Prognostic importance of anaemia in HIV-1 infected patients starting antiretroviral therapy: collaborative analysis of prospective cohort studies in industrialized countries

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Abstract

Background—In HIV-1 infected patients starting highly active antiretroviral therapy (HAART), the prognostic value of haemoglobin when starting HAART, and of changes in haemoglobin levels, are not well defined.

Methods—We combined data from 10 prospective studies of 12,100 previously untreated individuals (25% women). A total of 4,222 patients (35%) were anaemic: 131 patients (1.1%) had severe (<8.0 g/dl), 1120 (9%) had moderate (males: 8.0 to <11.0 g/dl, females 8.0 to <10.0 g/dl) and 2971 (25%) had mild anaemia (males 11.0 to <13.0 g/dl, females 10.0 to <12.0 g/dl). We separately analyzed progression to AIDS or death from baseline and six months using Weibull models, adjusting for CD4 cell count, age, sex, and other variables.

Results—During 48,420 person-years of follow-up 1,448 patients developed at least one AIDS event and 857 patients died. Anaemia at baseline was independently associated with higher mortality: the adjusted hazard ratio (95% confidence interval) for mild anaemia was 1.42 (1.17–1.73), for moderate anaemia 2.56 (2.07–3.18) and for severe anaemia 5.26 (3.55–7.81). Corresponding figures for progression to AIDS were 1.60 (1.37–1.86), 2.00 (1.66–2.40) and 2.24 (1.46–3.42). At 6 months the prevalence of anaemia had declined to 26%. Baseline anaemia continued to predict mortality (and to a lesser extent progression to AIDS) in patients with normal haemoglobin or mild anaemia at 6 months.

Conclusions—Anaemia at the start of HAART is an important prognostic factor for short and long term prognosis, including in patients whose haemoglobin levels improve or normalize during the first 6 months of HAART.

Keywords

HIV/AIDS; highly active antiretroviral therapy (HAART); anaemia; prognosis; mortality

Introduction

The prognosis of HIV-1 infected patients has been dramatically improved by highly active antiretroviral therapy (HAART), which typically consists of a combination of three drugs [1–4]. Based on data combined from HIV cohort studies in Europe and North America, the Antiretroviral Therapy (ART) Cohort Collaboration developed a prognostic model [5;6] that is widely used to estimate the risk of AIDS and death in treatment-naïve patients starting HAART. A low CD4 T lymphocyte cell (CD4 cell) count at the time of starting HAART was most strongly associated with progression to AIDS and death, and a previous AIDS-defining event, high viral load, transmission via injection drug use and older age are also associated with worse prognosis [5].

Several studies from North America and Europe [7–13] have shown that anaemia in HIV-infected patients is associated with higher rates of disease progression and death, independently of the CD4 cell count and other prognostic factors. These studies were mainly based on data from the pre-HAART era, or included patients who had been exposed to myelosuppressive antiretroviral drugs, particularly zidovudine, before starting HAART. The Anaemia in HIV Working Group recently reviewed the literature and concluded that more

research was needed on the long-term consequences and prognostic importance of anaemia as well as the impact of HAART [14].

We examined the value of anaemia as a prognostic marker in antiretroviral-naïve HIV-infected patients starting HAART, compared to other prognostic factors, and the prognostic importance of the haematological response to HAART using data from the ART Cohort Collaboration.

Patients and methods

The ART Cohort Collaboration

The Antiretroviral Therapy Cohort Collaboration (ART-CC, www.art-cohort-collaboration.org) has been described in detail elsewhere [5;15]. Prospective cohort studies were eligible to participate in the collaboration if they had enrolled at least 100 HIV-1 infected patients aged 16 years who had not previously received antiretroviral treatment and who had started antiretroviral therapy with a combination of at least three drugs, including nucleoside reverse transcriptase inhibitors (NRTIs), protease inhibitors (PIs), and/or non-nucleoside reverse transcriptase inhibitors (NNRTIs). Patients with HIV-1 RNA <1000 copies/ml at initiation of therapy were excluded since they might not have been treatment naïve. All cohorts provided anonymized data on a predefined set of demographic, laboratory and clinical variables, which were then pooled and analyzed centrally. Data included prognostic factors at the time of starting HAART (baseline measurements) and, where available, the values of CD4 cell count, HIV-1 RNA and haemoglobin that were measured at the nearest time-point to six months (within a window from 3 to 9 months) after starting HAART (6-month measurements).

Cohorts included in present analysis

The ART-CC dataset analyzed here includes data up to 2004, with the earliest date of starting HAART ranging from May 3, 1995 to March 12, 1997, and the last recorded clinic visit ranging from March 28, 2002 to August 11, 2003. Data are from clinical cohorts with follow-up during routine care, and visit schedules typically ranging from 3–6 months. Of 12 cohorts that contributed data, one did not supply haemoglobin measurements, while in another cohort haemoglobin values were not regularly recorded. Data from these two cohorts were omitted from all analyses. For one cohort, haemoglobin was only recorded regularly for a subset of patients from certain clinics; therefore only that part of the data was used. The analyses presented here are based on data from the following 10 cohorts: the AIDS Therapy Evaluation project, Netherlands (ATHENA), [16] the ANRS Co3 Aquitaine Cohort, [17] the British Columbia Centre for Excellence in HIV, [2] Collaborations in HIV Outcomes Research (CHORUS), [18] the French Hospital Database on HIV (FHDH), [19] the Italian Cohort of Antiretroviral-naïve patients (ICONA), [20] the Cologne/Bonn Cohort, [21] the Royal Free Hospital Cohort, [22] the South Alberta Clinic [23] and the Swiss HIV Cohort Study (SHCS) [1].

Definitions of anaemia and outcomes

We used a modified World Health Organisation (WHO) definition of anaemia [24], which included an intermediate category for moderate anaemia in order to examine the dose-response relationship in more detail. Definitions for females were severe anaemia <8.0 g/dl, moderate anaemia 8.0 to <10.0 g/dl, mild anaemia 10.0 to <12.0 g/dl, and for males severe <8 g/dl, moderate 8.0 to <11.0 g/dl, mild 11.0 to <13.0 g/dl. We considered progression to AIDS (based on the clinical part of the 1993 US Centers for Disease Control and Prevention revision of the AIDS case definition) [25] as well as progression to death from all causes.

Statistical methods

We used linear regression to examine the effect of initial HAART regimen on 6-month haemoglobin levels, adjusting for baseline haemoglobin, age, sex, disease stage and presumed transmission via injection drug use (IDU). We compared zidovudine (AZT) containing regimens with other regimens and examined whether there were differences between AZT combined with lamivudine (3TC) and AZT combined with didanosine (DDI).

We used Weibull proportional hazards models, stratified by cohort, to estimate hazard ratios for the effect of prognostic factors. The following were candidate prognostic variables: anaemia (categorized as described), CD4 cell count (categorized as <50, 50–99, 100–199, 200–350 and > 350 cells/μl), HIV-1 plasma RNA (categorized as < 4, 4–5 and >5 log10 copies/ml), sex, age (<50 or >=50 years), CDC disease stage, presumed transmission groups (injection drug use or other) and HAART regimen including AZT. Interactions between anaemia, sex and CD4 count were examined. To assess whether haemoglobin could be suitably modelled as a continuous variable, we divided haemoglobin into seven equally sized groups (septiles) and fitted one model using these groups as a categorical variable and one as a linear variable, modelling the linear relationship with the median in each group. Likelihood ratio tests were performed to assess departures from linearity.

We measured time from the start of HAART to the date the endpoints occurred. The same censoring strategy was used for all cohorts: for AIDS, follow-up was censored on the date of the most recent follow-up visit, for mortality on the date the patient was last known to be alive. Patients were deemed lost to follow-up if their last clinic visit was more than one year before the date of the last record in that cohort. In the model for prognosis from start of treatment, only baseline measurements were included. Models examining prognosis following initial response to therapy were based on delayed entry, with follow-up time starting six months after starting HAART or the date of the 6 month post-HAART measurements, whichever was later. In these models we examined the prognostic value of change in anaemia at baseline to 6 months while adjusting for other prognostic variables at baseline and 6 months. Anaemia was re-categorized to none, mild and moderate/severe at baseline and 6 months to produce 9 possible categories for change in anaemia.

Sensitivity analyses

We repeated analyses of the change in anaemia status from baseline to six months excluding patients who did not reach viral load <500 copies/ml by 6 months. We also conducted sensitivity analyses using multiple imputation [26] to deal with missing data. We derived 20

imputed datasets. Estimates from the imputed datasets were combined based on Rubin's rules [26;27].

In all analyses, we used an "intent-to-continue-treatment" approach and thus ignored changes to treatment regimen, including treatment interruptions and terminations. All analyses were carried out using Stata version 9.0 (StataCorp, College Station, TX, USA). Results are presented as hazard ratios (HR) or medians, with 95% confidence intervals (95% CI) or interquartile ranges (IQR).

Results

The combined dataset included data on 16,235 patients, of whom 12,100 (75%) had measurements of haemoglobin at the time they started HAART. Patients without baseline haemoglobin values were more likely to have had AIDS before starting HAART (26% compared with 23%), had higher median CD4 cell counts (239 cells/μl compared with 228 cells/μl) and were less likely to have presumed mode of transmission via IDU (13% vs. 19%). Of these 12,100 patients, 3,389 (28%) were lost to follow-up; varying from 5% to 75% across cohorts.

Table 1 shows the characteristics of 12,100 patients with measurements of haemoglobin at baseline. The median age was 36 years, and a quarter of patients were women. Heterosexual contact was the most frequent risk factor for HIV transmission. The median baseline CD4 cell count was 228 cells/μl and 23% of patients had AIDS. The median haemoglobin concentration was 13.4 g/dl (men 13.6 g/dl, women 11.9 g/dl). A total of 4,222 (35%) patients were anaemic: 2,971 (25%) had mild, 1,120 (9%) had moderate and 131 (1%) had severe anaemia. The prevalence of anaemia was higher in women than in men, higher in older patients than in younger patients, and higher in patients with AIDS compared to patients free of AIDS. The prevalence increased with decreasing CD4 cell counts (see webtable 1).

Prognostic value of baseline haemoglobin

During 48,420 person-years of follow-up, 1,448 patients developed at least one AIDS event and 857 patients died. In Kaplan-Meier plots there was a strong and graded association of baseline anaemia with rates of progression to both AIDS and death (figure 1). A graded association with progression to both AIDS and death continued to be observed in multivariable analyses adjusted for CD4 cell count, viral load and other prognostic variables (table 2). HRs for AIDS were 2.24 (95% CI 1.46 to 3.41) for severe anaemia compared with no anaemia and 2.00 (1.66 to 2.40) for moderate anaemia compared with no anaemia. Corresponding HRs for death were 5.26 (3.55 to 7.81) and 2.56 (2.07 to 3.18) respectively. There was little evidence of interactions of the effects of anaemia with sex or with CD4 cell counts (p=0.25 and 0.13 respectively for AIDS, p=0.80 and p=0.95 for death). Results were similar in analyses based on 16,235 patients, allowing for missing haemoglobin measurements using multiple imputation (data available from the authors). The analysis of septiles defined by haemoglobin levels indicated that the relation between haemoglobin level and risk of progression to AIDS or death was not linear (p=0.042 in men and p=0.015

in women). Plots of hazard ratios (webfigure 1) indicated that the risk increased when mild anaemia occurred.

Changes in haemoglobin after starting HAART

A total of 14,959 patients were followed up for longer than 6 months (175 patients died and 1101 patients had their last follow-up visit in the first 6 months after starting HAART). 9,096 (61%) of the patients followed for longer than 6 months had complete data on all variables measured at baseline and at 6 months. Table 3 shows the distribution of these patients across categories of anaemia at baseline and 6 months. The prevalence of any type of anaemia declined from 35% to 26% (p<0.0001). The median difference in haemoglobin levels between baseline and six months was 0.2 g/dl (interquartile range −0.6 to 1.1 g/dl). It was 0.0 g/dl (interquartile range −0.8 to 0.9 g/dl) in patients whose regimen included AZT, 0.0 g/dl (−0.8 to 0.9 g/dl) in patients on AZT + 3TC and 0.0 g/dl (−0.8 to 0.7 g/dl) in patients on AZT + DDI.

Prognostic value of anaemia at baseline and 6 months of HAART

We examined rates of progression to AIDS and death from 6 months, treating patients with no anaemia both at baseline and 6 months as the reference group (table 4). In general, improvement in anaemia during the first months on HAART was associated with lower subsequent rates of AIDS and death compared with patients whose anaemia status does not improve. However, patients who started HAART with moderate or severe anaemia at baseline continued to experience higher rates of AIDS and death than those with no anaemia at baseline. Patients with no anaemia at baseline and moderate/severe anaemia at 6 months experienced substantially higher mortality rates (hazard ratio 3.93 compared with patients with no anaemia at either time). The risk of AIDS was not elevated in this group (hazard ratio 1.00), though the confidence interval for this comparison was wide (0.43 to 2.33). Sensitivity analyses based on 6,456 patients who suppressed HIV-1 viral replication to <500 copies/ml at 6 months showed stronger associations (webtable 2). Finally, hazard ratios were somewhat higher in analyses based on 14,959 patients, allowing for missing haemoglobin measurements using multiple imputation (data available from the authors).

Discussion

In this large collaborative study, we found that about a third of HIV-1 infected patients who started HAART in European or North American settings were anaemic. Our results show that there is a strong and graded association of the severity of anaemia with rates of progression to AIDS and death, both at the time of starting HAART and after the first six months of therapy. This association was independent of other prognostic factors, including CD4 cell count, an AIDS diagnosis before starting HAART and transmission risk group. An increased risk of progression continued to be observed in patients who were anaemic at baseline but reached normal or near-normal haemoglobin concentrations within the first six months of HAART.

Strengths and weaknesses

The database of the ART Cohort Collaboration includes patients who were treated in many different clinics in Europe and North America. The range of patients was broad: men and women, from teenagers to elderly people were included, and the major exposure categories were well represented. Our findings should therefore be generalisable across different settings and patient groups in industrialized countries. The large number of patients included, and consequently large numbers of AIDS events and deaths, meant that we were able to estimate associations with precision, and examine the nature of the relationship between haemoglobin concentrations and outcomes in detail.

All patients were treatment naïve: baseline haemoglobin levels were therefore not affected by antiretroviral drugs. Our analyses did not, however, take other drugs into account that cause anaemia in HIV-infected patients, for example antiviral agents such as ganciclovir, antifungal agents or drugs used in the chemotherapy of malignancies [14;28]. In patients co-infected with Hepatitis C virus, treatment with ribavirine and interferon is associated with anaemia [29]. At present, the collaborative database does not include information on drugs other than antiretroviral treatments, or on HCV co-infection. Another limitation is the absence of information on ethnicity: several studies found a higher prevalence of anaemia in HIV-1 patients of African American ethnicity than in other races [13;30;31].

Results in context with other studies

In accordance with a recent systematic review of the literature [32], the prevalence of anaemia in our study population varied, depending on sex, age and clinical stage. Direct comparisons with other studies are difficult, because of differences in study populations, presentation of data and definitions used for anaemia. We used the definition that was proposed by a WHO expert committee nearly 40 years ago [24]. Based on NHANES III (the third US National Health and Nutrition Examination Survey) and the Scripps-Kaiser database, these limits should be corrected upwards for white populations in the USA: to 13.7 g/dl in men aged 20 to 59 years and to 12.2 g/dl in women of the same age group [33]. However, our study included many individuals from populations known to have lower average haemoglobin concentrations [33], including many patients form Southern Europe, as well as African American and African patients.

Our results confirm the association of anaemia with poorer outcomes that was observed in previous studies in North America and Europe [7–13]. These studies were based on data from the pre-HAART era, or on patients who had been exposed to antiretroviral monotherapy or dual therapy, and in particular zidovudine, before starting HAART. Of note, in many of these studies results were expressed as the increase in the risk of death per 1 g/dl reduction in haemoglobin, with estimates ranging from a 14% to a 57% increase in risk [28]. We found that the relation between disease progression and levels of haemoglobin is not linear, and this variation in risk estimates may thus to some extent reflect differences in distributions of haemoglobin levels across studies.

The increased risk of anaemia associated with zidovudine has been well documented in the pre-HAART era [34;35]. Our results indicate that the detrimental effect of zidovudine on

haemoglobin levels may be more modest in the context of HAART: the difference in haemoglobin levels at six months was 0.2 g/dl. A recent analysis from the Swiss HIV Cohort Study showed that patients on zidovudine-containing HAART had haemoglobin levels that were 0.5 to 0.8 g/dl lower than in patients not receiving zidovudine up to four years after starting HAART [36]. A retrospective chart review at Toronto Hospital indicated that the combination of AZT with 3TC may increase the risk of anaemia [37], but this was not observed in the present study.

Mechanisms

Anaemia in HIV-infected patients is related to anaemia of chronic disease, infection of auxiliary cells in the bone marrow with HIV and other bone marrow infections, neoplasms infiltrating the bone marrow, malabsorbtion and myelosuppressive drugs [14;28;38]. The Adult and Adolescent Spectrum of HIV Disease Surveillance Project found that 22% of anaemia diagnoses recorded 1990 to 1996 were drug-related [8]. In patients successfully treated with HAART, the suppression of HIV replication and decline in opportunistic infections and neoplasms probably means that antiretroviral drugs will become a relatively more important contributor to anaemia [28].

In resource-constrained settings, where 90% of people with HIV/AIDS live, anaemia related to causes other than antiretroviral drugs will continue to dominate in the coming years. A study in Malawi found that most patients admitted to hospital with severe anaemia had HIV infection, but two thirds also had another treatable condition likely to contribute to anaemia, most commonly tuberculosis or bloodstream infections [39]. Although HAART is effective in these settings [40] and access has improved in recent years, only a minority of patients in urgent need of antiretroviral treatment receive it at present [41].

The causal pathways linking anaemia with poor outcomes are not fully understood. It seems unlikely that anaemia directly leads to HIV associated disease progression. Rather, anaemia may be a proxy measure for more advanced illness. The association with worse prognosis that is observed for other chronic diseases, for example cancer [42], supports this notion. Interestingly, a recent analysis of the Royal Free cohort found that low serum albumin levels predict disease progression and death, independently of haemoglobin concentration [43].

Implications and conclusions

Accurate information on prognosis is of obvious importance to patients and their carers. It is also required to inform decisions on when to start HAART: such decisions should be based on estimates of progression with and without therapy that take several prognostic factors into account, and should not be based only on the CD4 cell count. We will revise the ART Cohort Collaboration's prognostic model to include haemoglobin and thus hope to provide more accurate estimates in the future. We emphasize that neither the present study or an improved prognostic model can determine the optimum time for starting therapy. Specific studies are needed to answer this question. Further research is also required to elucidate the mechanisms underlying the prognostic importance of anaemia. Finally, research is needed to clarify the place of interventions specifically aimed at correcting anaemia in HIV-1 infected patients, including the role of epoietin alfa [44].
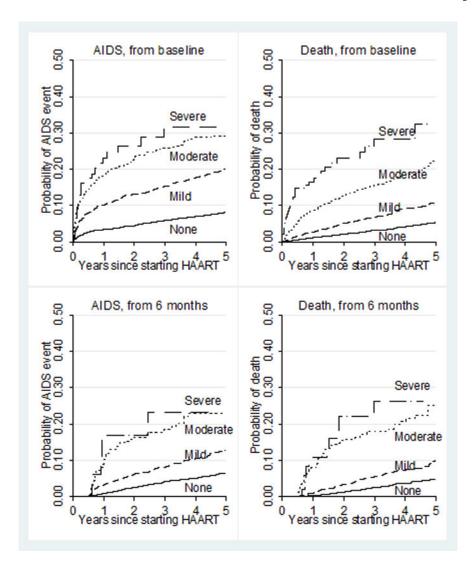
Acknowledgments

Reference List

1. Egger M, Hirschel B, Francioli P, Sudre P, Wirz M, Flepp M, et al. Impact of new antiretroviral combination therapies in HIV infected patients in Switzerland: prospective multicentre study. BMJ. 1997; 315:1194–9. [PubMed: 9393221]

2. Hogg RS, Yip B, Kully C, Craib KJ, O'Shaughnessy MV, Schechter MT, et al. Improved survival among HIV-infected patients after initiation of triple-drug antiretroviral regimens. CMAJ. 1999; 160:659–65. [PubMed: 10102000]

3. Mocroft A, Ledergerber B, Katlama C, Kirk O, Reiss P, d'Arminio MA, et al. Decline in the AIDS and death rates in the EuroSIDA study: an observational study. Lancet. 2003; 362:22–9. [PubMed: 12853195]

4. Sterne JA, Hernan MA, Ledergerber B, Tilling K, Weber R, Sendi P, et al. Long-term effectiveness of potent antiretroviral therapy in preventing AIDS and death: a prospective cohort study. Lancet. 2005; 366:378–84. [PubMed: 16054937]

5. Egger M, May M, Chêne G, Phillips AN, Ledergerber B, Dabis F, et al. Prognosis of HIV-1-infected patients starting highly active antiretroviral therapy: a collaborative analysis of prospective studies. Lancet. 2002; 360:119–29. [PubMed: 12126821]

6. May M, Sterne JA, Sabin C, Costagliola D, Justice AC, Thiebaut R, et al. Prognosis of HIV-1-infected patients up to 5 years after initiation of HAART: collaborative analysis of prospective studies. AIDS. 2007; 21:1185–97. [PubMed: 17502729]

7. Saah AJ, Hoover DR, He Y, Kingsley LA, Phair JP. Factors influencing survival after AIDS: report from the Multicenter AIDS Cohort Study (MACS). J Acquir Immune Defic Syndr. 1994; 7:287–95. [PubMed: 8106968]

8. Sullivan PS, Hanson DL, Chu SY, Jones JL, Ward JW. Epidemiology of anemia in human immunodeficiency virus (HIV)-infected persons: results from the multistate adult and adolescent spectrum of HIV disease surveillance project. Blood. 1998; 91:301–8. [PubMed: 9414298]

9. Moore RD, Keruly JC, Chaisson RE. Anemia and survival in HIV infection. J Acquir Immune Defic Syndr Hum Retrovirol. 1998; 19:29–33. [PubMed: 9732065]

10. Mocroft A, Kirk O, Barton SE, Dietrich M, Proenca R, Colebunders R, et al. Anaemia is an independent predictive marker for clinical prognosis in HIV-infected patients from across Europe. EuroSIDA study group. AIDS. 1999; 13:943–50. [PubMed: 10371175]

11. Thiebaut R, Malvy D, Marimoutou C, Dabis F. Anthropometric indices as predictors of survival in AIDS adults. Aquitaine Cohort, France, 1985–1997. Groupe d'Epidemiologie Clinique du Sida en Aquitaine (GECSA). Eur J Epidemiol. 2000; 16:633–9. [PubMed: 11078120]

12. Lundgren JD, Mocroft A, Gatell JM, Ledergerber B, d'Arminio MA, Hermans P, et al. A clinically prognostic scoring system for patients receiving highly active antiretroviral therapy: results from the EuroSIDA study. J Infect Dis. 2002; 185:178–87. [PubMed: 11807691]

13. Semba RD, Shah N, Klein RS, Mayer KH, Schuman P, Vlahov D. Prevalence and cumulative incidence of and risk factors for anemia in a multicenter cohort study of human immunodeficiency virus-infected and -uninfected women. Clin Infect Dis. 2002; 34:260–6. [PubMed: 11740716]

14. Volberding PA, Levine AM, Dieterich D, Mildvan D, Mitsuyasu R, Saag M. Anemia in HIV infection: clinical impact and evidence-based management strategies. Clin Infect Dis. 2004; 38:1454–63. [PubMed: 15156485]

15. May M, Royston P, Egger M, Justice AC, Sterne JA. Development and validation of a prognostic model for survival time data: application to prognosis of HIV positive patients treated with antiretroviral therapy. Stat Med. 2004; 23:2375–98. [PubMed: 15273954]
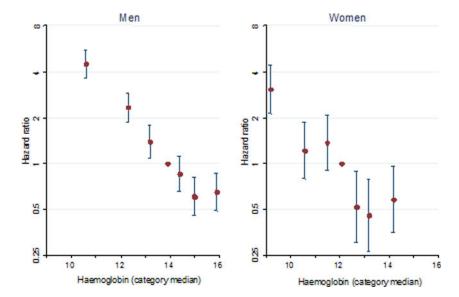
16. Nieuwkerk PT, Sprangers MA, Burger DM, Hoetelmans RM, Hugen PW, Danner SA, et al. Limited patient adherence to highly active antiretroviral therapy for HIV-1 infection in an observational cohort study. Arch Intern Med. 2001; 161:1962–8. [PubMed: 11525698]

17. Binquet C, Chene G, Jacqmin-Gadda H, Journot VV, Saves M, Lacoste D, et al. Modeling Changes in CD4-positive T-Lymphocyte Counts after the Start of Highly Active Antiretroviral Therapy and the Relation with Risk of Opportunistic Infections The Aquitaine Cohort, 1996–1997. Am J Epidemiol. 2001; 153:386–93. [PubMed: 11207157]

18. Becker SL, Raffanti SR, Hansen NI, Fusco JS, Fusco GP, Slatko GH, et al. Zidovudine and stavudine sequencing in HIV treatment planning: findings from the CHORUS HIV cohort. J Acquir Immune Defic Syndr. 2001; 26:72–81. [PubMed: 11176271]

19. Grabar S, Le Moing V, Goujard C, Leport C, Kazatchkine MD, Costagliola D, et al. Clinical outcome of patients with HIV-1 infection according to immunologic and virologic response after 6 months of highly active antiretroviral therapy. Ann Intern Med. 2000; 133:401–10. [PubMed: 10975957]

20. d'Arminio MA, Lepri AC, Rezza G, Pezzotti P, Antinori A, Phillips AN, et al. Insights into the reasons for discontinuation of the first highly active antiretroviral therapy (HAART) regimen in a cohort of antiretroviral naive patients. I.CO.N.A. Study Group. Italian Cohort of Antiretroviral-Naive Patients. AIDS. 2000; 14:499–507. [PubMed: 10780712]

21. Fatkenheuer G, Theisen A, Rockstroh J, Grabow T, Wicke C, Becker K, et al. Virological treatment failure of protease inhibitor therapy in an unselected cohort of HIV-infected patients. AIDS. 1997; 11:F113–F116. [PubMed: 9386799]

22. Mocroft A, Barry S, Sabin CA, Lepri AC, Kinloch S, Drinkwater A, et al. The changing pattern of admissions to a London hospital of patients with HIV: 1988–1997. Royal Free Centre for HIV Medicine. AIDS. 1999; 13:1255–61. [PubMed: 10416531]

23. Mocroft A, Gill MJ, Davidson W, Phillips AN. Predictors of a viral response and subsequent virological treatment failure in patients with HIV starting a protease inhibitor. AIDS. 1998; 12:2161–7. [PubMed: 9833857]

24. World Health Organization. Nutritional Anaemias: Report of a WHO Scientific Group. WHO Tech Rep Ser. 1968; 405:1–40.

25. Centers for Disease Control. 1993 revised classification system for HIV infection and expanded surveillance case definition for AIDS among adolescents and adults. MMWR. 1992; 41(RR-17):1–20.

26. Rubin, DB. Multiple Imputation for Nonresponse in Surveys. New York: John Wiley; 1987.

27. Royston P. Multiple imputation of missing values. Stata Journal. 2004; 4:241.

28. Moyle G. Anaemia in persons with HIV infection: prognostic marker and contributor to morbidity. AIDS Rev. 2002; 4:13–20. [PubMed: 11998779]

29. Hadziyannis SJ, Sette H Jr, Morgan TR, Balan V, Diago M, Marcellin P, et al. Peginterferon-alpha2a and ribavirin combination therapy in chronic hepatitis C: a randomized study of treatment duration and ribavirin dose. Ann Intern Med. 2004; 140:346–55. [PubMed: 14996676]

30. Levine AM, Berhane K, Masri-Lavine L, Sanchez M, Young M, Augenbraun M, et al. Prevalence and correlates of anemia in a large cohort of HIV-infected women: Women's Interagency HIV Study. J Acquir Immune Defic Syndr. 2001; 26:28–35. [PubMed: 11176266]

31. Creagh T, Mildvan D, Leitz G. Prevalence of anemia and correlation with biomarkers and specific antiretroviral regimens in 9690 human-immunodeficiency-virus-infected patients: findings of the Anemia Prevalence Study. Current Medical Research & Opinion. 2007; 23:343–55. [PubMed: 17288689]

32. Belperio PS, Rhew DC. Prevalence and outcomes of anemia in individuals with human immunodeficiency virus: a systematic review of the literature. Am J Med. 2004; 116(Suppl 7A): 27S–43S. [PubMed: 15050884]

33. Beutler E, Waalen J. The definition of anemia: what is the lower limit of normal of the blood hemoglobin concentration? Blood. 2006; 107:1747–50. [PubMed: 16189263]

34. Richman DD, Fischl MA, Grieco MH, Gottlieb MS, Volberding PA, Laskin OL, et al. The toxicity of azidothymidine (AZT) in the treatment of patients with AIDS and AIDS-related complex. A double-blind, placebo-controlled trial. N Engl J Med. 1987; 317:192–7. [PubMed: 3299090]

35. Volberding PA, Lagakos SW, Koch MA, Pettinelli C, Myers MW, Booth DK, et al. Zidovudine in asymptomatic human immunodeficiency virus infection. A controlled trial in persons with fewer than 500 CD4-positive cells per cubic millimeter. The AIDS Clinical Trials Group of the National Institute of Allergy and Infectious Diseases. N Engl J Med. 1990; 322:941–9. [PubMed: 1969115]

36. Huttner AC, Kaufmann GR, Battegay M, Weber R, Opravil M. Treatment initiation with zidovudine-containing potent antiretroviral therapy impairs CD4 cell count recovery but not clinical efficacy. AIDS. 2007; 21:939–46. [PubMed: 17457087]

37. Tseng A, Conly J, Fletcher D, Keystone D, Salit I, Walmsley S. Precipitous declines in hemoglobin levels associated with combination zidovudine and lamivudine therapy. Clin Infect Dis. 1998; 27:908–9. [PubMed: 9798060]

38. Moses A, Nelson J, Bagby GC Jr. The influence of human immunodeficiency virus-1 on hematopoiesis. Blood. 1998; 91:1479–95. [PubMed: 9473211]

39. Lewis DK, Whitty CJ, Walsh AL, Epino H, Broek NR, Letsky EA, et al. Treatable factors associated with severe anaemia in adults admitted to medical wards in Blantyre, Malawi, an area of high HIV seroprevalence. Trans R Soc Trop Med Hyg. 2005; 99:561–7. [PubMed: 15893781]

40. Braitstein P, Brinkhof MW, Dabis F, Schechter M, Boulle A, Miotti P, et al. Mortality of HIV-1-infected patients in the first year of antiretroviral therapy: comparison between low-income and high-income countries. Lancet. 2006; 367:817–24. [PubMed: 16530575]

41. World Health Organization. Progress report 2008. Geneva: World Health Organization, UNAIDS, Unicef; 2008. Towards universal access. Scaling up priority HIV/AIDS interventions in the health sector. Ref Type: Report

42. Caro JJ, Salas M, Ward A, Goss G. Anemia as an independent prognostic factor for survival in patients with cancer: a systemic, quantitative review. Cancer. 2001; 91:2214–21. [PubMed: 11413508]

43. Shah S, Smith CJ, Lampe F, Youle M, Johnson MA, Phillips AN, et al. Haemoglobin and albumin as markers of HIV disease progression in the highly active antiretroviral therapy era: relationships with gender. HIV Med. 2007; 8:38–45. [PubMed: 17305931]

44. Henry DH, Volberding PA, Leitz G. Epoetin alfa for treatment of anemia in HIV-infected patients: past, present, and future. J Acquir Immune Defic Syndr. 2004; 37:1221–7. [PubMed: 15385728]

Figure 1.
Kaplan Meier estimates of the cumulative probability of AIDS (left panels) and death (right panels) according to anaemia at baseline (upper panels) or anaemia at 6 months (lower panels).

See table 1 for definitions of mild, moderate and severe anaemia.

Webfigure 1.

Hazard ratios for progression to AIDS or death by septiles of haemoglobin levels.

Median (range) haemoglobin levels in men were: septile 1: 10.6 g/dl (4.1 to 11.6 g/dl); septile 2: 12.3 g/dl (11.7 to 12.8 g/dl); septile 3: 13.2 g/dl (12.9 to 13.5 g/dl); septile 4: 13.9 g/dl (13.6 to 14.1 g/dl); septile 5: 14.4 g/dl (14.2 to 14.7 g/dl); septile 6: 15.0 g/dl (14.7 to 15.3 g/dl); septile 7: 15.9 g/dl (15.4 to 22.6 g/dl).

Median (range) haemoglobin levels in women were: septile 1: 9.2 g/dl (2.1 to 10.0 g/dl); septile 2: 10.6 g/dl (10.1 to 11.0 g/dl); septile 3: 11.5 g/dl (11.1 to 11.8 g/dl); septile 4: 12.1 g/dl (11.8 to 12.4 g/dl); septile 5: 12.7 g/dl (12.4 to 12.9 g/dl); septile 6: 13.2 g/dl (13.0 to 13.6 g/dl); septile 7: 14.2 g/dl (13.7 to 18.0 g/dl).

Table 1

Baseline characteristics of 12,100 patients with haemoglobin measurements at the time of starting HAART.

Age (years)	Median (IQR)	36 (31 to 42)
	>50	1,219 (10%)
Sex	Female	3,138 (25%)
Presumed mode of transmission	Male homosexual contact	4,263 (35%)
	Heterosexual contact	4,581 (38%)
	Injection-drug use	2,262 (19%)
	Other or unknown	994 (8%)
Clinical AIDS		2,737 (23%)
CD4 count (cells/μl)	Median (IQR)	228 (93–372)
	> 350	3,422 (28%)
	200–349	3,236 (27%)
	100–199	2,244 (19%)
	50–99	1,213 (10%)
	<50	1,965 (16%)
Plasma HIV-1 RNA ($\log_{10}$ copies/ml)	Median (IQR)	4.9 (4.4–5.4)
	< 4	1,423 (12%)
	4 – 5	5,168 (43%)
	> 5	5,509 (46%)
Haemoglobin (g/dl)	Median (IQR)	13.4 (11.9 to 14.6)
Anaemia[*]	Mild	2,971 (25%)
	Moderate	1,120 (9%)
	Severe	131 (1%)
Initial HAART regimen	NNRTI-based	3,087 (26%)
	PI-based	8,108 (67%)
Year of initiation of HAART	1995–6	584 (5%)
	1997	2,659 (22%)
	1998	2,681 (22%)
	1999	1,994 (16%)
	2000	1,775 (15%)
	2001	1,290 (11%)
	2002–3	1,117 (9%)
Duration of follow-up (years)	Median (IQR)	2.9 (1.41 to 4.43)
	< 2	4,066 (34%)
	2–4	6,068 (50%)
	>5	1,966 (16%)

IQR, interquartile range

[*] Mild – males: 11.0 to <13.0 g/dl, females: 10.0 to <12 g/dl; moderate – males: 8.0 to <11.0 g/dl, females: 8.0 to <10.0 g/dl; severe – males and females: < 8.0 g/dl.

Table 2

Hazard ratios for AIDS and death for anaemia and other prognostic factors measured at initiation of HAART.

	Adjusted		Unadjusted	
	AIDS	**Death**	**AIDS**	**Death**
Anaemia				
None	1	1	1	1
Mild	1.60 (1.37–1.86)	1.42 (1.17–1.73)	2.81 (2.45, 3.23)	2.09 (1.74, 2.51)
Moderate	2.00 (1.66–2.40)	2.56 (2.07–3.18)	5.28 (4.46, 6.24)	5.08 (4.19, 6.15)
Severe	2.24 (1.46–3.42)	5.26 (3.55–7.81)	6.10 (4.04, 9.21)	10.22 (7.03, 14.84)
CD4 count (cells/μl)				
>350	1	1	1	1
200–349	1.50 (1.16–1.92)	1.40 (1.06–1.85)	1.68 (1.31, 2.16)	1.62 (1.23, 2.13)
100–199	2.38 (1.86–3.05)	1.71 (1.28–2.27)	3.52 (2.78, 4.47)	2.69 (2.06, 3.53)
50–99	3.90 (3.01–5.04)	1.94 (1.42–2.65)	7.37 (5.79, 9.37)	3.83 (2.88, 5.09)
<50	4.87 (3.81–6.24)	2.30 (1.72–3.07)	11.81 (9.51, 14.67)	5.62 (4.40, 7.18)
Plasma viral load ($\log_{10}$ copies/ml)				
3 – 4	1	1	1	1
4 – 5	1.16 (0.88–1.52)	0.81 (0.62–1.07)	1.35 (1.03, 1.78)	0.87 (0.66, 1.14)
> 5	1.46 (1.12–1.91)	0.81 (0.62–1.07)	3.13 (2.41, 4.06)	1.53 (1.18, 1.98)
Other risk factors				
Female	0.79 (0.68–0.92)	0.77 (0.63–0.93)	0.81 (0.70, 0.94)	0.77 (0.64, 0.93)
Age > 50	1.12 (0.94–1.35)	1.96 (1.60–2.39)	1.32 (1.10, 1.57)	1.92 (1.59, 2.32)
CDC stage C	2.92 (2.49–3.41)	2.14 (1.79–2.56)	6.73 (5.85, 7.73)	3.90 (3.35, 4.54)
Presumed transmission from IDU	1.44 (1.23–1.69)	2.44 (2.05–2.91)	1.40 (1.20, 1.64)	2.09 (1.77, 2.48)

Analysis based on 12,100 individuals with complete data on all variables. *Hazard ratios adjusted for other variables in the table and zidovudine (AZT) in initial HAART regimen, stratified by year of starting HAART and cohort

CDC, Centers for Disease Control and Prevention; IDU, injection drug use

See table 1 for definitions of mild, moderate and severe anaemia.

Table 3

Distribution of patients across categories of anaemia at baseline and 6 months after starting HAART.

Anaemia status at baseline	Anaemia status at 6 months				
	None	Mild	Moderate	Severe	Total
None	5508 (86.8%)	763 (12.0%)	59 (0.9%)	15 (0.2%)	6345 (66.3%)
Mild	1274 (54.9%)	910 (39.2%)	126 (5.4%)	12 (0.5%)	2322 (24.3%)
Moderate	316 (38.5%)	346 (39.2%)	148 (18.0%)	10 (1.2%)	820 (8.6%)
Severe	20 (25.0%)	30 (37.5%)	25 (31.3%)	5 (6.7%)	80 (0.8%)
Total	7118 (74.4%)	2049 (21.4%)	358 (3.7%)	42 (0.4%)	9,567 (100%)

See table 1 for definitions of mild, moderate and severe anaemia.

Table 4

Hazard ratios for AIDS and death for change in anaemia status from baseline to six months after starting HAART, adjusting for other factors.

Anaemia status at baseline	Anaemia status at 6 months	Number of patients	AIDS	Death
None	None	5358	1 (ref)	1 (ref)
Mild	None	1194	0.98 (0.74, 1.31)	0.85 (0.61, 1.20)
Moderate/severe	None	297	1.37 (0.90, 2.09)	1.52 (0.99, 2.34)
None	Mild	723	1.20 (0.82, 1.77)	1.30 (0.88, 1.91)
Mild	Mild	857	1.55 (1.17, 2.06)	1.49 (1.08, 2.06)
Moderate/severe	Mild	339	1.84 (1.28, 2.65)	1.89 (1.31, 2.73)
None	Moderate/severe	61	1.00 (0.43, 2.33)	3.93 (2.18, 7.07)
Mild	Moderate/severe	112	2.51 (1.50, 4.19)	3.47 (2.23, 5.40)
Moderate/severe	Moderate/severe	155	2.59 (1.60, 4.19)	3.14 (2.06, 4.77)

Analyses based on 9,096 individuals with complete data on all prognostic variables.

Hazard ratios adjusted for CD4 cell count and plasma viral load at baseline and six months, age, sex, CDC stage, AIDS in first six months, presumed HIV transmission injection drug use and regimen including zidovudine (AZT) at baseline, stratified by year of starting HAART and cohort.

See table 1 for definitions of mild, moderate and severe anaemia.

Webtable 1

Prevalence of anaemia among 12,100 patients at the time of starting HAART.

		Anaemia*				
A		**Severe**	**Moderate**	**Mild**	**None**	**Total**
Age (years)	50	1.1%	8.7%	23.8%	66.4%	10,881
	>50	1.1%	13.7%	30.2%	55.0%	1,219
Sex	Female	2.1%	11.3%	33.4%	53.2%	3,138
	Male	0.7%	8.5%	21.5%	69.3%	8,962
Presumed mode of transmission	Male homosexual contact	0.5%	5.8%	19.1%	74.5%	4,263
	Heterosexual contact	1.4%	12.0%	29.6%	57.1%	4,581
	Injection-drug use	1.1%	9.2%	23.3%	66.4%	2,262
	Other or unknown	1.9%	11.8%	27.8%	58.6%	994
AIDS before start of HAART	Yes	0.9%	25.2%	28.8%	45.0%	111
	No	0.4%	4.9%	13.3%	81.4%	285
CD4 count (cells/µl)	> 350	0.3%	1.9%	12.1%	85.8%	3,442
	200–349	0.5%	4.3%	20.9%	74.3%	3,236
	100–199	1.4%	9.8%	28.9%	59.9%	2,244
	50–99	2.1%	17.0%	36.6%	44.4%	1,213
	<50	2.6%	24.9%	40.0%	32.5%	1,965
Plasma HIV-1 RNA ($\log_{10}$ copies/ml)	< 4	1.1%	5.9%	18.6%	74.4%	1,399
	4 – 5	0.7%	6.0%	19.2%	74.2%	5,131
	> 5	1.5%	13.1%	31.0%	54.4%	5,570
Initial HAART regimen	NNRTI-based	1.3%	6.4%	21.7%	70.6%	3,087
	PI-based	1.1%	10.3%	26.1%	62.5%	8,108
Year of initiation of HAART	1995–6	0.7%	9.7%	25.2%	64.3%	575
	1997	0.9%	8.2%	21.3%	69.6%	2,598
	1998	0.9%	8.2%	22.1%	68.9%	2,637
	1999	1.3%	9.9%	23.3%	65.5%	1,955
	2000	0.9%	9.9%	28.9%	60.3%	1,729
	2001	1.8%	9.1%	31.3%	57.7%	1,254

A

		Anaemia[*]				
		Severe	Moderate	Mild	None	Total
Duration of follow-up (years)	2002–3	1.3%	11.5%	25.3%	62.0%	1,352
	< 2	1.8%	11.5%	28.0%	58.7%	4,066
	2–4	0.8%	8.6%	24.1%	66.5%	6,068
	>5	0.4%	6.8%	18.8%	74.0%	1,966

See table 1 for definitions of mild, moderate and severe anaemia.

Webtable 2

Hazard ratios for AIDS and death for change in anaemia status from baseline to six months after starting HAART, adjusting for other factors. Analyses based on 6,456 patients who suppressed HIV-1 viral replication to <500 copies/ml at 6 months.

Anaemia status at baseline	Anaemia status at 6 months	N	AIDS	Death
None	None	3877	1 (ref)	1 (ref)
Mild	None	823	0.88 (0.57, 1.37)	1.08 (0.68, 1.70)
Moderate/severe	None	203	1.13 (0.61, 2.10)	1.96 (1.08, 3.55)
None	Mild	539	1.05 (0.58, 1.90)	1.18 (0.68, 2.05)
Mild	Mild	568	2.14 (1.45, 3.16)	1.54 (0.96, 2.46)
Moderate/severe	Mild	242	2.39 (1.43, 3.99)	2.27 (1.38, 3.75)
None	Moderate/severe	41	0.91 (0.13, 6.57)	8.70 (4.32, 17.50)
Mild	Moderate/severe	67	1.35 (0.42, 4.32)	5.99 (3.26, 11.02)
Moderate/severe	Moderate/severe	96	1.90 (0.89, 4.04)	3.35 (1.73, 6.46)

Hazard ratios adjusted for CD4 cell count and plasma viral load at baseline and six months, age, sex, CDC stage, AIDS in first six months, presumed HIV transmission injection drug use and regimen including zidovudine (AZT) at baseline, stratified by year of starting HAART and cohort.

See table 1 for definitions of mild, moderate and severe anaemia.