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Predicting risk of cancer during HIV infection: the role of inflammatory and coagulation biomarkers

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Abstract

Objective—To investigate the relationship between inflammatory [interleukin-6 (IL-6) and C-reactive protein (CRP)] and coagulation (D-dimer) biomarkers and cancer risk during HIV infection.

Design—A prospective cohort.

Methods—HIV-infected patients on continuous antiretroviral therapy (ART) in the control arms of three randomized trials (N = 5023) were included in an analysis of predictors of cancer (any type, infection-related or infection-unrelated). Hazard ratios for IL-6, CRP and D-dimer levels ($\log_2$-transformed) were calculated using Cox models stratified by trial and adjusted for demographics and $CD4^+$ cell counts and adjusted also for all biomarkers simultaneously. To assess the possibility that biomarker levels were elevated at entry due to undiagnosed cancer,

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analyses were repeated excluding early cancer events (i.e. diagnosed during first 2 years of follow-up).

Results—During approximately 24 000 person-years of follow-up (PYFU), 172 patients developed cancer (70 infection-related; 102 infection-unrelated). The risk of developing cancer was associated with higher levels (per doubling) of IL-6 (hazard ratio 1.38, $P < 0.001$), CRP (hazard ratio 1.16, $P = 0.001$) and D-dimer (hazard ratio 1.17, $P = 0.03$). However, only IL-6 (hazard ratio 1.29, $P = 0.003$) remained associated with cancer risk when all biomarkers were considered simultaneously. Results for infection-related and infection-unrelated cancers were similar to results for any cancer. Hazard ratios excluding 69 early cancer events were 1.31 ($P = 0.007$), 1.14 ($P = 0.02$) and 1.07 ($P = 0.49$) for IL-6, CRP and D-dimer, respectively.

Conclusion—Activated inflammation and coagulation pathways are associated with increased cancer risk during HIV infection. This association was stronger for IL-6 and persisted after excluding early cancer. Trials of interventions may be warranted to assess whether cancer risk can be reduced by lowering IL-6 levels in HIV-positive individuals.

Keywords

biomarkers; cancer; C-reactive protein; D-dimer; HIV; interleukin-6

Introduction

Since the beginning of the HIV pandemic, an increased risk of cancer has been observed in HIV-infected individuals. Some infection-related cancers, namely Kaposi sarcoma, non-Hodgkin lymphoma (NHL) and invasive cervical cancer (ICC), were found to have a particularly high incidence in patients with advanced HIV infection. With the aim of tracking the emerging epidemic, these cancers were included in the case definition of AIDS and, since then, have been classically referred to as AIDS-defining malignancies (ADMs) [1]. Epidemiological surveillance, however, subsequently broadened the spectrum of malignancies associated with HIV infection. HIV-infected individuals have been found to be at a higher risk of an ever-increasing range of both infection-related and infection-unrelated non-AIDS defining malignancies (NADMs) [2–5].

Although many reasons have been postulated, the mechanisms by which HIV infection increases the risk of cancer remain poorly understood. A higher prevalence of traditional cancer risk factors (e.g. smoking, alcohol use, oncogenic virus coinfection) in HIV-positive persons seems to play an important role [6]. Furthermore, clinical data suggest that HIV-associated immunodeficiency may lead to accelerated viral oncogenesis and reduced immune surveillance of malignant cells. Not only infection-related ADM but also infection-unrelated NADM have been shown to occur more frequently in HIV-infected persons, particularly in individuals with lower $CD4^+$ cell counts [4,7–10]. Finally, the scale up of antiretroviral therapy (ART) has greatly improved survival outcomes, and as HIV-infected individuals age, the incidence of cancer is expected to increase [11], as it does in the general population.

Evidence has recently accrued suggesting that activated inflammatory and coagulation pathways may also contribute to cancer risk in HIV-infected individuals. In the Strategies for Management of Antiretroviral Therapy (SMART) study [12], structured interruptions of ART were associated with a significantly higher incidence of cancer [13] and a rise in plasma levels of D-dimer, a fibrin degradation product, and of interlukin-6 (IL-6), an inflammatory cytokine [14]. IL-6 production has been shown to be an important component of autocrine [15] and paracrine [16] circuits that fuel the growth of solid tumours. In the general population, elevated plasma levels of IL-6 and C-reactive protein (CRP), a marker of inflammation whose production by hepatocytes is driven by IL-6 [17], are associated with an increased risk of developing cancer [18–20]. Except for a few reports linking elevated IL-6 levels with the risk of future NHL [21,22], no systematic studies examining the interplay between inflammation, coagulation and cancer in the setting of HIV infection have been carried out.

The purpose of this study is to investigate the relationship between inflammatory (IL-6 and CRP) and coagulation (D-dimer) biomarkers and the risk of cancer during HIV infection. Our main *a priori* hypothesis was that activated inflammation and coagulation, as demonstrated by elevated plasma levels of IL-6, CRP and D-dimer, contribute to the risk of developing both infection-related and infection-unrelated cancer in HIV-infected individuals.

Materials and methods

Study design and study population

This is a cohort study involving participants in the control arms of three randomized controlled trials, who had consented to storing blood for future research and whose plasma levels of IL-6, CRP and D-dimer were measured at study entry ($N = 5023$). The methodology of SMART [12], the Evaluation of Subcutaneous Proleukin in a Randomized International Trial (ESPRIT) and the Subcutaneous Recombinant, Human Interleukin-2 in HIV-Infected Patients with Low $CD4^+$ Counts under Active Antiretroviral Therapy (SILCAAT) [23] trial has been described in detail elsewhere. Briefly, the SMART trial compared, in 5472 individuals with $CD4^+$ cell count more than 350 cells/μl at baseline, continuous use of ART versus structured treatment interruption guided by $CD4^+$ cell count. The ESPRIT and SILCAAT trials compared IL-2 and ART with ART alone in 4111 individuals with $CD4^+$ cell count more than 300 cells/μl and 1695 individuals with $CD4^+$ cell count between 50 and 299 cells/μl, respectively. In the three trials, individuals in the control arms received standard of care according to HIV guidelines and were to be continuously maintained on ART.

Inflammatory and coagulation biomarkers

On the basis of strong associations of CRP, IL-6 and D-dimer with all-cause mortality in a nested case–control study [14] and the observation that these biomarkers were elevated in HIV-infected individuals compared with the general population [24], IL-6, CRP and D-dimer were measured on stored plasma at baseline for all consenting participants in ESPRIT, SILCAAT and SMART. For SMART participants, biomarkers were measured at the

Laboratory for Clinical Biochemistry Research at the University of Vermont (Burlington). In the ESPRIT and SILCAAT trials, laboratory measurements were performed by SAIC-Frederick (Frederick, Maryland, USA).

IL-6 was measured by the same method at each laboratory (Chemiluminescent Sandwich ELISA; R&D Systems, Minneapolis, Minnesota, USA). D-dimer levels were measured by ELISA on the Sta-R analyser, Liatest D-DI (Diagnostic Stago, Parsippany, New Jersey, USA) for SMART participants and on a VIDAS instrument (BioMerieux Inc., Durham, North Carolina, USA) for ESPRIT and SILCAAT participants. These assays, while different, compared very well on 20 duplicate samples. CRP was measured by ELISA by both laboratories. For SMART participants, an NBTMII nephelometer, N Antiserum to Human CRP (Siemens Diagnostics, Deerfield, Illinois, USA) was used. For ESPRIT and SILCAAT participants, an R&D Systems ELISA assay was used. The assays used were different, but as for D-dimer, they compared very well on duplicate samples. Lower limits of detection for IL-6, CRP and D-dimer were 0.16 pg/ml, 0.16 and 0.01 μg/ml for SMART. In ESPRIT and SILCAAT, lower limits of detection were 0.156 pg/ml, 0.078 and 0.045 μg/ml. All samples were analysed blinded to the treatment group and cancer event status.

Follow-up and cancer ascertainment

Patients were followed from study entry until first cancer event, death, loss to follow-up or the closing date of each study, whichever occurred first. Clinical assessment intervals and total follow-up time varied by study. Median follow-up time was 29 months in SMART, 81 months in ESPRIT and 91 months in SILCAAT; overall, median follow-up was 59 months for the entire cohort. In SMART and ESPRIT, all malignancies were systematically reported to and centrally adjudicated by the Endpoint Review Committee (ERC). In SILCAAT, only ADM and Hodgkin lymphoma were centrally adjudicated, whereas other NADMs were identified from the adverse event reporting system. In sensitivity analyses, the exclusion of SILCAAT had no major impact on findings (data not shown).

The following malignancies were considered to be infection-related: ADM, which have viral causes [Kaposi sarcoma, NHL and ICC are related to human herpes virus 8 (HHV-8), Epstein–Barr virus (EBV) and human papillomavirus (HPV), respectively); vagina, vulva, penis, anal and oral cavity/pharynx cancers (HPV); Hodgkin lymphoma (EBV); liver cancer (hepatitis B and C viruses) and stomach cancer (*Helicobacter pylori*) [25]. No pharyngeal lymphoepitheliomas were identified in this study. All other malignancies were considered to be infection-unrelated cancers.

Statistical analyses

All analyses were restricted to patients in the control arms of each study for whom baseline biomarker data were available. Other than the events identified through adverse event reporting, cancer events were included if they were considered ‘confirmed or ‘probable’ by the ERC, or if the patient died and the ERC attributed the underlying cause of death to cancer. Cancers were characterized by type on the basis of the Medical Dictionary for Regulatory Activities (MedDRA) High Level Term (HLT) assigned.

Kaplan–Meier curves giving the cumulative percentage of participants with cancer are shown for biomarker quartiles. To control for any differences in biomarker distributions among studies, quartiles were defined separately for participants in each study. Associations between IL-6, CRP and D-dimer levels at study entry and the risk of the first incident cancer event were estimated using proportional hazards (Cox) models from which hazard ratios corresponding to one $\log_2$ increase in biomarker (i.e. a doubling) and 95% confidence intervals (CIs) were estimated. Biomarkers were $\log_2$-transformed because their distributions were right-skewed. Cox models for each biomarker were stratified by study to account for differences in underlying risk for the different cohorts. This stratification also served to stratify on the two laboratories that measured the biomarkers. Three models were considered: (1) unadjusted; (2) adjusting for age, sex, race, continent of enrolment, study entry and time-updated $CD4^+$ counts; and (3) adjusting for the same covariates and all biomarkers simultaneously.

In order to compare the association of each biomarker with different types of cancer, a competing risk model for multiple unordered events of different types was fit [26]. With this approach, the fit of a model that assumed a common association for each biomarker with infection-related and infection-nonrelated cancers was compared with the fit of a model that allowed the association to vary. A similar approach was taken for assessing the association of the biomarkers with type-specific cancers of interest.

With the proportional hazards model, we assume that the risk of cancer associated with higher versus lower biomarker levels is constant over the follow-up period. This assumption was tested in a model that included an interaction term between each $\log_2$-transformed biomarker and log follow-up time along with the baseline covariates in model (2) above. In addition, two other analyses were carried out: first, we produced graphic displays of log (–log) of the adjusted survival curves for participants with biomarker levels above versus below the median (departure from parallelism of these graphs indicates that the risk of cancer for those with higher versus lower levels is not constant over follow-up); and second, we performed analyses excluding cancer events diagnosed in the first 2 years of follow-up. These analyses were performed because clinically undetected malignancies may lead to raised plasma levels of inflammatory and coagulation biomarkers. In that case, one would expect that risk ratios for cancer events closer to the time of measurement of the biomarkers would differ from those events more remote from the measurement.

In other sensitivity analyses, additional important traditional cancer risk factors were adjusted for obesity/BMI (data available for participants in the three trials); diabetes (SMART and ESPRIT participants) and smoking (SMART only). All statistical analyses were performed using SAS software (version 9.2; SAS Institute, Cary, North Carolina, USA)

Results

A total of 5023 HIV-infected individuals were included. Six hundred and fifteen individuals without biomarker measurements were excluded. Excluded individuals were more likely to be younger, black and female and have lower nadir $CD4^+$ cell counts (data not shown).

During approximately 24 000 persons-years of follow-up, 172 patients developed cancer (70 infection-related; 102 infection-unrelated). The most common infection-unrelated cancers were lung, prostate and colorectal; the most common infection-related cancers were NHL, anal and Hodgkin lymphoma (Table 1). Patients who developed cancer were more likely to be older males, have hepatitis B/C coinfection and diabetes, as well as to smoke. Patients who developed cancer were also found to have higher baseline levels of CRP, D-dimer and IL-6, and lower nadir $CD4^+$ cell counts (Table 2).

Kaplan–Meier curves for any type, infection-related and infection-unrelated cancers are shown for quartiles of each biomarker in Supplementary Figure 1, http://links.lww.com/QAD/A313. When biomarkers were modelled as continuous variables, all three biomarkers demonstrated significant unadjusted associations (model 1) with increased risk of any type, infection-related and infection-unrelated cancers (see Table 3). After adjustment for demographics (age, race, sex and continent) and for study entry and time-updated $CD4^+$ cell counts (model 2), each biomarker remained significantly associated with any type of cancer. When all biomarkers were adjusted for (model 3), only IL-6 remained independently associated with cancer risk (hazard ratio 1.29, 95% CI 1.09–1.52, $P = 0.003$). There was no evidence that biomarker associations varied between infection-related and infection-unrelated cancers. With the model 2 adjustment, P values for differences between the associations with infection-related and infection-unrelated cancers were 0.90, 0.90 and 0.89, for CRP, D-dimer and IL-6, respectively.

Table 4 summarizes the association between inflammatory and coagulation biomarkers and specific types of cancer that occurred in at least 10 patients. On the basis of the adjusted competing risk model summarizing cancers by type (NHL, Hodgkin lymphoma, HPV-related cancers, lung, prostate, colorectal, Kaposi sarcoma and all other types), there was no evidence that biomarker associations varied by type of cancer for D-dimer ($P = 0.12$) or IL-6 ($P = 0.65$); however, there was evidence for unequal effects across cancer types for CRP ($P = 0.04$). Higher baseline plasma levels of CRP were independently associated with risk of developing any type of lymphoma (hazard ratio 1.35, $P = 0.005$), Hodgkin lymphoma (HR = 1.72, $P = 0.004$) and colorectal cancer (hazard ratio 1.41, $P = 0.04$). Elevated IL-6, on the contrary, was found to be statistically associated with risk of lung cancer (hazard ratio 1.62, $P = 0.01$). However, owing to the limited number of cancer events and the consequent wider CI, our estimates are imprecise and need to be interpreted with caution.

Sensitivity analyses

For all cancers and for infection-related and infection-unrelated cancers, there was no evidence that hazard ratios for higher versus lower levels of CRP and D-dimer varied over follow-up; P values testing the proportionality assumption ranged between 0.25 and 0.92. For IL-6, however, the hazard ratios varied across time for all cancers ($P = 0.04$) and infection-unrelated cancers ($P = 0.03$). On the basis of model (2), the hazard ratio for any type of cancer in the first 2 years of follow-up (68 cancer events) associated with a doubling of IL-6 was 1.50 (95% CI 1.21–1.85; $P < 0.001$); for follow-up after 2 years (103 cancer events), the hazard ratio was 1.31 (95% CI 1.08–1.60; $P = 0.007$). For infection-unrelated cancers, the hazard ratios were 1.52 (95% CI 1.17–1.97, $P = 0.002$) and 1.24 (95% CI 0.95–

1.62, $P = 0.12$) for the two time periods. For IL-6, this is graphically illustrated in Supplementary Figure 2, http://links.lww.com/QAD/A313. During the early follow-up period, differences between the quartiles were greater, whereas after 1–2 years, the curves were more parallel.

Data on some important traditional cancer risk factors, namely obesity/BMI, diabetes and smoking, were not uniformly collected in the three trials. Consequently, these factors had to be adjusted for in smaller datasets (see Table 5). Their inclusion in multivariable models did not alter the associations between the biomarkers and risk of any type, infection-related and infection-unrelated cancer.

Discussion

This is the largest prospective study to investigate, in the setting of HIV infection, the relationship between plasma levels of coagulation and inflammatory biomarkers and cancer risk. Here, we report that activated inflammation and coagulation pathways, as measured by higher IL-6, CRP and D-dimer levels, are associated with an increased risk of cancer. This association was strongest for IL-6 and evident for both infection-related and infection-unrelated cancer after excluding early events and adjusting for demographics, $CD4^+$ cell counts and traditional risk factors. During the entirety of follow-up, IL-6 had the steepest risk gradients for all cancer endpoints and, when the three biomarkers were entered into a multivariable Cox regression model, only IL-6 remained significantly associated with cancer risk.

The strong association between elevated plasma levels of IL-6 and cancer risk that we observed is corroborated by a body of evidence suggesting that IL-6 is a tumourigenic cytokine that influences, through autocrine [15] and paracrine [16] pathways, all stages of cancer development, including initiation, promotion, progression and dissemination [27]. Gene association studies using principles of Mendelian randomization have provided further evidence to support a role of IL-6 in cancer aetiogenesis, with IL-6 gene polymorphisms associated with colorectal [28], cervical [29] and oral cancer [30].

In the present study, associations between CRP and cancer risk differed according to cancer type. Site-specific associations between inflammatory markers and cancer have also been described in the general population. Elevated plasma levels of CRP were found to be linked with lung [31,32] and colorectal cancer [31], but not with breast cancer [33]. On the contrary, a significantly increased risk of lung and colorectal cancer [18], but not of prostate cancer [34], was observed in HIV-uninfected individuals with raised circulating levels of IL-6. In contrast with the few previous reports in HIV-infected populations [21,22], we did not find a significant association between IL-6 and NHL. Instead, we did observe an association between CRP and Hodgkin lymphoma and all lymphomas grouped. Nevertheless, we had limited ability to examine site-specific associations, which should be confirmed in larger studies.

The possibility that raised plasma levels of biomarkers, rather than predicting clinical endpoints, just reflect the presence of subclinical disease (i.e. reverse causality) is difficult to

exclude in cancer or other diseases with long latency. In this study, risk gradients with CRP did not vary over follow-up and although risk gradients with IL-6 did vary over follow-up, hazard ratios associated with higher IL-6 levels remained significant for all cancers after exclusion of early cancer events. Furthermore, HIV-positive patients frequently have more aggressive cancer and poorer clinical outcomes [35]. Therefore, we consider it unlikely that our results can be explained by occult cancer.

The main strengths of our study are its large sample size, long follow-up period and carefully collected clinical data. A number of limitations need to be considered. First, the measurement of a more extensive panel of inflammatory and coagulation biomarkers would have been helpful to further clarify the relationship between activated inflammatory and coagulation pathways and the risk of cancer in HIV-infected individuals. Second, our analyses were not adjusted for some important traditional cancer risk factors, namely alcohol use, diet and sun exposure. Third, given the small numbers of cases, we were unable to carry out separate analyses for less common cancers and had no alternative but to use, as endpoints, broader cancer categories involving widely heterogeneous and etiologically distinct malignancies. However, this implies that the strong associations observed for IL-6 are conservative and may be stronger for individual cancers within broad categories. Finally, this study does not provide definitive evidence for a causal relationship between activated coagulation/inflammation and cancer during HIV infection; prospective gene association studies in HIV-infected individuals or a protective clinical effect on risk of cancer development from medical interventions able to specifically reduce the production or effect of IL-6 may be useful to elucidate this.

Our results indicate that activated inflammation and coagulation, as demonstrated by higher IL-6, CRP and D-dimer levels, are associated with the development of cancer during HIV infection. This association was strongest for IL-6 and was present for both infection-related and infection-unrelated malignancies after the exclusion of early events and after adjustment for demographics, $CD4^+$ cell counts and traditional cancer risk factors. Trials of interventions may be warranted to assess whether cancer risk can be reduced by lowering IL-6 levels in HIV-positive individuals.

Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

Acknowledgments

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References

1. Centers for Disease Control and Prevention. 1993 revised classification system for HIV infection and expanded surveillance case definition for AIDS among adolescents and adults. MMWR Recomm Rep. 1992; 41:1–19.
2. Grulich AE, van Leeuwen MT, Falster MO, Vajdic CM. Incidence of cancers in people with HIV/AIDS compared with immunosuppressed transplant recipients: a meta-analysis. Lancet. 2007; 370:59–67. [PubMed: 17617273]
3. Patel P, Hanson DL, Sullivan PS, Novak RM, Moorman AC, Tong TC, et al. Incidence of types of cancer among HIV-infected persons compared with the general population in the United States, 1992–2003. Ann Intern Med. 2008; 148:728–736. [PubMed: 18490686]
4. Bedimo RJ, McGinnis KA, Dunlap M, Rodriguez-Barradas MC, Justice AC. Incidence of non-AIDS-defining malignancies in HIV-infected versus noninfected patients in the HAART era: impact of immunosuppression. J Acquir Immune Defic Syndr. 2009; 52:203–208. [PubMed: 19617846]
5. Silverberg MJ, Chao C, Leyden WA, Xu L, Tang B, Horberg MA, et al. HIV infection and the risk of cancers with and without a known infectious cause. AIDS. 2009; 23:2337–2345. [PubMed: 19741479]
6. Grulich AE, Jin F, Poynten IM, Vajdic CM. HIV, cancer, and aging. Sex Health. 2011; 8:521–525. [PubMed: 22127038]
7. Guiguet M, Boué F, Cadranel J, Lang JM, Rosenthal E, Costagliola D. Clinical Epidemiology Group of the FHDH-ANRS CO4 cohort. Effect of immunodeficiency, HIV viral load, and antiretroviral therapy on the risk of individual malignancies (FHDH-ANRS CO4): a prospective cohort study. Lancet Oncol. 2009; 10:1152–1159. [PubMed: 19818686]
8. Silverberg MJ, Chao C, Leyden WA, Xu L, Horberg MA, Klein D, et al. HIV infection, immunodeficiency, viral replication, and the risk of cancer. Cancer Epidemiol Biomarkers Prev. 2011; 20:2551–2559. [PubMed: 22109347]
9. Kesselring A, Gras L, Smit C, van Twillert G, Verbon A, de Wolf F, et al. Immunodeficiency as a risk factor for non-AIDS-defining malignancies in HIV-1-infected patients receiving combination antiretroviral therapy. Clin Infect Dis. 2011; 52:1458–1465. [PubMed: 21628488]
10. Worm, S.; Bower, M.; Reiss, P.; Grulich, A.; Fontas, E.; Bonnet, F., et al. Non-AIDS defining malignancies (NADM) and immunosuppression: the D:A:D study; 19th Conference on Retroviruses and Opportunistic Infections; 5–8 March 2012; Seattle, Washington. Washington 2012
11. Shiels MS, Pfeiffer RM, Gail MH, Hall HI, Li J, Chaturvedi AK, Bhatia K, et al. Cancer burden in the HIV-infected population in the United States. J Natl Cancer Inst. 2011; 103:753–762. [PubMed: 21483021]
12. El-Sadr WM, Lundgren JD, Neaton JD, Gordin F, Abrams D, Arduino RC, et al. CD4+ count-guided interruption of antiretroviral treatment. N Engl J Med. 2006; 355:2283–2296. [PubMed: 17135583]
13. Silverberg MJ, Neuhaus J, Bower M, Gey D, Hatzakis A, Henry K, et al. Risk of cancers during interrupted antiretroviral therapy in the SMART study. AIDS. 2007; 21:1957–1963. [PubMed: 17721103]
14. Kuller LH, Tracy R, Belloso W, De Wit S, Drummond F, Lane HC, et al. Inflammatory and coagulation biomarkers and mortality in patients with HIV infection. PLoS Med. 2008; 5:e203. [PubMed: 18942885]
15. Dougan M, Li D, Neuberg D, Mihm M, Googe P, Wong KK, et al. A dual role for the immune response in a mouse model of inflammation-associated lung cancer. J Clin Invest. 2011; 121:2436–2446. [PubMed: 21537082]
16. Stone RL, Nick AM, McNeish IA, Balkwill F, Han HD, Bottsford-Miller J, et al. Paraneoplastic thrombocytosis in ovarian cancer. N Engl J Med. 2012; 366:610–618. [PubMed: 22335738]
17. Heinrich PC, Castell JV, Andus T. Interleukin-6 and the acute phase response. Biochem J. 1990; 265:621–636. [PubMed: 1689567]

18. Il'yasova D, Colbert LH, Harris TB, Newman AB, Bauer DC, Satterfield S, et al. Circulating levels of inflammatory markers and cancer risk in the health aging and body composition cohort. Cancer Epidemiol Biomarkers Prev. 2005; 14:2413–2418. [PubMed: 16214925]
19. Siemes C, Visser LE, Coebergh JW, Splinter TA, Witteman JC, Uitterlinden AG, et al. C-reactive protein levels, variation in the C-reactive protein gene, and cancer risk: the Rotterdam Study. J Clin Oncol. 2006; 24:5216–5222. [PubMed: 17114654]
20. Pine SR, Mechanic LE, Enewold L, Chaturvedi AK, Katki HA, Zheng YL, et al. Increased levels of circulating interleukin 6, interleukin 8, C-reactive protein, and risk of lung cancer. J Natl Cancer Inst. 2011; 103:1112–1122. [PubMed: 21685357]
21. Breen EC, van der Meijden M, Cumberland W, Kishimoto T, Detels R, Martínez-Maza O. The development of AIDS-associated Burkitt's/small noncleaved cell lymphoma is preceded by elevated serum levels of interleukin 6. Clin Immunol. 1999; 92:293–2999. [PubMed: 10479534]
22. Breen EC, Hussain SK, Magpantay L, Jacobson LP, Detels R, Rabkin CS, et al. B-cell stimulatory cytokines and markers of immune activation are elevated several years prior to the diagnosis of systemic AIDS-associated non-Hodgkin B-cell lymphoma. Cancer Epidemiol Biomarkers Prev. 2011; 20:1303–1314. [PubMed: 21527584]
23. Abrams D, Lévy Y, Losso MH, Babiker A, Collins G, Cooper DA, et al. Interleukin-2 therapy in patients with HIV infection. N Engl J Med. 2009; 361:1548–1559. [PubMed: 19828532]
24. Neuhaus J, Jacobs DR Jr, Baker JV, Calmy A, Duprez D, La Rosa A, et al. Markers of inflammation, coagulation, and renal function are elevated in adults with HIV infection. J Infect Dis. 2010; 201:1788–1795. [PubMed: 20446848]
25. Bouvard V, Baan R, Straif K, Grosse Y, Secretan B, El Ghissassi F, et al. A review of human carcinogens – Part B: biological agents. Lancet Oncol. 2009; 10:321–322. [PubMed: 19350698]
26. Wei LJ, Lin DY, Weissfeld L. Regression analysis of multivariate incomplete failure time data by modelling marginal distributions. J Am Stat Assoc. 1989; 84:1065–1073.
27. Grivennikov SI, Karin M. Inflammatory cytokines in cancer: tumour necrosis factor and interleukin 6 take the stage. Ann Rheum Dis. 2011; 70(Suppl 1):104–108. [PubMed: 20679474]
28. Theodoropoulos G, Papaconstantinou I, Felekouras E, Nikiteas N, Karakitsos P, Panoussopoulos D, et al. Relation between common polymorphisms in genes related to inflammatory response and colorectal cancer. World J Gastroenterol. 2006; 12:5037–5043. [PubMed: 16937502]
29. Nogueira de Souza NC, Brenna SM, Campos F, Syrjänen KJ, Baracat EC, Silva ID. Interleukin-6 polymorphisms and the risk of cervical cancer. Int J Gynecol Cancer. 2006; 16:1278–1282. [PubMed: 16803517]
30. Vairaktaris E, Yiannopoulos A, Vylliotis A, Yapijakis C, Derka S, Vassiliou S, et al. Strong association of interleukin-6-174 G>C promoter polymorphism with increased risk of oral cancer. Int J Biol Markers. 2006; 21:246–250. [PubMed: 17177164]
31. Allin KH, Bojesen SE, Nordestgaard BG. Baseline C-reactive protein is associated with incident cancer and survival in patients with cancer. J Clin Oncol. 2009; 27:2217–2224. [PubMed: 19289618]
32. Chaturvedi AK, Caporaso NE, Katki HA, Wong HL, Chatterjee N, Pine SR, et al. C-reactive protein and risk of lung cancer. J Clin Oncol. 2010; 28:2719–2726. [PubMed: 20421535]
33. Zhang GJ, Adachi I. Serum interleukin-6 levels correlate to tumor progression and prognosis in metastatic breast carcinoma. Anticancer Res. 1999; 19:1427–1432. [PubMed: 10365118]
34. Baillargeon J, Platz EA, Rose DP, Pollock BH, Ankerst DP, Haffner S, et al. Obesity, adipokines, and prostate cancer in a prospective population-based study. Cancer Epidemiol Biomarkers Prev. 2006; 15:1331–1335. [PubMed: 16835332]
35. Spano JP, Costagliola D, Katlama C, Mounier N, Oksenhendler E, Khayat D. AIDS-related malignancies: state of the art and therapeutic challenges. J Clin Oncol. 2008; 26:4834–4842. [PubMed: 18591544]

Table 1
Frequency distribution of cancers ESPRIT, SILCAAT and SMART control patients

	Number	Frequency (%)
Infection-unrelated cancers	102	100
Lung	26	26
Prostate	14	14
Colorectal	12	12
Breast	8	8
Melanoma	7	7
Urinary tract	6	6
Pancreas	3	3
Oesophagus	2	2
Leukemia	2	2
Unknown primary site	9	9
Other[a]	13	13
Infection-related cancers	70	100
Non-Hodgkin lymphoma	21	30
Anus	13	19
Hodgkin lymphoma	10	14
Kaposi sarcoma	10	14
Liver	5	7
Oral cavity/pharynx	4	6
Cervix	4	6
Other[b]	3	4

[a] Female reproductive tract, mesotheliomas, larynx, small intestine, thyroid, testicle, adrenal gland.

[b] Penis, vagina, stomach.

Table 2
Baseline characteristics ESPRIT, SILCAAT and SMART control patients

	Developed cancer		
	Yes	**No**	***P****
Age in years (median, IQR)	48 (41, 56)	42 (36, 49)	<0.001
Female sex (%)	12.2	22.7	0.005
Race (%)			0.05
Black	16.3	19.6	
White	76.7	67.2	
Other	7.0	13.1	
BMI (median, IQR)	24 (22, 26)	24 (22, 27)	0.67
AIDS (%)	25.0	26.1	0.67
Hepatitis B/C coinfection (%)[a]	27.9	18.6	0.04
Diabetes[a]	8.6	4.9	0.02
Smoking[b]	60.3	40.5	0.003
$CD4^+$ cell count (cells/μl) (median, IQR)	421 (305, 585)	488 (370, 672)	0.27
$CD4^+$ nadir (cells/μl)(median, IQR)	143 (44, 262)	200 (87, 319)	0.01
HIV RNA ≤500 copies/ml (%)	72.7	76.7	0.15
CRP (μg/ml) (median, IQR)	2.51 (1.25, 5.24)	1.54 (0.67, 3.54)	<0.001
D-dimer (μg/ml) (median, IQR)	0.28 (0.22, 0.47)	0.24 (0.15, 0.38)	<0.001
IL-6 (pg/ml) (median, IQR)	2.40 (1.80, 3.58)	1.80 (1.17, 2.80)	<0.001
Study			0.03
ESPRIT participant	40.7	35.2	
SILCAAT participant	25.6	13.6	
SMART participant	33.7	51.2	
No. of patients	172	4851	

CRP, C-reactive protein; IQR, interquartile range.

[a] Not ascertained for patients in SILCAAT.

[b] Not ascertained for patients in SILCAAT or ESPRIT.

* *P* value from a univariate Cox regression model stratified by study.

Table 3

ESPRIT, SMART and SILCAAT hazard ratios associated with baseline biomarkers [per one $\log_2$ (i.e. doubling) increment] cohort: all control patients

	Any cancer						Infection-related cancers[a]						Infection-unrelated cancers					
	CRP		D-dimer		IL-6		CRP		D-dimer		IL-6		CRP		D-dimer		IL-6	
	HR(95%CI)*	*P*	HR (95% CI)*	*P*	HR(95%CI)*	*P*	HR (95% CI)*	*P*	HR(95%CI)*	*P*	HR (95% CI)*	*P*	HR (95% CI)*	*P*	HR(95%CI)*	*P*	HR(95%CI)*	*Pe*
Model 1[b]	1.21 (1.11–1.32)	<0.001	1.25 (1.10–1.43)	<0.001	1.48 (1.30–1.69)	<0.001	1.22 (1.07–1.39)	0.003	1.25 (1.01–1.54)	0.04	1.49 (1.21–1.84)	<0.001	1.20 (1.08–1.34)	<0.001	1.26 (1.06; 1.49)	0.007	1.47 (1.24; 1.74)	<0.001
Model 2[c]	1.16 (1.06–1.26)	0.001	1.17 (1.01–1.35)	0.03	1.38 (1.19–1.59)	<0.001	1.18(1.03–1.35)	0.02	1.19 (0.95–1.50)	0.13	1.42 (1.14–1.78)	0.002	1.15 (1.02–1.28)	0.02	1.15 (0.96; 1.38)	0.12	1.35 (1.12; 1.63)	0.002
Model 3[d]	1.06 (0.96–1.17)	0.22	1.06 (0.91–1.23)	0.43	1.29 (1.09–1.52)	0.003	1.08 (0.92–1.26)	0.34	1.06 (0.83–1.36)	0.64	1.32 (1.01–1.71)	0.04	1.06 (0.93–1.20)	0.40	1.06 (0.88; 1.28)	0.53	1.27 (1.02; 1.58)	0.03
No. of patients[e]	5022		5006		4994		5022		5006		4994		5022		5006		4994	
No. of events[e]	172		171		171		70		70		70		102		101		101	

CI, confidence interval; CRP, C-reactive protein; HR, hazard ratio; IL-6, interleukin-6.

[a] All AIDS-defining-malignancies along with vagina, vulva, penis, anal and oral cavity/pharynx cancers; Hodgkin lymphoma; liver and stomach cancer. All other malignancies were considered to be infection-unrelated cancers.

[b] Stratified for study and unadjusted.

[c] Stratified for study and adjusted for demographics (age, race, sex and continent), and study entry and time-updated $CD4^+$ cell counts.

[d] As in model 2 and also adjusted for all biomarkers.

[e] Due to missing data, numbers are reduced in model containing all biomarkers to 4982 participants with 171 any cancer, 70 infection-related cancer and 101 infection-unrelated cancer events.

* HR, hazard ratio for one $\log_2$ (i.e. doubling) increase in indicated biomarker.

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Table 4

ESPRIT, SMART and SILCAAT hazard ratios for one $\log_2$ (i.e. doubling) increment in biomarkers for cancer subsets

	CRP			D-dimer			IL-6		
	Adj HR[a]	**95% CI**	***P***	**Adj HR[a]**	**95% CI**	***P***	**Adj HR[a]**	**95% CI**	***P***
Any cancer	1.16	1.06–1.26	0.001	1.17	1.01–1.35	0.03	1.38	1.19–1.59	<0.001
No. of patients (events)		5022 (172)			5006 (171)			4994 (171)	
Lymphoma – any	1.35	1.09–1.66	0.005	1.31	0.93–1.85	0.12	1.34	0.95–1.88	0.10
No. of patients (events)		5022 (31)			5006 (31)			4994 (31)	
Non-Hodgkin lymphoma	1.21	0.94–1.56	0.13	1.49	1.00–2.21	0.05	1.27	0.83–1.93	0.27
No. of patients (events)		5022 (21)			5006 (21)			4994 (21)	
Hodgkin lymphoma	1.72	1.18–2.50	0.004	0.98	0.51–1.91	0.96	1.52	0.85–2.72	0.16
No. of patients (events)		5022 (10)			5006 (10)			4994 (10)	
HPV-related cancers[b]	1.06	0.84–1.35	0.61	0.99	0.65–1.50	0.96	1.47	0.98–2.19	0.06
No. of patients (events)		5022 (23)			5006 (23)			4994 (23)	
Lung cancer	1.26	1.00–1.58	0.05	0.84	0.58–1.20	0.34	1.62	1.12–2.35	0.01
No. of patients (events)		5022 (26)			5006 (25)			4994 (25)	
Prostate cancer	0.84	0.62–1.15	0.28	0.91	0.59–1.42	0.68	0.62	0.33–1.17	0.14
No. of patients (events)		*5022 (14)*			*5006 (14)*			*4994 (14)*	
Colorectal cancer	1.41	1.02–1.94	0.04	0.95	0.56–1.59	0.84	1.32	0.77–2.26	0.31
No. of patients (events)		5022 (12)			5006 (12)			4994 (12)	
Kaposi sarcoma	1.05	0.72–1.51	0.81	1.55	0.93–2.57	0.09	1.58	0.92–2.69	0.09
No. of patients (events)		5022 (10)			5006 (10)			4994 (10)	

CI, confidence interval; CRP, C-reactive protein; HPV, human papillomavirus.

[a] HR, hazard ratio for one $\log_2$ increase in indicated biomarker. Model is stratified by study and adjusted for demographics (age, race, sex and continent), and study entry and time-updated $CD4^+$ cell counts.

[b] Invasive cervical cancer, anal, vagina, vulva, penis and oral cavity/pharynx.

Table 5

Sensitivity analysis – adjustment for BMI, diabetes and smoking hazard ratios for one $\log_2$ (i.e. doubling) increment in biomarkers ESPRIT, SMART and SILCAAT

	CRP		D-dimer		IL-6	
	HR[b]	*P*	HR[b]	*P*	HR[b]	*P*
Any cancer						
Full adjustment[a]	1.16	0.001	1.17	0.03	1.38	<0.001
No. of patients (events)	5022 (172)		5006 (171)		4994 (171)	
Full adjustment + BMI	1.16	<0.001	1.16	0.04	1.37	<0.001
No. of patients (events)	4948 (170)		4932 (169)		4920 (169)	
Full adjustment + smoking (SMART)	1.10	0.20	1.12	0.28	1.37	0.006
No. of patients (events)	2541 (58)		2526 (57)		2514 (57)	
Full adjustment + diabetes (SMART and ESPRIT)	1.15	0.008	1.14	0.10	1.42	<0.001
No. of patients (events)	4307 (128)		4291 (127)		4279 (127)	
Infection-related cancer						
Full adjustment[a]	1.18	0.02	1.19	0.13	1.42	0.002
No. of patients (events)	5022 (70)		5006 (70)		4994 (70)	
Full adjustment + BMI	1.18	0.02	1.19	0.15	1.40	0.004
No. of patients (events)	4948 (69)		4932 (69)		4920 (69)	
Full adjustment + smoking (SMART)	1.15	0.30	1.25	0.22	1.45	0.07
No. of patients (events)	2541 (18)		2526 (18)		2514 (18)	
Full adjustment + diabetes (SMART & ESPRIT)	1.20	0.03	1.25	0.08	1.56	<0.001
No. of patients (events)	4307 (51)		4291 (51)		4279 (51)	
Infection-unrelated cancer						
Full adjustment[a]	1.15	0.02	1.15	0.12	1.35	0.002
No. of patients (events)	5022 (102)		5006 (101)		4994 (101)	
Full adjustment + BMI	1.15	0.02	1.15	0.13	1.35	0.002
No. of patients (events)	4948 (101)		4932 (100)		4920 (100)	
Full adjustment + smoking (SMART)	10.08	0.39	1.06	0.65	1.35	0.04
No. of patients (events)	2541 (40)		2526 (39)		2514 (39)	

	CRP		D-dimer		IL-6	
	HR[b]	*P*	HR[b]	*P*	HR[b]	*P*
Full adjustment + diabetes (SMART & ESPRIT)	1.12	0.10	1.08	0.45	1.32	0.02
No. of patients (events)	4307 (77)		4291 (76)		4279 (76)	

CRP, C-reactive protein.

[a] Stratified by study and adjusted for demographics (age, race, sex, continent), and study entry and time-updated $CD4^+$ cell counts.

[b] HR, hazard ratio for one $\log_2$ increase in indicated biomarker.

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