

INITIAL PLASMA HIV-1 RNA LEVELS AND PROGRESSION TO AIDS IN WOMEN AND MEN

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ABSTRACT

Background It is unclear whether there are differences between men and women with human immunodeficiency virus type 1 (HIV-1) infection in the plasma level of viral RNA (the viral load). In men, the initial viral load after seroconversion predicts the likelihood of progression to the acquired immunodeficiency syndrome (AIDS), but the relation between the two has not been assessed in women. Currently, the guidelines for initiating antiretroviral therapy are applied uniformly to women and men.

Methods From 1988 through 1998, the viral load and the CD4+ lymphocyte count were measured approximately every six months in 156 male and 46 female injection-drug users who were followed prospectively after HIV-1 seroconversion.

Results The median initial viral load was 50,766 copies of HIV-1 RNA per milliliter in the men but only 15,103 copies per milliliter in the women ($P < 0.001$). The median initial CD4+ count did not differ significantly according to sex (659 and 672 cells per cubic millimeter, respectively). HIV-1 infection progressed to AIDS in 29 men and 15 women, and the risk of progression did not differ significantly according to sex. For each increase of 1 log in the viral load (on a base 10 scale), the hazard ratio for progression to AIDS was 1.55 (95 percent confidence interval, 0.97 to 2.47) among the men and 1.43 (95 percent confidence interval, 0.76 to 2.69) among the women. The median initial viral load was 77,822 HIV-1 RNA copies per milliliter in the men in whom AIDS developed and 40,634 copies per milliliter in the men in whom it did not; the corresponding values in the women were 17,149 and 12,043 copies per milliliter. Given the recommendation that treatment should be initiated when the viral load reaches 20,000 copies per milliliter, 74 percent of the men but only 37 percent of the women in our study would have been eligible for therapy at the first visit after seroconversion ($P < 0.001$).

Conclusions Although the initial level of HIV-1 RNA was lower in women than in men, the rates of progression to AIDS were similar. Treatment guidelines that are based on the viral load, rather than the CD4+ lymphocyte count, will lead to differences in eligibility for antiretroviral treatment according to sex. (N Engl J Med 2001;344:720-5.)

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STUDIES of a possible difference between men and women in the plasma level of human immunodeficiency virus type 1 (HIV-1) RNA (the viral load) have had conflicting results. Some cross-sectional^{1,2} and longitudinal³⁻⁵ studies have found lower plasma HIV-1 RNA levels in women than in men after controlling for the CD4+ lymphocyte count, but two cross-sectional studies did not find a difference.^{6,7} We previously observed that the difference between men and women in the viral load was greatest soon after seroconversion and diminished over time, suggesting different viral dynamics in women and men.⁴ Previous studies of difference between men and women in the viral load have been limited by small samples,^{3,6} a cross-sectional^{1,2,7,8} or nested case-control⁴ design, or use of different assays to determine the viral load in men and women.⁹

In men, viral load after HIV-1 seroconversion is an independent predictor of the risk of progression to the acquired immunodeficiency syndrome (AIDS).¹⁰⁻¹⁶ Viral load is the basis for the current guidelines for the initiation of antiretroviral therapy, which apply uniformly to women and men.^{17,18} The relation between the initial viral load and the risk of progression to AIDS in women has not been studied.

In a prospective cohort study of injection-drug users, we measured the viral load by means of the reverse-transcriptase polymerase chain reaction (RT-PCR) in all participants who had HIV-1 seroconversion. We also assessed the association between the initial viral load and the rate of progression to AIDS in women and in men and determined the effects of the current guidelines for antiretroviral therapy on eligibility for treatment.

METHODS

Study Population

Between February 1988 and March 1989, injection-drug users in Baltimore were enrolled in a longitudinal study of HIV-1 infection.¹⁹ There was a second period of enrollment from August 1994 to June 1995. Persons were eligible for enrollment if they were at least 18 years old and free of AIDS and if they had used injection drugs at least once since 1977. A total of 3380 injection-

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drug users were enrolled. All participants had blood drawn for HIV serologic testing semiannually; seroconversion was determined only by means of this semiannual testing. When participants were first identified as HIV-seropositive, they were asked to return to the study center so that blood could be drawn for measurement of the CD4+ lymphocyte count, with plasma frozen for later testing. This procedure and a physical examination were then repeated semiannually. Women and men were evaluated at the same location, and the processing of blood specimens did not differ according to sex. The study was approved by the institutional review board of the Johns Hopkins University School of Public Health, and written informed consent was obtained from all participants.

The criteria for inclusion in the current study were documented HIV-1 seroconversion within 12 months after the last visit at which the participant was seronegative and before December 1, 1997; measurement of the viral load within 12 months after the estimated date of seroconversion (defined as the midpoint between the last visit at which the participant was seronegative and the first visit at which he or she was seropositive); and at least three measurements of the viral load after seroconversion. All seropositive participants were referred to primary care providers for management of HIV infection.

Data Collection

For each participant in the study, demographic data and the medical history were obtained at the initial visit, and self-reported information on use of injection drugs and medications (including antiretroviral therapy) during the previous six months was obtained semiannually. Using standard forms, trained nurses abstracted information on AIDS-defining diagnoses and AIDS-related deaths from medical records and death certificates, respectively, and an end-points committee led by a physician established the final diagnoses. AIDS-defining diagnoses were based on the 1993 clinical case definition established by the Centers for Disease Control and Prevention,²⁰ except that a CD4+ lymphocyte count of less than 200 per cubic millimeter was excluded as a sufficient condition for a diagnosis of AIDS. Outcomes that occurred before December 31, 1998, were included in the analysis. All measurements of viral load and CD4+ lymphocyte counts obtained before AIDS was diagnosed (or before December 31, 1998, in persons in whom AIDS did not develop) were included in the analysis. Measurements of viral load and CD4+ lymphocyte counts from medical records not obtained as part of the study were not included. The initial viral load was defined as the first viral load measured within 12 months after the estimated date of seroconversion. The follow-up time was calculated as the interval between the estimated date of seroconversion and the diagnosis of AIDS or the last date on which the viral load was measured in participants in whom AIDS did not develop.

Laboratory Studies

Antibodies to HIV-1 were measured with a commercially available enzyme-linked immunosorbent assay kit (Genetic Systems, Seattle), and positive results were confirmed by a Western blot assay (Dupont, Wilmington, Del.). T-cell subpopulations were measured by means of whole-blood staining methods and flow-cytometric procedures.^{21,22} All plasma specimens were stored at -70°C until testing was performed. Levels of HIV-1 RNA in plasma were quantified by means of an RT-PCR assay (Roche Molecular Systems, Branchburg, N.J.) according to the manufacturer's protocol for thawed plasma. For heparin-treated plasma (collected through April 1997), viral RNA was obtained by means of a silica-based method of extraction.²³ After April 1997, plasma was collected in tubes containing EDTA. The minimal detectable level of HIV-1 RNA was 400 copies per milliliter, and the dynamic range of the assay was approximately 4 log on a base 10 scale. Undetectable viral loads were coded as 200 copies per milliliter.

Statistical Analysis

The Wilcoxon rank-sum test was used for comparisons of continuous variables. The chi-square test was used for comparisons

of categorical variables, with Fisher's two-tailed exact test used when the sample was small. Generalized estimating equations were used to compare repeated measures. We used multiple linear regression analysis, while adjusting for confounders and testing for interactions, to identify predictors of the initial HIV-1 RNA level. A Kaplan-Meier analysis of the time to the diagnosis of AIDS according to sex was performed; the significance of the difference between the curves was assessed with the log-rank test. Multivariate proportional-hazards models were used to determine which factors that were present shortly after seroconversion were independent predictors of the progression to AIDS. Cross-sectional comparisons of viral load and CD4+ lymphocyte categories were made at yearly intervals after seroconversion. If there were multiple measurements for one of these intervals, the value obtained closest to the beginning of the interval was used. All reported P values are two-sided.

RESULTS

Study Participants

There were 295 participants in the longitudinal study who underwent HIV-1 seroconversion during the study period (222 men and 73 women). Of these, 93 participants were ineligible for this study because more than 12 months had elapsed between the last visit at which the participant was seronegative and the first visit at which the participant was seropositive (in 39 participants), there were fewer than three measurements of the viral load (in 35), or more than 12 months had elapsed between the estimated time of seroconversion and the first measurement of the viral load (in 19). Of the 202 participants with HIV-1 seroconversion who met the criteria for inclusion, 156 were men (77 percent) and 46 were women (23 percent), a sex distribution similar to that for all the participants in whom seroconversion occurred.

The clinical characteristics of the study participants are shown in Table 1. The men and the women were similar except that the women were younger at the time of seroconversion and had somewhat longer follow-up than the men. There were no significant differences between men and women in the frequency of missed study visits before seroconversion occurred or the time that elapsed between the estimated date of seroconversion and the first measurement of viral load. Data on pregnancy were available for 146 visits by women; at 7 of these visits (4.8 percent), women reported being pregnant, and at 5 of these visits (3.4 percent), women reported having had a miscarriage or an abortion during the previous six months.

At the initial visit after seroconversion, none of the participants reported that they were receiving antiretroviral therapy. Overall, the use of one or more antiretroviral drugs was reported at 22 percent of the study visits by men and at 23 percent of the visits by women ($P=0.93$) (Table 1). Of the reports of antiretroviral-drug use, 80 percent involved therapy with nucleoside analogues only (a single nucleoside in 55 percent and two nucleosides in 25 percent). These figures did not differ according to sex. The receipt of highly active antiretroviral therapy (defined as a reg-

TABLE 1. CLINICAL CHARACTERISTICS OF THE 202 PARTICIPANTS WITH HIV-1 SEROCONVERSION.*

CHARACTERISTIC	MEN (N=156)	WOMEN (N=46)	P VALUE
Black race (%)	94	93	0.99†
Median age at seroconversion (yr)	36.7 (32.1–41.8)	32.6 (29.7–37.4)	0.002‡
Median ratio of no. of years between study entry and seroconversion to no. of visits before seroconversion	0.55 (0.46–0.63)	0.53 (0.46–0.56)	0.12‡
Median interval from seroconversion to first viral-load measurement (mo)	4.3 (3.6–5.2)	4.1 (3.2–5.0)	0.45‡
Median follow-up (yr)	4.6 (3.2–6.8)	6.0 (3.3–7.9)	0.10‡
Antiretroviral drug use reported (% of study visits)	22	23	0.93§

*Values in parentheses are interquartile ranges.

†Fisher's two-tailed exact test was used.

‡The Wilcoxon rank-sum test was used.

§Generalized estimating equations were used.

imen that included an HIV-1 protease inhibitor or a non-nucleoside reverse-transcriptase inhibitor) was reported at less than 5 percent of the study visits by both women and men.

Initial Viral Load and Progression to AIDS According to Sex

The median initial viral load after seroconversion was significantly lower in women than in men (15,103 vs. 50,766 copies of HIV-1 RNA per milliliter, $P < 0.001$); CD4+ lymphocyte counts did not differ according to sex (Table 2). The median initial viral load remained approximately 0.5 log lower in women than in men after adjustment for the age at seroconversion, the time between the estimated date of seroconversion and the first measurement of the viral load ($P = 0.001$), and the CD4+ lymphocyte count at seroconversion ($P = 0.001$) in multivariate linear models. The difference according to sex in viral load persisted at the second visit after seroconversion (data not shown).

HIV infection progressed to AIDS in 29 men and 15 women; a Kaplan–Meier analysis of the time to progression did not demonstrate a significant difference according to sex ($P = 0.18$ by the log-rank test) (Fig. 1). In addition, a Cox proportional-hazards model of the time to a diagnosis of AIDS, in which sex was a covariate, showed that the risk of AIDS was not significantly greater for women than for men (hazard ratio for women, 1.53; 95 percent confidence interval, 0.8 to 2.9; $P = 0.18$). The relative proportions of AIDS-defining diagnoses did not differ according to sex, with the exception of *Pneumocystis carinii* pneumonia, which accounted for 3 of the 29 AIDS-defining diagnoses in men (10 percent) but for 6 of 15 in women (40 percent, $P = 0.04$). Among the participants with fewer than 200 CD4+ lymphocytes per cubic millimeter, self-reports of medications used routinely for prophylaxis against *P. carinii* pneumonia

TABLE 2. INITIAL PLASMA HIV-1 RNA LEVELS AND CD4+ LYMPHOCYTE COUNTS AFTER SEROCONVERSION.

VARIABLE	MEN	WOMEN	P VALUE*
Median plasma HIV-1 RNA level (copies/ml)	50,766	15,103	<0.001
Median CD4+ lymphocyte count (per mm ³)	659	672	0.48

*P values were determined with the Wilcoxon rank-sum test.

(trimethoprim–sulfamethoxazole, dapsone, or pentamidine) did not differ significantly according to sex (reported in 23 percent of visits by men and 31 percent of visits by women, $P = 0.29$).

To determine whether the initial viral load was a predictor of progression to AIDS, we used proportional-hazards models in which the time to the diagnosis of AIDS was the dependent variable. In separate univariate models for each sex, for each 1-log increase in the initial viral load, the hazard ratio for progression to AIDS was 1.55 in men (95 percent confidence interval, 0.97 to 2.47; $P = 0.07$) and 1.43 in women (95 percent confidence interval, 0.76 to 2.69; $P = 0.27$). In separate multivariate proportional-hazards models for men and women, in which the initial CD4+ lymphocyte count and age were controlled for, the hazard ratios for progression to AIDS for each 1-log increase in the initial viral load remained similar but were not statistically significant (Table 3).

Among the 29 men in whom HIV infection progressed to AIDS, the median initial viral load was 77,822 copies per milliliter, as compared with 40,634 copies per milliliter among the 127 men in whom

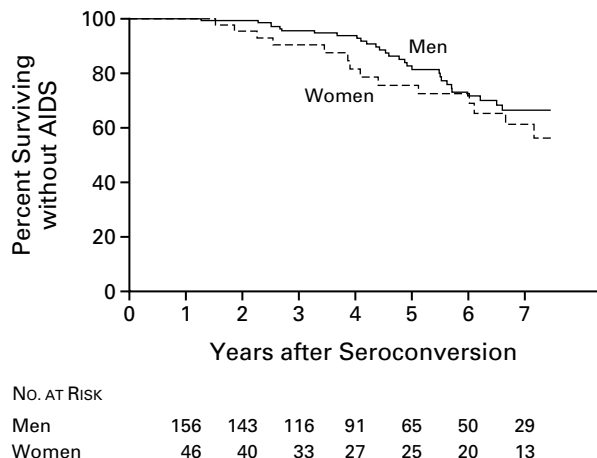


Figure 1. Kaplan-Meier Estimates of Survival without Progression to AIDS According to Sex.

The curves represent the percentages of patients surviving without AIDS during the seven years after seroconversion. The numbers of men and women at risk during each 12-month interval are given below the graph. There was no significant difference between men and women in the risk of progression to AIDS ($P=0.18$ by the log-rank test).

the infection did not progress to AIDS. Among the 15 women with progression to AIDS and the 31 women without progression, the corresponding values were 17,149 and 12,043 copies per milliliter. The median initial CD4+ lymphocyte count after seroconversion did not differ significantly between the participants in whom AIDS developed and those in whom it did not, and the result was not affected by stratification according to sex (data not shown).

DISCUSSION

Our study is one of the largest cohort studies to date of men and women followed from the time of HIV-1 seroconversion, and it confirms earlier reports that plasma HIV-1 RNA levels are lower in women than in men.^{1,3,6,9} This difference remained significant after we had controlled for age, the interval between seroconversion and the initial measurement of the viral load, and the CD4+ lymphocyte count. In addition, the difference between men and women in the viral load persisted for several years after seroconversion.

Since the initial viral load after seroconversion predicts the likelihood of progression to AIDS in men, one would expect that women would be at lower risk for AIDS than men, given their initially lower viral load. However, several studies have found that the risk of AIDS does not differ significantly between men and women,²⁴⁻²⁶ and in our study, there was also no significant difference in the risk of AIDS according to sex. The mechanism by which HIV infec-

TABLE 3. HAZARD RATIOS FOR PROGRESSION TO AIDS IN MEN AND WOMEN.*

VARIABLE	HAZARD RATIO (95% CI)	P VALUE
Men		
HIV-1 viral load (per 1-log increase)	1.55 (0.95–2.52)	0.08
CD4+ lymphocyte count (per 100-cell decrease)	1.01 (0.88–1.15)	0.95
Age (per 1-yr increase)	1.08 (1.03–1.13)	0.002
Women		
HIV-1 viral load (per 1-log increase)	1.86 (0.94–3.67)	0.08
CD4+ lymphocyte count (per 100-cell decrease)	0.77 (0.58–1.03)	0.07
Age (per 1-yr increase)	0.93 (0.83–1.03)	0.18

*Separate multivariate proportional-hazards models were used for men and women, on the basis of data from the initial visit after seroconversion. CI denotes confidence interval.

tion in women progresses to AIDS at the same rate as it does in men despite the lower initial viral load in women is unknown. The median initial viral load in the men in our study (50,766 HIV-1 RNA copies per milliliter) was similar to that reported by the Multicenter AIDS Cohort Study for the first year after seroconversion (33,759 copies per milliliter),¹⁶ which suggests that the results can be generalized to men with HIV-1 infection in the United States.

In our study, for each increase of 1 log in the initial viral load, the hazard ratio for progression to AIDS was similar in men and women (1.55 and 1.43, respectively). The hazard ratio for men was of borderline statistical significance, but the hazard ratio for the smaller sample of women was not significant. However, the median initial viral load was higher in both the men and the women in whom HIV infection subsequently progressed to AIDS than in those in whom it did not.

Although the relative viral load appeared to have a similar predictive value for progression to AIDS, the same absolute viral load conferred different risks of AIDS among women and men. For example, an initial viral load of 17,149 copies per milliliter was associated with progression to AIDS in women but not in men. The median initial viral load among the men in whom HIV infection did not progress to AIDS was 40,634 copies per milliliter. This difference is important because of the cutoff value (more than 20,000 copies per milliliter) used in current guidelines for the initiation of antiretroviral therapy in both women and men.¹⁸

To assess the effect of our findings with respect to the guidelines of the Department of Health and Human Services, we compared the proportions of men and women who had an initial viral load of more than 20,000 copies per milliliter after seroconversion (Table 4). Given this cutoff value, 115 of the 156 men

TABLE 4. ELIGIBILITY OF MEN AND WOMEN FOR ANTIRETROVIRAL THERAPY ON THE BASIS OF CURRENT TREATMENT GUIDELINES.*

YEAR AFTER SEROCONVERSION	TOTAL		VIRAL LOAD >20,000 COPIES/ml			VIRAL LOAD >30,000 COPIES/ml			CD4+ LYMPHOCYTE COUNT <500 PER mm ³		
	MEN	WOMEN	MEN	WOMEN	P VALUE	MEN	WOMEN	P VALUE	MEN	WOMEN	P VALUE
	no.		% eligible			% eligible			% eligible		
First	156	46	74	37	<0.001	61	33	0.001	31	28	0.68
Second	144	42	59	43	0.06	45	33	0.17	52	48	0.58
Third	132	36	56	28	0.003	45	28	0.07	60	56	0.61
Fourth	108	32	52	34	0.08	41	28	0.20	65	66	0.93
Fifth	80	25	54	40	0.23	44	28	0.16	66	68	0.87
Sixth	64	23	47	39	0.52	39	30	0.46	67	70	0.83

*P values were determined with the chi-square test.

(74 percent; 95 percent confidence interval, 67 to 81 percent) and 17 of the 46 women (37 percent; 95 percent confidence interval, 23 to 51 percent) would have been eligible for antiretroviral therapy during the first year after seroconversion ($P < 0.001$). With the use of a cutoff value of more than 30,000 copies per milliliter, as recommended by the International AIDS Society,¹⁷ more men than women would still have been eligible for antiretroviral therapy, but the difference would have been smaller. The difference between men and women in terms of eligibility was statistically significant during the first few years after seroconversion and then became less pronounced (Table 4).

There was no significant difference in the proportions of men and women who would have been eligible for therapy solely on the basis of an initial CD4+ lymphocyte count of less than 500 per cubic millimeter. During the first year after seroconversion, 31 percent of men (95 percent confidence interval, 24 to 38 percent) and 28 percent of women (95 percent confidence interval, 13 to 43 percent) would have been eligible ($P = 0.68$). These comparisons of eligibility changed very little after adjustment for self-reports of the receipt of antiretroviral therapy and after the exclusion of the data on viral loads and CD4 counts obtained at visits at which antiretroviral therapy was reported (data not shown).

Given the lower initial viral load in women, questions have been raised about whether antiretroviral therapy should be initiated at a lower viral load in women than in men. Some investigators suggest, in contrast, that the cutoff values for CD4+ lymphocytes and plasma viral load used in the current guidelines^{17,18} lead to premature use of antiretroviral therapy in both women and men.²⁷ To address these two issues, several factors must be considered. First, despite the lower initial viral load in women than in men, several large prospective studies have found no

difference according to sex in the progression to AIDS.²⁴⁻²⁶ Second, a viral-load cutoff value above which rapid progression to AIDS can be predicted has not been identified for either women or men. Third, during the first few years after seroconversion, when the difference between men and women in the viral load is greatest, the risk of progression to AIDS is lowest. Later in the course of infection, when the risk of AIDS is greater, there is no longer a sex-based difference in the viral load.⁴ Fourth, the CD4+ lymphocyte count is critical in predicting the risk of opportunistic infections and is a better predictor of mortality than is the viral load.^{28,29} Finally, the survival advantage associated with highly active antiretroviral therapy is compromised only if such therapy is withheld until the CD4+ lymphocyte count is less than 200 per cubic millimeter.^{29,30} Although we did not determine the optimal time for initiating therapy, these factors suggest that the cutoff values in the current guidelines should be reassessed in the light of the equal risk of disease progression for men and women and their equal eligibility for therapy if the CD4+ lymphocyte count is the criterion.

There are several limitations to this study. First, the receipt of antiretroviral therapy was reported by the study participants, and they were not asked about the duration of such therapy. Although participants' reports may not be completely accurate, the most likely error would be an overestimate of antiretroviral-drug use, not an underestimate. Thus, confounding due to unreported antiretroviral therapy is unlikely. Second, the study cohort was composed entirely of injection-drug users. However, a recent, large collaborative study in subjects with HIV-1 seroconversion found no difference in the risk of AIDS or death according to the category of exposure to HIV.²⁶ In addition, more than 90 percent of the participants in our study were black, a fact that could limit the generalizability of the results. However, race has not been

found to affect viral load or the natural history of HIV-1 infection.^{31,32}

The implications of the difference in viral load between men and women for the initiation of antiretroviral therapy require further investigation. In addition, studies are needed to determine whether there is a threshold value for the viral load that predicts progression to AIDS. Finally, investigation into the biologic mechanism underlying the lower initial viral load in women may provide insight into the pathogenesis of HIV-1 infection in both women and men.

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REFERENCES

- Katzenstein DA, Hammer SM, Hughes MD, et al. The relation of virologic and immunologic markers to clinical outcomes after nucleoside therapy in HIV-infected adults with 200 to 500 CD4 cells per cubic millimeter. *N Engl J Med* 1996;335:1091-8. [Erratum, *N Engl J Med* 1997;337:1097.]
- Farzadegan H, Hoover DR, Astemborski J, et al. Sex differences in HIV-1 viral load and progression to AIDS. *Lancet* 1998;352:1510-4.
- Evans JS, Nims T, Cooley J, et al. Serum levels of virus burden in early-stage human immunodeficiency virus type 1 disease in women. *J Infect Dis* 1997;175:795-800.
- Sterling TR, Lyles CM, Vlahov D, Astemborski J, Margolick JB, Quinn TC. Sex differences in longitudinal human immunodeficiency virus type 1 RNA levels among seroconverters. *J Infect Dis* 1999;180:666-72.
- Lyles CM, Dorrucchi M, Vlahov D, et al. Longitudinal human immunodeficiency virus type 1 load in the Italian Seroconversion Study: correlates and temporal trends of virus load. *J Infect Dis* 1999;180:1018-24.
- Bush CE, Donovan RM, Markowitz N, Baxa D, Kvale P, Saravolatz LD. Gender is not a factor in serum human immunodeficiency virus type 1 RNA levels in patients with viremia. *J Clin Microbiol* 1996;34:970-2.
- Moore RD, Cheever L, Keruly JC, Chaisson RE. Lack of sex difference in CD4 to HIV-1 RNA viral load ratio. *Lancet* 1999;353:463-4.
- Junghans C, Ledergerber B, Chan P, Weber R, Egger M. Sex differences in HIV-1 viral load and progression to AIDS: Swiss HIV Cohort Study. *Lancet* 1999;353:589.
- Anastos K, Gange SJ, Lau B, et al. Association of race and gender with HIV-1 RNA levels and immunologic progression. *J Acquir Immune Defic Syndr* 2000;24:218-26.
- Jurriaans S, van Gemen B, Weverling GJ, et al. The natural history of HIV-1 infection: virus load and virus phenotype independent determinants of clinical course? *Virology* 1994;204:223-33.
- Mellors JW, Kingsley LA, Rinaldo CR Jr, et al. Quantitation of HIV-1 RNA in plasma predicts outcome after seroconversion. *Ann Intern Med* 1995;122:573-9.
- Farzadegan H, Henrard DR, Kleiberger CA, et al. Virologic and serologic markers of rapid progression to AIDS after HIV-1 seroconversion. *J Acquir Immune Defic Syndr Hum Retrovirol* 1996;13:448-55.
- Henrard DR, Phillips JF, Muenz LR, et al. Natural history of HIV-1 cell-free viremia. *JAMA* 1995;274:554-8.
- Craib KJ, Strathdee SA, Hogg RS, et al. Serum levels of human immunodeficiency virus type 1 (HIV-1) RNA after seroconversion: a predictor of long-term mortality in HIV infection. *J Infect Dis* 1997;176:798-800.
- Mellors JW, Munoz A, Giorgi JV, et al. Plasma viral load and CD4+ lymphocytes as prognostic markers of HIV-1 infection. *Ann Intern Med* 1997;126:946-54.
- Lyles RH, Munoz A, Yamashita TE, et al. Natural history of human immunodeficiency virus type 1 viremia after seroconversion and proximal to AIDS in a large cohort of homosexual men: Multicenter AIDS Cohort Study. *J Infect Dis* 2000;181:872-80.
- Carpenter CCJ, Cooper DA, Fischl MA, et al. Antiretroviral therapy in adults: updated recommendations of the International AIDS Society-USA Panel. *JAMA* 2000;283:381-90.
- Report of the NIH Panel to Define Principles of Therapy of HIV Infection and guidelines for the use of antiretroviral agents in HIV-infected adults and adolescents. *MMWR Morb Mortal Wkly Rep* 1998;47(RR-5):1-82. (Updated January 2000 as a living document. [See <http://www.hivatis.org/trtdlms.html>].)
- Vlahov D, Anthony JC, Munoz A, et al. The ALIVE study: a longitudinal study of HIV infection in intravenous drug users: description of methods. *J Drug Issues* 1991;21:759-76.
- 1993 Revised classification system for HIV infection and expanded surveillance case definition for AIDS among adolescents and adults. *MMWR Morb Mortal Wkly Rep* 1992;41(RR-17):1-19.
- Hoffman RA, Kung PC, Hansen WP, Goldstein G. Simple and rapid measurement of human T-lymphocytes and their subclasses in peripheral blood. *Proc Natl Acad Sci U S A* 1980;77:4914-7.
- Giorgi JV, Cheng HL, Margolick JB, et al. Quality control in the flow cytometric measurement of T-lymphocyte subsets: the Multicenter AIDS Cohort Study experience. *Clin Immunol Immunopathol* 1990;55:173-86.
- Boom R, Sol CJA, Salimans MMM, Jansen CL, Wertheim-van Dillen PME, van der Noordaa J. Rapid and simple method for purification of nucleic acids. *J Clin Microbiol* 1990;28:495-503.
- Melnick SL, Sherer R, Louis TA, et al. Survival and disease progression according to gender of patients with HIV infection: the Terry Bein Community Programs for Clinical Research on AIDS. *JAMA* 1994;272:1915-21.
- Chaisson RE, Keruly JC, Moore RD. Race, sex, drug use, and progression of human immunodeficiency virus disease. *N Engl J Med* 1995;333:751-6.
- Collaborative Group on AIDS Incubation and HIV Survival. Time from HIV-1 seroconversion to AIDS and death before widespread use of highly-active antiretroviral therapy: a collaborative re-analysis. *Lancet* 2000;355:1131-7.
- Henry K. The case for more cautious, patient-focused antiretroviral therapy. *Ann Intern Med* 2000;132:306-11.
- Anastos K, Kalish LA, Hessel N, et al. The relative value of CD4 cell count and quantitative HIV-1 RNA in predicting survival in HIV-1 infected women: results of the Women's Interagency HIV Study. *AIDS* 1999;13:1717-26.
- Montaner J, Hogg R, Yip B, Chan K, Craib K, O'Shaughnessy M. To start or not to start? Diminished effectiveness of anti-retroviral therapy among patients initiating therapy with CD4+ cell counts below 200/mm³. In: Late breaker abstract book of the 13th International AIDS Conference, Durban, South Africa, July 9–14, 2000:45. abstract.
- Anastos K, Barron Y, Weiser B, et al. Risk of progression to AIDS and death in HIV infected women initiating HAART at different stages of disease. In: Late breaker abstract book of the 13th International AIDS Conference, Durban, South Africa, July 9–14, 2000:332. abstract.
- Brown AE, Malone JD, Zhou SYJ, Lane JR, Hawkes CA. Human immunodeficiency virus RNA levels in US adults: a comparison based on race and ethnicity. *J Infect Dis* 1997;176:794-7.
- Pezzotti P, Galai N, Vlahov D, Rezza G, Lyles CM, Astemborski J. Direct comparison of time to AIDS and infectious disease death between HIV seroconverter injection drug users in Italy and the United States: results from the ALIVE and ISS studies: AIDS Link to Intravenous Experiences: Italian Seroconversion Study. *J Acquir Immune Defic Syndr Hum Retrovirol* 1999;20:275-82.

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