Estimating HIV incidence, time to diagnosis, and the undiagnosed HIV epidemic using routine surveillance data

**eAppendix**

*Model structure*

The full structure of our model is shown in eFigure 1. The model is a deterministic compartmental model that describes HIV progression in the absence of antiretroviral treatment as a unidirectional flow through different stages of the infection. The HIV incidence over calendar time is given by , which is unknown and needs to be estimated by fitting the model to observed data. Immediately after infection, all individuals first enter a phase of primary infection, . After primary infection, individuals enter at a rate one of five compartments of undiagnosed HIV infection with . The first four stages to correspond to CD4 strata ≥500, 350-499, 200-349, and <200 cells/mm3, respectively, in the absence of an AIDS-defining illness, whilst corresponds to AIDS irrespective of CD4 count.

During each stage of undiagnosed HIV infection, patients can be diagnosed at a rate , which depends on the stage of infection and on calendar time. Upon diagnosis, patients enter a stage of diagnosed HIV infection. In the absence of antiretroviral treatment, both undiagnosed and diagnosed patients experience the same rate of progression to the next CD4 stratum. Progression in diagnosed patients will be different after they start treatment. Therefore, the model only describes HIV progression in undiagnosed patients or in diagnosed patients before the introduction of combination antiretroviral treatment in 1996.

*Model equations*

The model is formulated as a set of ordinary differential equations that describe the changes over time *t* in the number of individuals in each compartment in eFigure 1. The equations are solved numerically using a fourth order Runge-Kutta algorithm. Parameter values are given in eTable 1.

The change in the total number of individuals diagnosed in each stage, , is given by

Before 1996 when combination antiretroviral treatment became available, the cumulative number of individuals diagnosed with AIDS, , consists of those with a concurrent HIV and AIDS diagnosis (stage to ) and diagnosed individuals who progress to AIDS (stage to ). Therefore,

The number of infected men who die without being diagnosed, , is given by

Taking into account missing CD4 measurements and the probability that diagnosed individuals survive up to 1996, the observed number of HIV diagnoses in each CD4 stratum is written as

where is the probability of having a CD4 count measurement available when diagnosed in CD4 stratum in calendar year and is the probability that an untreated individual diagnosed in CD4 stratum in calendar year will survive up to 1996.

*Equation for time from infection to diagnosis*

The average time from infection to diagnosis, , if diagnosis rates remain the same as at the time of infection , can be calculated as

This equation is the sum of the average duration of all possible pathways in eFigure 1 from infection to each of the five stages of diagnosed HIV infection weighted by the probability of taking each pathway. The first term at the right hand side of the equation is the average duration of primary infection. In the second term, is the probability of being diagnosed whilst in stage and thus going to , is the probability of remaining undiagnosed and progressing from to , and is the average duration patients stay in stage .

The actual time from infection to diagnosis, , i.e., the duration patients have been infected by the time they are diagnosed in year , is approximated by

where is the total number of HIV diagnoses in year , and is the number of HIV diagnoses in stage and year among individuals infected in year .

*HIV incidence curve*

The HIV incidence curve is approximated using cubic M-splines, which are piecewise polynomials of degree 4 defined by a knot sequence , where is the number of internal knots. The knot sequence is defined such that , , and where is 1 January 1980 and is 31 December 2012. M-splines are defined such that spline is positive in the interval and zero elsewhere and has the normalisation . Adjacent splines are required to join at the boundaries of the intervals with equal first-order derivatives and continuous second-order derivatives. Formulae for M-spline are taken from Ramsay [1]. The incidence curve is approximated by a linear combination of the M-splines: with parameters that need to be estimated by fitting to data [2]. We take the internal knots to be equidistant between and , whilst is chosen such that fewer knots would give a model with a worse fit to the data.

*Diagnosis because of HIV-related symptoms*

In a secondary analysis, we assume that diagnosis rates are the same for the first three CD4 strata () and that with the rate of being diagnosed because of HIV-related symptoms. The rate of developing HIV-related symptoms is approximately double the rate of AIDS, i.e. [3-5]. However, not all HIV-related symptoms are severe enough to lead to testing for HIV [6,7]. We therefore assume that 50% of HIV infections with symptoms are missed, such that 0.4 per year, which is approximately 50% of the rate of HIV-related symptoms (eTable 1).

*Fitting procedure*

As described in the main text, the model needs to estimate 16 parameters relating to the probability of HIV diagnosis and parameters associated with the incidence curve, which is modelled as a superposition of cubic M-splines, where is the number of internal knots. Maximum likelihood methods are used to find the set of parameters that best fit the observed data. To define the likelihood, we assume that all data items are distributed according to a negative binomial distribution around a mean defined by the model and a dispersion parameter which is initially set at a value of 1000. For convenience, instead of maximising the likelihood, we minimise the equivalent deviance measure. A downhill simplex optimisation algorithm is used to find the minimum value. The algorithm is started from various starting values to ensure that the optimisation is robust and that local optima are avoided.

In the first step of the fitting procedure, all parameters are estimated except for which is fixed to 0 in order to ensure that the incidence curve starts at zero. To improve the robustness and convergence of the fitting procedure, any whose estimated value is either less than 1 or less than a fraction (chosen to be 5%) of the estimated value for is fixed at zero. All parameters are then re-estimated and this procedure is repeated until no more fulfil these criteria. Note that the coefficient associated with the -th spline is allowed to be smaller than 1, because fixing it at zero would force the incidence to be zero.

In the second step, a new approximate value for the dispersion parameter is obtained by requiring that Pearson’s statistic equals with the number of data points used in the fit and the number of estimated parameters [8]. Using this updated dispersion parameter the set of parameters is re-estimated. This procedure is repeated until the dispersion parameter does not notably change anymore, which is the case after four times.

*Confidence intervals*

We estimated pointwise 95% confidence intervals for parameters and other derived quantities via a bootstrap procedure [9]. In brief, assuming that the data are distributed according to a negative binomial with a mean defined by the model, we generated a new dataset by sampling from this distribution for every year for each of the relevant data items. The model was then refitted to this new dataset starting from the parameter values found in the main fit. This sampling and refitting procedure was repeated 200 times. From these 200 fits, 95% confidence intervals around the main model fit were then determined as the 2.5th and 97.5th percentile.

*Simulated data*

Our approach was tested on three data sets of hypothetical patients generated by the HIV Synthesis progression model [10,11]. These hypothetical patients represented HIV epidemics for different risk groups in a typical European country generated with different pairs of incidence and diagnosis rate curves. For each diagnosed patient, the CD4 count at diagnosis was known, as well as the date of AIDS diagnosis, death, and date of emigration or loss to follow-up. Our model was tested by comparing the reconstructed HIV incidence curve with the true annual number of HIV infections that was used as an input in the simulation. Hypothetical patients who migrated before being diagnosed were not included in the true number of infections. When information on CD4 counts at the time of diagnosis was not included in the model, the estimated HIV infection curve looked very similar (eFigure 2 and 3).

*Model fits*

eFigures 4A and 4B show the curves that best fitted to the observed data on AIDS cases and HIV/AIDS diagnoses, as well as on annual total number of HIV diagnoses, reflecting the steep increase in annual HIV infections and shorter time to diagnosis. eFigure 5 shows the best-fitting curves to the number of new HIV diagnoses by CD4 stratum. Before 1996, the proportion of patients with a CD4 count was 34%; this increased to over 85% in recent years. The proportion of MSM with a measured CD4 count ≥500 cells/mm3 at the time of HIV diagnosis increased from 17% in 1996 to 38% in 2012.

eFigure 6 shows the estimated model outcomes when diagnosis rates in the period 1984-1995 were assumed to be a linear function of calendar time instead of being constant. The estimated annual number of HIV infections (eFigure 6A) was comparable to the main analysis, as was the cumulative number of 15,300 (95% CI, 14,800-16,000) infections by the end of 2011. As expected, the estimated average time from infection to diagnosis by year of infection was different for the period 1984-1995. eFigure 7 shows the estimated diagnosis rates by CD4 count interval. From 1996 onwards, diagnosis rates were very similar between the two models, with the steep increase reflecting adoption of a more active HIV testing strategy after the availability of combination antiretroviral therapy [12].

The HIV infection curve looked very similar although with wider confidence intervals in more recent calendar years when no information on CD4 counts at the time of diagnosis was used (eFigure 8A). The cumulative number of infections by the end of 2011 was 15,500 (95% CI, 14,700-16,200), which was comparable to the model with information on CD4 counts. The estimated time to diagnosis was also similar, 2.8 (2.4-3.4) years in 2011 (eFigure 8B).

*Multivariable sensitivity analysis*

We did a multivariable sensitivity analysis to investigate the impact of assumptions on input parameters on the model outcomes. For each of the input parameters, a range of plausible values was identified (eTable 1). Each parameter was partitioned into 250 equidistant possible values spanning its whole plausible range. The sensitivity of the model to the fixed input parameters was then evaluated by sampling from all possible parameter values. Parameter values were sampled using Latin hypercube sampling such that each possible value was sampled exactly once [13,14]. For each parameter set, we refitted the model to the data and re-estimated the unknown parameters. In this way we explored a wide range of input parameters, but only with the restricted set of scenarios that best fit the observed data [14].

Partial rank correlation coefficients (PRCCs) were calculated for the correlation between each input parameter and four model outputs: the estimated time to diagnosis, the cumulative number of HIV infections, and the number and proportion of undiagnosed infections. In general, PRCC values near 1 (or -1) indicate a strong positive (or negative) influence of the input parameter on the estimated model output, whilst values near 0 indicate little influence.

Higher disease progression rates were generally associated with a shorter estimated time to diagnosis as indicated by negative PRCCs (eTable 2). To understand these associations it should be noted that a higher value of means a shorter duration of primary infection and hence a shorter time to diagnosis. A higher rate of progressing to the next CD4 stratum is compensated for by a higher diagnosis rate, and thus a shorter time to diagnosis, for the current CD4 stratum in order for the model to generate a certain number of HIV diagnoses that can be compared with observed data. An analogous argument explains the positive correlation between the proportion in each disease stage immediately after primary infection and the time to diagnosis. A higher proportion of patients entering a disease stage needs to be compensated by a lower diagnosis rate and hence a longer time between infection and diagnosis. Parameters related to the early stages of HIV infection had the largest influence on estimated time to diagnosis.

Similar associations were observed between input parameters and the number of infections. Since faster disease progression is compensated for by a shorter time to diagnosis, new HIV infections will be diagnosed more rapidly. Hence, the estimated number of infections is smaller in order to generate the same number of HIV diagnoses. As a consequence, the proportion of undiagnosed infections will also be smaller.

*Missing CD4 counts*

In the main model we implicitly assumed that CD4 counts were missing at random such that patients without CD4 count measurement had the same CD4 distribution as those with a CD4 measurement. Simulated data were used to assess the effect of missing CD4 data by randomly or non-randomly deleting certain proportions of the observed number of HIV diagnoses per CD4 count stratum and then repeating the model fits assuming that CD4 counts were missing at random. We considered various scenarios: (i) CD4 counts missing at random for 10% to 90% of all HIV diagnoses in steps of 10%, (ii) CD4 counts missing for 10% to 90% of HIV diagnoses with CD4 count <200 cells/mm3 (stage 4 in eFigure 1) and for smaller but equal proportions for the other three CD4 stages. For both types of scenarios, estimates of the annual number of HIV infections for all three simulated HIV epidemics were within the confidence intervals estimated in the main mode. The estimated time between infection to diagnosis by year of infection or by year of diagnosis was almost identical and well with the confidence intervals for scenario (i). However, for scenario (ii) estimates of time to diagnosis were consistently lower, up to 50% or 2 years, the higher the proportion of missing CD4 counts in stage 4 and the larger the difference between the proportions missing in stage 4 and the other three stages. This is because by (wrongly) assuming that CD4 counts were missing at random, the annual number of diagnoses with CD4 count <200 cells/mm3 is underestimated while the number of diagnoses in the other three CD4 strata is overestimated. Thus, the time between infection and diagnosis appears to be shorter.

**eTable 1**: Parameters used in the mathematical model for estimating HIV incidence and diagnosis rates. The range of was the reported 95% confidence interval. For the other rates the range was chosen to be 0.8 to 1.2 times the main value. Years are continuous variables with whole years representing 1 January-31 December. In the sensitivity analyses, new values were sampled within the given ranges using Latin hypercube sampling. Rates are per year.

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
| Description | Symbol | Value | Range | Source |
| Rate of progression from acute to chronic infection (per year) |  | 4.14 | 2.00 – 9.76 | [15] |
| Proportion in each disease stage directly after primary infection |  | 0.58 |  | [16,17] |
|  |  | 0.23 | 0.19 – 0.27 | [16,17] |
|  |  | 0.16 | 0.14 – 0.18 | [16,17] |
|  |  | 0.03 | 0.00 – 0.05 | [16,17] |
|  |  | 0 | – |  |
| Rate of progression to the next disease stage (per year) |  | 1/6.37 | 0.13 – 0.19 | [16,17] |
|  |  | 1/2.86 | 0.28 – 0.42 | [16,17] |
|  |  | 1/3.54 | 0.23 – 0.34 | [16,17] |
|  |  | 1/2.30 | 0.35 – 0.52 | [16,17] |
| Rate of progression from AIDS to death (per year) |  | 1/1.89 | 0.42 – 0.63 | [18,19] |
| Diagnosis rate by disease stage (per year) |  | estimated | – | – |
|  |  | estimated | – | – |
|  |  | estimated | – | – |
|  |  | estimated | – | – |
|  |  | 12 | – | [18,19] |
|  |  | 0 (<1984), | – | assumption |
|  |  | 0.4 (≥1984) | 0.2 – 0.6 | assumption |
| Mortality rate due to causes other than HIV (per year) |  | 0 | – | assumption |
| Start year of historical intervals for diagnosis rates |  | 1980 | – |  |
|  |  | 1984 | – |  |
|  |  | 1996 | 1995 – 1997 |  |
|  |  | 2000 | 1999 – 2001 |  |
|  |  | 2005 | 2004 – 2006 |  |
| Number of internal knots |  | 2 to 6 | – |  |

**eTable 2**: Partial rank correlation coefficients for the association between each input parameter and four model outcomes: the estimated time to diagnosis, the cumulative number of infections since 1980, the number of undiagnosed infections, and the proportion of undiagnosed infections. All model outcomes were evaluated at the end of 2011. \*\**p*<0.001; \**p*<0.01.

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
| Parameter | Time to diagnosis | Cumulative number of infections | Undiagnosed infections | % undiagnosed infections |
|  | \*\*-0.87 | \*\*-0.87 | \*\*-0.86 | \*\*-0.86 |
|  | \*\*-0.95 | \*\*-0.93 | \*\*-0.96 | \*\*-0.96 |
|  | \*\*-0.78 | \*\*-0.79 | \*\*-0.77 | \*\*-0.77 |
|  | \*\*-0.69 | \*\*-0.84 | \*\*-0.69 | \*\*-0.66 |
|  | \*-0.17 | \*\*-0.75 | -0.12 | 0.05 |
|  | 0.02 | \*\*0.72 | 0.07 | -0.07 |
|  | \*\*0.94 | \*\*0.95 | \*\*0.93 | \*\*0.93 |
|  | \*\*0.69 | \*\*0.83 | \*\*0.60 | \*\*0.55 |
|  | \*\*0.28 | \*\*0.53 | \*0.18 | 0.11 |
|  | \*0.19 | 0.11 | \*\*0.21 | \*\*0.25 |
|  | 0.07 | 0.03 | 0.04 | 0.04 |
|  | \*-0.20 | \*\*-0.52 | \*\*-0.35 | \*\*-0.38 |

**eFigure 1**: Complete model structure. HIV incidence over calendar time is denoted by . Immediately after infection, all individuals first enter a phase of primary infection, . After primary infection, individuals enter at a rate () one of five compartments of undiagnosed HIV infection that represent compartments with CD4 ≥500, 350-499, 200-349, and <200 cells/mm3, and AIDS, respectively. In the absence of treatment, individuals progress to the next compartment at a rate until they develop AIDS, stage , and then die at a rate . and are stages of AIDS-related death among undiagnosed and diagnosed HIV-infected patients, respectively. During each stage except primary infection individuals can be diagnosed at a rate , which depends on the stage and on calendar time, and then enter a compartment of diagnosed HIV infection. Dashed lines indicate that HIV progression is different in diagnosed individuals after starting antiretroviral treatment and that the model only considers HIV progression among diagnosed individuals in the period before 1996.



**eFigure 2**: Estimated and true number of infections for three different simulated HIV epidemics when fitting to the total annual number of HIV diagnoses instead of diagnoses by CD4 count stratum. Black solid lines show the model estimates, and dashed lines are 95% confidence intervals. Thin grey lines show results of multivariable sensitivity analyses. Grey dots are the true annual number of infections that were used as input in the simulations.



**eFigure 3**: Estimated and true number of undiagnosed infections for three different simulated HIV epidemics when fitting to the total annual number of HIV diagnoses instead of diagnoses by CD4 count stratum. Black solid lines show the model estimates, and dashed lines are 95% confidence intervals. Thin grey lines show results of multivariable sensitivity analyses. Grey dots are the true annual proportions undiagnosed infections.



**eFigure 4**: Model fits to reported HIV and AIDS cases in MSM in the Netherlands. (A) Annual number of new AIDS cases (+ signs) and concurrent HIV and AIDS diagnoses (dots); (B) annual number of HIV diagnoses. Black solid lines show the best model fit to the data, whilst black dashed lines are 95% confidence intervals. Black shapes are data points used for fitting, grey shapes are not used for fitting. Thin grey lines show results of multivariable sensitivity analyses. In panel B, the thick dashed grey line is the model estimate for the actual number of HIV diagnoses taking into account patients who did not survive up to 1996.



**eFigure 5**: Model fits to reported HIV diagnoses by CD4 count. The panels show the annual number of observed HIV diagnoses (black dots) with CD4 counts (A) ≥500 cells/mm3, (B) 350-499 cells/mm3, (C) 200-349 cells/mm3, and (D) <200 cells/mm3. Black solid lines show the model fit, and dashed lines are 95% confidence intervals. Thick grey lines are the actual number of HIV diagnoses taking into account patients who did not survive up to 1996 and patients for whom no CD4 was available. Thin grey lines show results of multivariable sensitivity analyses.



**eFigure 6**: Model outcomes for men who have sex with men (MSM) in the Netherlands when diagnosis rates in the period 1984-1995 were assumed to be a linear function of calendar time. (A) Annual number of new HIV infections; (B) average time from HIV infection to diagnosis by year of infection if diagnosis rates would remain the same as in the year of infection; (C) average time from HIV infection to diagnosis by year of diagnosis; (D) total number of individuals living with HIV and number of diagnosed and undiagnosed HIV infections, with dots representing the number of diagnosed MSM living with HIV according to the AIDS Therapy Evaluation in the Netherlands (ATHENA) database. Dashed grey lines in (A), (B), and (C) are results for the main model with constant diagnosis rate in 1984-1995, while thin grey lines show results of multivariable sensitivity analyses.



**eFigure 7**: Estimated diagnosis rates for the four CD4 count intervals (A) ≥500 cells/mm3, (B) 350-499 cells/mm3, (C) 200-349 cells/mm3, and (D) <200 cells/mm3. Solid lines show the estimated diagnosis rate, and dashed lines are 95% confidence intervals. Black lines are results when assuming a constant diagnosis rate in the period 1984-1995, while grey lines represent results when assuming a linear function of calendar time.



**eFigure 8**: Model outcomes for men who have sex with men in the Netherlands when no information on CD4 counts was used. (A) Annual number of new HIV infections; (B) average time from HIV infection to diagnosis by year of infection if parameters would not change. Black solid lines show the best model fit to the data, whilst black dashed lines are 95% confidence intervals. Thin grey lines show results of multivariable sensitivity analyses.



References

1. Ramsay JO. Monotone Regression Splines in Action. *Statistical Science* 1988; **3**:425-441.

2. Alioum A, Commenges D, Thiebaut R, Dabis F. A multistate approach for estimating the incidence of human immunodeficiency virus by using data from a prevalent cohort study. *Appl Statist* 2005; **54**:739-752.

3. Lee CA, Phillips AN, Elford J, Janossy G, Griffiths P, Kernoff P. Progression of HIV disease in a haemophilic cohort followed for 11 years and the effect of treatment. *BMJ* 1991; **303**:1093-1096.

4. Morgan D, Mahe C, Mayanja B, Whitworth JA. Progression to symptomatic disease in people infected with HIV-1 in rural Uganda: prospective cohort study. *BMJ* 2002; **324**:193-196.

5. Flegg PJ. Barnett Christie Lecture (1993). The natural history of HIV infection: a study in Edinburgh drug users. *J Infect* 1994; **29**:311-321.

6. Schouten M, van Velde AJ, Snijdewind IJ, Verbon A, Rijnders BJ, van der Ende ME. [Late diagnosis of HIV positive patients in Rotterdam, the Netherlands: risk factors and missed opportunities]. *Ned Tijdschr Geneeskd* 2013; **157**:A5731.

7. British HIV Association Audit & Standards Sub-Committee. 2010-11 survey of HIV testing policy and practice and audit of new patients when first seen post-diagnosis. 2011 Available at: www.bhiva.org/documents/ClinicalAudit/FindingsandReports/HIVdiagnosisWebVersion.ppt.

8. Mccullagh P. Quasi-Likelihood Functions. *Annals of Statistics* 1983; **11**:59-67.

9. Efron B, Tibshirani RJ. An Introduction to the Bootstrap. New York: Chapman & Hall/CRC, **1993**.

10. Phillips AN, Sabin C, Pillay D, Lundgren JD. HIV in the UK 1980-2006: reconstruction using a model of HIV infection and the effect of antiretroviral therapy. *HIV Med* 2007; **8**:536-546.

11. Phillips AN, Cambiano V, Nakagawa F *et al*. Increased HIV incidence in men who have sex with men despite high levels of ART-induced viral suppression: analysis of an extensively documented epidemic. *PLoS ONE* 2013; **8**:e55312.

12. Health Council of the Netherlands: Standing Committee on Infectious Diseases and Immunology. Reconsidering the policy on HIV testing. Publication no. 1999/02. The Hague, Health Council of the Netherlands, 1999.

13. Sanchez MA, Blower SM. Uncertainty and sensitivity analysis of the basic reproductive rate. Tuberculosis as an example. *Am J Epidemiol* 1997; **145**:1127-1137.

14. van Sighem A, Vidondo B, Glass TR *et al*. Resurgence of HIV infection among men who have sex with men in Switzerland: mathematical modelling study. *PLoS ONE* 2012; **7**:e44819.

15. Hollingsworth TD, Anderson RM, Fraser C. HIV-1 transmission, by stage of infection. *J Infect Dis* 2008; **198**:687-693.

16. Lodi S, Phillips A, Touloumi G *et al*. Time from human immunodeficiency virus seroconversion to reaching CD4+ cell count thresholds <200, <350, and <500 Cells/mm(3): assessment of need following changes in treatment guidelines. *Clin Infect Dis* 2011; **53**:817-825.

17. Cori A, Ayles H, Beyers N *et al*. HPTN 071 (PopART): A Cluster-Randomized Trial of the Population Impact of an HIV Combination Prevention Intervention Including Universal Testing and Treatment: Mathematical Model. *PLoS ONE* 2014; **9**:e84511.

18. Bezemer D, de Wolf F, Boerlijst MC *et al*. A resurgent HIV-1 epidemic among men who have sex with men in the era of potent antiretroviral therapy. *AIDS* 2008; **22**:1071-1077.

19. Bezemer D, de Wolf F, Boerlijst MC, van Sighem A, Hollingsworth TD, Fraser C. 27 years of the HIV epidemic amongst men having sex with men in the Netherlands: an in depth mathematical model-based analysis. *Epidemics* 2010; **2**:66-79.