Methods to Assess Population Effectiveness of Therapies in Human Immunodeficiency Virus Incident and Prevalent Cohorts

Two methods are presented for measuring population effectiveness (i.e., reduction of disease in a population in which only some receive treatment) of antiretroviral therapy among human immunodeficiency virus (HIV)-infected men at risk for acquired immunodeficiency syndrome (AIDS) and followed between January 1, 1986, and June 30, 1999, in the Multicenter AIDS Cohort Study. Method I, requiring use of a seroincident cohort, estimates relative hazards of AIDS for persons at equal duration of infection. Method II, allowing use of a seroprevalent cohort, estimates relative hazards since the beginning of therapy eras for persons starting at equal levels of prognostic markers of disease stage (CD4 cell count and HIV type 1 RNA). The follow-up interval was divided into four calendar periods to characterize different eras of antiretroviral therapy. For method I, the relative hazards were 1.52 (95% confidence interval (CI): 0.93, 2.49), 0.91 (95% CI: 0.66, 1.26), and 0.30 (95% CI: 0.18, 0.51) for the eras of no therapy, dual nucleoside therapy, and potent combination antiretroviral therapy, respectively (monotherapy was the reference era). For method II, the corresponding relative hazards were 1.52 (95% CI: 1.10, 2.09), 1.03 (95% CI: 0.77, 1.38), and 0.31 (95% CI: 0.21, 0.45). These results extend the measurement of population effectiveness from incident to prevalent cohorts and demonstrate the ability of cohort studies to complement information provided by clinical trials. Am J Epidemiol 2001;154:675–81.

Assessment of the effect of treatment based on data from clinical trials versus observational studies has long been a topic of debate in epidemiologic research (1). To date, the consensus opinion has been that clinical trials are the “gold standard” for the measurement of efficacy. Recently, however, there have been a number of reports using observational studies to assess the efficacy of therapies as they are used in the general population, as opposed to the controlled study populations enrolled in clinical trials (2, 3).

Observational studies provide estimates of two types of treatment effects: “individual effectiveness” and “population effectiveness” (4). Individual effectiveness compares responses between groups of persons receiving treatment and groups not receiving treatment. In observational studies, analyses via stratification and/or regression overcome the lack of randomization (5). These adjusted analyses provide alternative, or supplemental, estimates of efficacy as measured by clinical trials. In contrast, population effectiveness compares the incidence of disease in populations in which some and typically the most ill are given treatment with the incidence of disease in a population in which no one, or almost no one, receives treatment. Measures of population effectiveness differ from measures of individual effectiveness by providing an estimate of the reduction in disease burden in a population after treatment has been introduced into the population at large (4).

At the population level, measures of effectiveness are susceptible to ecologic fallacy, because exposures other than treatments can differ between the populations being compared or even among the time periods when different treatments were used in the same population (6, 7). Cohort studies, with the advantage of prospective and direct follow-up of subjects, standardized protocols for the collection of data and specimens, and closely monitored outcomes are able to observe and measure changes in important variables such as health care access and utilization, use of prophylaxis, and adherence to treatments. Therefore, to provide a contextual framework for the inferences in population level studies, it is important to fully document descriptors of the cohort in different treatment eras to rule out, to the extent the data allow, the possibility of ecologic fallacy. The primary objective of the analysis is to ascribe changes in incidence to changes in the therapies defining different eras. As in all observational studies, there...
is the possibility of unmeasured changes between periods, but this is less likely to occur when contiguous and relatively short subperiods (e.g., 2–3 years in length) are compared.

The efficacy (8–10) and effectiveness of antiretroviral therapy for human immunodeficiency virus (HIV) type 1-infected persons have been well demonstrated at both the individual (11–13) and population (14–19) levels. The primary aim of studies that assess population effectiveness is to compare the incidence of acquired immunodeficiency syndrome (AIDS) across calendar periods when cohorts are exposed to different antiretroviral therapies. The later periods tend to have persons observed at later stages of disease (e.g., lower CD4 cell counts) and have higher percentages of persons who have remained AIDS free for long durations. These factors operate in opposite directions: persons at later stages of disease will inevitably be subject to a higher hazard of AIDS than persons at an earlier stage of infection, while persons who have been free of AIDS for long durations (and hence are indicative of some level of immunity) will have a lower hazard of AIDS. Therefore, analyses of population effectiveness of HIV treatment should be adjusted for duration of infection and/or stage of disease to control for survival bias.

The objective of the study reported here was to compare two methods of adjustment for disease stage and risk of disease progression. In method I, the incidences of AIDS in persons reaching the same duration of infection at different calendar periods of antiretroviral therapy were contrasted. In method II, the incidences of AIDS in persons having similar levels of prognostic markers (CD4+ lymphocyte count and plasma HIV RNA level) at the start of different therapy eras were contrasted.

MATERIALS AND METHODS

Because the types of antiretroviral therapy that have been prescribed are closely tied to calendar dates, analyses of population effectiveness can use calendar periods to characterize the eras of different antiretroviral therapies. For this analysis, four treatment eras were defined: no therapy, nucleoside monotherapy, dual nucleoside therapy, and potent combination antiretroviral therapy. The calendar periods used for these eras were as follows: January 1986 through June 1989 as the era of no antiretroviral therapy; July 1989 through December 1992 as the era of nucleoside monotherapy; January 1993 through June 1996 as the era of dual nucleoside therapy; and July 1996 through June 1999 as the era of potent combination antiretroviral therapy. These calendar periods were used as the primary exposure variables in time-to-event (e.g., AIDS) models (5, 15, 20, 21). The methods and calendar periods used in this analysis are similar to those reported in recent studies of population effectiveness of antiretroviral therapy (15, 21–24).

Method I: adjustment by duration of infection

Study population. Adjustment for disease progression by duration of infection requires knowledge of each person’s date of seroconversion; that is, the study population must consist of seronegative persons who subsequently seroconverted during study follow-up (i.e., an incident cohort). The Multicenter AIDS Cohort Study enrolled 5,622 gay men, of whom 3,427 were seronegative at entry. Of the seronegatives, 546 seroconverted during the study follow-up (1984–1999), and they constitute the appropriate study population for method I. The primary aim of this analysis was to compare the incidence of AIDS in persons who reached the same duration of infection in different calendar periods representing previously defined eras of antiretroviral therapy. Hence, differences in the incidence of AIDS in different calendar periods cannot be ascribed to differences in duration of infection but instead to other variables that are different among the various periods.

Exposure and outcome. Exposure to therapy as measured by the calendar periods that characterize population level exposure to different therapies was treated as an external time-dependent covariate (i.e., an instrumental variable) (25). The primary outcome was the hazard of AIDS in different calendar periods. The time scale used in method I was the time to AIDS from the date of seroconversion (i.e., duration of infection). As previously reported (15), we defined seroconversion as occurring at one third of the time interval between the date of last known negative status and the date of first known HIV-positive status, to reflect declining incidence more accurately than using the midpoint. That is, if \( a \) and \( b \) represent the dates of the latest and first positive HIV tests, the date of seroconversion was defined as \( e' = (2a + b)/3 \). After \( e' \), each person was seen for \( t' \) years either because of AIDS onset (\( \delta = 1 \)) or because the person was last to follow-up while AIDS free or reached the date of analysis (June 1999) as AIDS free (\( \delta = 0 \)).

Analysis. The statistical analysis for the external time-dependent covariate was accomplished by the use of survival analysis (i.e., proportional hazards model) with staggered entries. The proportional hazards model allows the comparison of incidences of AIDS among persons at a similar duration of HIV infection. Specifically, for each calendar period beginning on date \( c_1 \) and ending on date \( c_2 \) (e.g., for the first calendar period \( c_1 = January 1, 1986 \), and \( c_2 = June 30, 1989 \)), a person contributes information to that period if \( t' > c_1 \) (i.e., person neither developed AIDS nor was lost to follow-up before \( c_1 \) ) and if \( e' < c_2 \) (i.e., seroconverted before \( c_2 \)). A person is included in the analysis from \( e = \max(2(\delta - a)/3, c_1 - e') \) to \( t = \min(\delta, c_2 - e', t') \), and the censoring information for the calendar period is given by \( \delta = \min(\delta, \delta') \), indicator of whether \( t = t' \).

In summary, the contribution of a person for a given period (from \( c_1 \) to \( c_2 \)) was characterized by three variables: 1) \( e \), the larger of two thirds of the seroconversion lag and the duration of infection at the beginning of an era; 2) \( t \), the duration of infection at exit in the given period; and 3) \( \delta \), the AIDS status at exit in that period, whereby \( \delta = 1 \) for only the persons who developed AIDS between \( c_1 \) and \( c_2 \). With the outcome defined by the triplet \((e, t, \delta)\), we used a proportional hazards regression model with periods as a categorical variable. Antilogs of regression coefficient represent the relative hazards of periods relative to the period chosen as the reference category. Software for proportional hazards
regression for survival data with staggered entries \((e)\) is widely available \((26)\).

**Method II: adjustment by markers of disease progression**

**Study population.** The primary objective of this method was to compare the incidence of AIDS in persons with the same key prognostic markers (CD4 cell count and plasma HIV RNA) of HIV progression at the beginning of different therapeutic eras. All HIV-seropositive participants with available data on CD4 cell count and HIV RNA were eligible for this study. Thus, this study design could include both seroincident and seroprevalent cohorts. In principle, all HIV-infected persons in the study population could be used, but HIV RNA data were not measured prospectively in the Multicenter AIDS Cohort Study. To optimize testing and efficiently represent the seropositive population, a random sample of 300 participants was taken from the beginning of each of the four calendar periods (i.e., the 6-month interval before the start of an era) from the set of all seropositive AIDS-free individuals with available specimens for testing. As a result, the study population comprises four groups of 300 persons, each selected at random among all persons seen AIDS free from July 1985 through December 1985, from January 1989 through June 1989, from July 1992 through December 1992, and from January 1996 through June 1996, respectively. HIV RNA was quantified on the 1,200 plasma samples in a central laboratory with the use of reverse transcriptase polymerase chain reaction \((27)\).

**Exposure and outcome.** Exposure to therapy, as measured by calendar periods that characterize population level exposure to different therapy regimens, was treated as a fixed covariate via indicator variables. The markers of disease progression measured at the beginning of each period were included in the model as covariates. The primary outcome was the time to AIDS or censoring from the beginning of each era for each of the four groups of 300 persons. The hazard of AIDS in different periods was compared using persons with similar values of CD4 cell count and HIV RNA measured at the beginning of each era.

**Analysis.** The statistical analysis for method II is simpler than the analysis for method I. While both utilize proportional hazards models, method II does not include the complexity associated with the staggered entry model used for adjustment by duration of infection. For method II, the contribution of a person for a given period is characterized by only two variables: 1) the duration of follow-up at exit in the given period and 2) the AIDS status at exit within the period. As in method I, only those persons who developed AIDS in a given period were treated as uncensored for that period. The adjustment by markers as covariates allows appropriate comparisons between persons with equal CD4 cell counts and HIV RNA levels at the beginning of each period (i.e., persons at similar disease stages and risk of disease progression). A person could have contributed AIDS-free time to more than one calendar period if he was chosen via random selection for more than one therapy era. Robust standard errors were used to account for possible statistical dependencies of persons who contributed multiple times.

**RESULTS**

Figure 1 illustrates the percentages of the study populations who, while AIDS free, were taking potent combination

![Figure 1](https://academic.oup.com/aje/article-abstract/154/7/675/107524) Use of antiretroviral therapy among the two study populations: method I (seroconverters, \(n = 546\)) and method II (random samples of seropositive participants, \(n = 300\)) across four calendar periods, Multicenter AIDS Cohort Study, 1984–1999. AIDS, acquired immunodeficiency syndrome.
antiretroviral therapy, dual nucleoside therapy, nucleoside monotherapy, or no antiretroviral therapy. For all visits that a person attended in a given period, the most intense reported antiretroviral therapy was taken; for example, if a person was initially on dual antiretroviral therapy but began potent combination antiretroviral therapy, exposure was taken as potent combination antiretroviral therapy. For the four calendar periods, the predominant forms of antiretroviral therapy were none, nucleoside monotherapy, dual nucleoside therapy, and potent combination antiretroviral therapy, respectively. In addition, the therapies received by the two study populations in each era were quite similar, except for the first era, where more than 25 percent of the seroprevalent samples used monotherapy versus only about 10 percent of seroconverters. However, because it has been established that monotherapy (especially when such a small percentage of the population is affected) did not significantly affect the incidence of AIDS, this difference probably did not impact the results of this study.

Table 1 shows descriptive statistics of the 546 seroconverters who comprised the study population for method I. It clearly demonstrates the dramatic increase in the duration of infection across calendar periods, indicating the need for adjustment. Moreover, in the last calendar period there were a marked decline in the number of person-years at risk of AIDS and, consequently, a decline of observed AIDS cases. The increase of disease severity among HIV-infected men over time was observed as a decreasing median CD4 cell count with respect to the calendar periods. However, in the last period the median HIV RNA level showed a marked decrease, which was probably the result of early use of potent combination antiretroviral therapy before July 1996, as seen in figure 1. The percentage of seroconverters in each sample increased across the calendar periods, representing the increase of the proportion of incident cases of HIV infection among all seropositive persons in the Multicenter AIDS

Table 2 presents the relative hazards of AIDS for the analysis of the study population for method I (seroconverters), both unadjusted and adjusted by duration of infection. The unadjusted analysis estimated the relative hazard of AIDS during the era of nucleoside monotherapy as being higher than the hazard during the era of no therapy. This results from the fact that persons in the second calendar period had experienced a longer duration of infection than those in the first calendar period. Adjusting for duration of infection allowed for the comparison of persons with equal duration of infection and thus shows the appropriate directionality of the effect of no therapy on the population; that is, the hazard of AIDS was 52 percent higher in the era of no antiretroviral therapy compared with the era of monotherapy. The adjusted model identified the directionality of the effect of dual nucleoside therapy as reported by clinical trials. The unadjusted model also slightly underestimated the effect of potent combination antiretroviral therapy on the population; that is, the relative hazard (unadjusted) = 0.37 versus the relative hazard (adjusted) = 0.30.

Table 3 shows descriptive statistics for the four groups of persons randomly selected from visits occurring at the start of different therapy eras for method II. The incidences of AIDS in this seropositive study population were similar to the incidences observed for the seroincident study population. The increase of disease severity among HIV-infected men over time was observed as a decreasing median CD4 cell count with respect to the calendar periods. However, in the last period the median HIV RNA level showed a marked decrease, which was probably the result of early use of potent combination antiretroviral therapy before July 1996, as seen in figure 1. The percentage of seroconverters in each sample increased across the calendar periods, representing the increase of the proportion of incident cases of HIV infection among all seropositive persons in the Multicenter AIDS

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**TABLE 1. Descriptive statistics of the 546 seroconverters (study population for method I) by calendar periods corresponding to four different eras of therapy, Multicenter AIDS* Cohort Study, 1984–1999**

<table>
<thead>
<tr>
<th>Calendar period</th>
<th>No. seen while AIDS free</th>
<th>No. of AIDS cases in period</th>
<th>AIDS-free person-years in period</th>
<th>Median date of seroconversion†</th>
<th>Median duration of infection at beginning of period (years)</th>
</tr>
</thead>
<tbody>
<tr>
<td>January 1986–June 1989</td>
<td>341</td>
<td>36</td>
<td>887</td>
<td>July 1985</td>
<td>0.68</td>
</tr>
<tr>
<td>January 1993–June 1996</td>
<td>378</td>
<td>100</td>
<td>1,017</td>
<td>January 1988</td>
<td>5.05</td>
</tr>
</tbody>
</table>

* AIDS, acquired immunodeficiency syndrome.
† For all men seen while AIDS free in each period.

**TABLE 2. Results of method I: unadjusted and adjusted relative hazards of AIDS* by therapy era for the 546 seroconverters, Multicenter AIDS Cohort Study, 1984–1999**

<table>
<thead>
<tr>
<th>Calendar period</th>
<th>Era of therapy</th>
<th>Unadjusted</th>
<th>Adjusted by duration of infection</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>Relative hazard</td>
<td>95% CI*</td>
</tr>
<tr>
<td>January 1986–June 1989</td>
<td>No therapy</td>
<td>0.49</td>
<td>0.34, 0.73</td>
</tr>
<tr>
<td>July 1989–December 1992</td>
<td>Monotherapy</td>
<td>1</td>
<td>1</td>
</tr>
<tr>
<td>January 1993–June 1996</td>
<td>Dual therapy</td>
<td>1.19</td>
<td>0.89, 1.58</td>
</tr>
<tr>
<td>July 1996–June 1999</td>
<td>Potent therapy</td>
<td>0.37</td>
<td>0.23, 0.60</td>
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</tbody>
</table>

* AIDS, acquired immunodeficiency syndrome; CI, confidence interval.
Cohort Study; that is, a higher incidence of infection occurred earlier in the study among seroconverters.

Table 4 presents the effect of adjustments by different markers for disease progression for study population II. The unadjusted analysis, as was previously noted, inappropriately suggests the hazard of AIDS as being lower in the no therapy period than in the monotherapy period. The fully adjusted analysis, including both HIV RNA and CD4 cell count, yielded almost identical measures of population effectiveness as those in the model adjusting for the duration of infection (method I). It is observed that adjusting for CD4 cell count alone provides closer estimates to those obtained by adjusting for both CD4 cell count and HIV RNA than does adjusting by HIV RNA alone. Even though adjusting for CD4 cell count does closely estimate the effectiveness of therapies observed in the era of potent antiretroviral therapy, the magnitude of the estimate of the relative hazard of AIDS for the no therapy era was lessened.

DISCUSSION

Because analyses of population effectiveness of antiretroviral therapies for HIV-infected persons require adjustment for stage and risk of disease progression (4), the aim of the current study was to compare two methods of analyses with adjustment by duration of infection requiring cohorts of seroconverters (method I) and prognostic markers of disease risk allowing the inclusion of prevalent cohorts (method II). The two methods produce almost identical estimates of the relative hazards of measuring population effectiveness. Because of the overwhelming effect of potent combination antiretroviral therapy regimens, failure to adjust for disease stage did not necessarily lead to false inferences, but it did underestimate the effect of potent combination antiretroviral therapy compared with the adjusted estimates (unadjusted and adjusted relative hazards = 0.37 and 0.30 for method I and 0.44 and 0.31 for method II, respectively). Such an underestimation suggests that persons in the latest period were, on the average, faced with an intrinsically higher risk of AIDS because of longer durations of infection and/or a higher degree of immune deficiency and higher HIV RNA. Furthermore, the lack of adjustment does lead to incorrect inferences regarding the era of no therapy. The apparent protective effect (relative hazard < 1) in the era of no therapy for the unadjusted analysis in method I was an unanticipated finding, and it was a consequence of the fact that persons in that period had been infected for a relatively short period of time, and the corresponding hazard of AIDS was low (28).

The similarities of the findings from the application of methods I and II are reassuring. They suggest that both methods control for the various stages of disease in populations in different eras. These findings are in consonance with a number of natural history studies reporting that, for the prediction of the onset of AIDS, knowledge of duration of infection does not add significant information beyond that provided by CD4 cell count levels (29–32). Even if some residual effect of duration of infection is acknowledged (32), the impact on our inferences is further reduced because we also controlled by HIV RNA. The similarity of our findings from methods I and II is an accolade of mark-

<table>
<thead>
<tr>
<th>TABLE 3. Descriptive statistics of the four random samples of HIV*-positive men (study population for method II) by calendar periods corresponding to four different eras of therapy, Multicenter AIDS* Cohort Study, 1984–1999</th>
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<tbody>
<tr>
<td>Calendar period</td>
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<tr>
<td></td>
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<tr>
<td>January 1986–June 1989</td>
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<td>July 1996–June 1999</td>
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</table>

* HIV, human immunodeficiency virus; AIDS, acquired immunodeficiency syndrome.
† One person was removed because of missing CD4 cell count.

<table>
<thead>
<tr>
<th>TABLE 4. Results of method II: unadjusted and adjusted relative hazards of AIDS* by therapy era for the four random samples among HIV*-positive men at beginning of era, Multicenter AIDS Cohort Study, 1984–1999</th>
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<tbody>
<tr>
<td>Calendar period</td>
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<tr>
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* AIDS, acquired immunodeficiency syndrome; HIV, human immunodeficiency virus; CI, confidence interval.
ers of HIV disease progression and of the power of the cohort design to compare methods. Markers for other diseases may not yield the same consistency that we have documented for HIV/AIDS.

In population-level observational studies, the danger that ecologic fallacies may influence interpretation of the relative hazards of primary exposure (e.g., calendar periods that characterize treatment eras) cannot be overemphasized. Observational studies that lack comprehensive longitudinal data may be unable to eliminate ecologic fallacy and, thus, may incorrectly ascribe the impact of changes in other exposures to the therapy itself. Therefore, it is essential to document changes in other exposures, such as prophylaxis for opportunistic infections and access and utilization of health care, during the follow-up period. Indeed, in a previous report from the Multicenter AIDS Cohort Study (15), we have documented that the primordial change in the last period of the analysis was the introduction of potent antiretroviral therapy. Furthermore, we have also previously reported a decline in the proportion of persons using prophylaxis against opportunistic infections in the same period when potent antiretroviral therapy was introduced (21). In addition, to assess the effect of possible changes in the pathogenicity of HIV type 1, we extended the analysis in method I to include the year of seroconversion as a categorical variable with 1987 (median) and 1990 (third quartile) as cutpoints. In spite of the analysis including the redundancies of age, period, and cohort effects, the inferences of periods were practically unchanged. This gives stronger grounds to the inference that changes in the incidence of AIDS are very likely to be due to the introduction of potent antiretroviral therapy.

Several HIV cohort studies involving persons with known dates of infection have been used to assess the effectiveness of interventions (e.g., antiretroviral therapies) at the population level (15, 18, 22, 23). As valuable as these contributions are, an obvious limitation arises from the fact that the more commonly available cohorts of prevalent persons cannot be used to directly assess population effectiveness. The focus of this report was to provide alternative methods based on adjustment by markers at the beginning of different eras so that population effectiveness can be assessed using cohorts of prevalent persons. Furthermore, using data from a cohort containing both subcohorts of seroconverters and seroprevalent persons, we compared the epidemiologic inferences reached using the two approaches. Illustrating the equality of adjustment by prognostic markers to that of adjustment by duration of infection opens a pathway for population effectiveness to be measured in seroprevalent study populations.

ACKNOWLEDGMENTS


The authors are grateful to David George for editorial assistance.

REFERENCES


