daily	258	65	25.2%	7	10.8%		8.0%	(2.6–20.1)
Incarceration						0.269		
No	1622	452	27.9%	88	19.5%		16.0%	(11.4–22.8)
Yes, no IDU	125	60	48.0%	7	11.7%		22.2%	(7.1–55.8)
Yes, IDU	218	82	37.6%	18	22.0%		26.7%	(13.4–50.2)
Time period						0.001		
2nd quarter	928	245	26.4%	36	14.7%		11.4%	(6.9–18.6)
3rd quarter	596	198	33.2%	31	15.7%		16.5%	(9.7–27.6)
4th quarter	440	151	34.3%	46	30.5%		31.3%	(20.1–48.9)
Total overall	1969	594	30.2%	113	19.0%		17.3%	(12.8–24.2)

[a]Serum specimens that tested HIV-1 seropositive during the period April–December 1996.
[b]Classified as recent infections by reactivity on the IgG Capture BED enzyme immunoassay.

There are a number of possible reasons why the estimated BED incidence may be higher than the incidence measured from the prospective cohort during the same time period. We have previously discussed some potential factors and biases affecting this and other STARHS assays.[13,14,19] Aside from assay-related factors that may bias the estimated BED incidence upward, the most important consideration is that the subset of individuals who consent to be monitored in a prospective cohort may differ on a number of important epidemiological and sociobehavioral risk factors from the overall population screened. As reported previously, participants enrolled into our prospective cohort tended to have characteristics associated with lower risk as compared with the HIV-seronegative persons who were screened but not enrolled. For example, in comparison with those not enrolled, the participants who were enrolled were older, more likely to be married, more likely to be in a methadone maintenance program, and less likely to inject more than once per day.[5] Furthermore, those persons who tested positive on standard EIA at screening, and whose blood specimens were subsequently tested with the BED-CEIA, tended to be at higher risk than participants who were initially seronegative and were willing to return for regular follow-up. Nevertheless, stratified comparisons of estimated BED and cohort incidence are consistent with previous research on incidence and transmission risk in this population.

Thus, while application of this assay may be useful for cross-sectional estimates of incidence in various populations and risk groups, it does not mitigate the need for careful consideration of persons tested (including sampling frequency) and for examination of potential biases. In general, it will be critically important to evaluate the performance characteristics and the predictive value of this and other assays in different populations and how these may affect estimates of incidence. Clearly, use of the BED-CEIA or other STARHS approaches for identification of recently infected persons and estimating incidence will be extremely useful in high-risk and/or resource-poor populations, where prospective follow-up of persons may be difficult. However, use of these testing strategies to identify populations with high incidence (such as for HIV vaccine trials) may overestimate incidence measured from prospective cohort studies.

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Address reprint requests to:
Dale J. Hu
Division of HIV/AIDS Prevention
Division of AIDS/STD/TB Laboratory Research
Centers for Disease Control and Prevention
Atlanta, Georgia 30333

E-mail: dhu@cdc.gov