

HIV and Aging: Impact on CD4 Gains, Frailty, Falls, Cognition, and MIs

Report on the 7th International Workshop on HIV and Aging, 26 – 27 September 2017, Washington, DC, USA

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In 2016 the recorded HIV epidemic had lasted 35 years. Many people with HIV have lasted much longer than that. The US Centers for Disease Control and Prevention (CDC) estimates that 42% of US residents diagnosed with HIV through 2013 were 50 years old or older, and 26% were 55 or older.¹ According to hiv-age.org, 55% of HIV-positive people in New York City 60% in San Francisco are 50 or older today.²

At the 7th International Workshop on HIV and Aging, held 26-27 September 2016 in Washington, DC, a modeling study based on data from 2982 HIV patients in Italy projected that people 65 and older will account for well over one third of the HIV population in 2030.³ (See “By 2030 over one third of HIV patients 65 or older” below.) Severe frailty, the model figured, will affect half of HIV patients in 2030 compared with one quarter today.

Survival of virally suppressed HIV-positive people in the West still lags survival of the general population, according to studies in Denmark⁴ and the United States.⁵ But longer survival with HIV thanks to better care largely explains the graying of the global HIV population. To further improve longevity with HIV infection, researchers are pursuing work on several fronts, many of them covered at the 7th Aging Workshop and detailed in this report—from CD4 gains, frailty, and falls, to myocardial infarction, neurocognitive function, and the epigenetic clock.

UNDERSTANDING AN AGING HIV EPIDEMIC

By 2030 over one third of HIV patients 65 or older

Many HIV clinicians will be de facto geriatricians by 2030, according to results of a modeling study involving the Modena HIV Metabolic Clinic in Italy.³ More than one third of HIV patients will be 65 years old or older, according to the model, elaborated in collaboration with the University College London in the context of the European Union COBRA study.

In the United States, the CDC figures, more than 40% of people diagnosed with HIV were 50 or older as long ago as 2013.¹ Other high-income countries with similar HIV epidemics probably have similar portions of people past 50. But what fraction of HIV patients are 65 or older today, and what fraction will be that old 15 years from now? The Modena researchers aimed to answer those questions; they stressed, however, that the most novel study goal was not how old HIV patients will be in 2030 or even how many of them will have comorbidities or multimorbidity. Rather, they maintained, the most interesting outcome was the extent to which these old patients will experience geriatric syndrome (indicated by falls) and disability in daily living activities.

To find out they created an individual patient-based model of the HIV population at the Modena HIV Metabolic Clinic from 2009 through 2015. They fed the model with data collected on clinical variables over time to generate the number, age, and gender of new individual entries up to 2030. The Modena team postulated that relationships between age, gender, falls, and disability seen in 2014-2015 would remain constant over time.

The Modena team figured a frailty index for each current patient by a deficit-accumulation model that counts clinical deficits on a list of 37 such conditions. The frailty index for each person is the number of deficits that person accumulates divided by 37. An index from 0.3 to 0.4 indicates frailty, and an

index above 0.4 indicates “most frail.” Current patients self-reported fall frequency in the past 12 months. The researchers defined disability of current patients as impairment in 1 or more of the 8 Instrumental Activities of Daily Living (IADL): housekeeping, money management, cooking, transportation, telephone use, shopping, laundry, and medication management.

The 2009-2015 population included 2982 patients with a median age of 49 years (interquartile range [IQR] 45 to 54). Almost one third of these people (31.9%) were women, and 40% smoked. Median body mass index measured 23.5 kg/m² and median waist circumference 87 cm. These clinic patients had taken antiretroviral therapy for a median of 5.2 years (IQR 2.6 to 7.8) and had a median CD4 count of 648 cells/mm³ (IQR 474 to 841). The most prevalent comorbidities were dyslipidemia (82%), lipodystrophy (76%), vitamin D insufficiency (69%), hypertension (37%), nonalcoholic fatty liver disease (24%), and impaired fasting glucose (20%). Rates of diabetes, cardiovascular disease, chronic kidney disease, osteoporosis, and AIDS malignancy were all below 15%.

The model determined that the proportion of HIV clinic patients 65 year old or older will climb from 4.3% in 2016, to 6.5% in 2020, to 15.8% in 2025, and to 37% in 2030 (**Figure 1**). About 95% of HIV patients will be 50 or older in 2030. Frail individuals make up about 30% of the clinic population from 2016 through 2030, the model projected. But 48.2% of clinic patients will be in the “most frail” group in 2030 compared with 24.5% today (**Figure 2**). Whereas 20.4% of individuals will experience falls in 2016, that proportion will jump to 29.8% in 2030. And while 22.2% of 2016 patients will have IADL-determined disability, 33.7% will have disability in 2030.

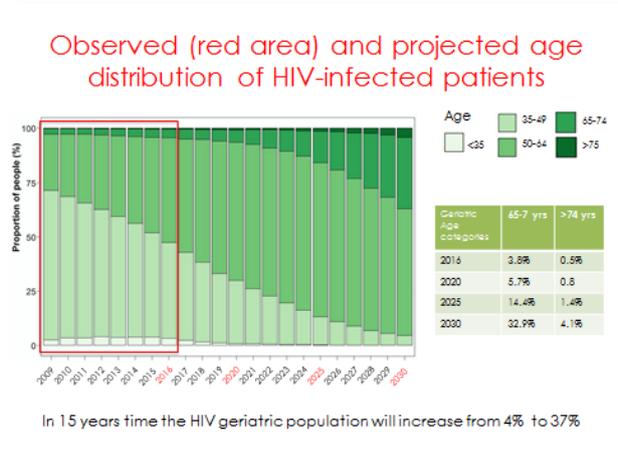


Figure 1. A modeling study at the Modena HIV Metabolic Clinic determined that more than one third of patients will be 65 years old or older in 2030, compared with 4.3% today. (Source: Giovanni Guaraldi and colleagues, Modena HIV Metabolic Clinic.³)

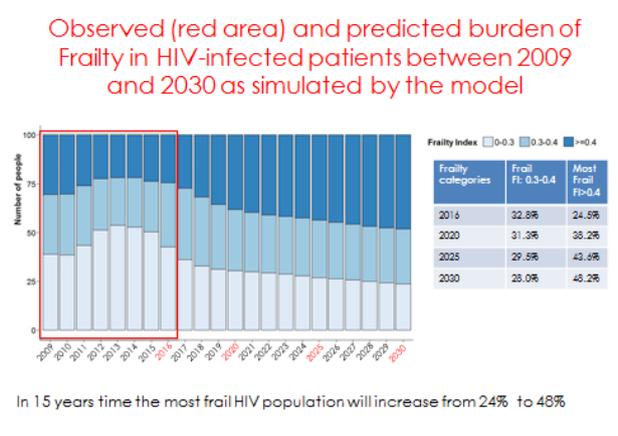


Figure 2. The Modena model predicted that the proportion of “most frail” HIV patients (frailty index > 0.4) will double from 24.5% in 2016 to 48.2% in 2030. (Source: Giovanni Guaraldi and colleagues, Modena HIV Metabolic Clinic.³)

The Modena investigators proposed that the HIV aging epidemic has moved “far beyond” a simple increase in age or a rising prevalence of age-related comorbidities. Instead, their model predicts a “geriatric HIV scenario” where multimorbidity is the norm and frailty becomes common. The researchers believe HIV care teams must add geriatricians and occupational therapists. They suggested that the HIV care model must address not only clinical outcomes, but also quality of life,

neurocognitive impairment, physical function, and geriatric syndromes.

High rates of untreated metabolic comorbidities in DC cohort

While the Modena model offered a look at the aging HIV epidemic 15 years from now³ (see preceding section), a study in Washington, DC provided hard numbers about age-related comorbidities in a large urban cohort today.⁶ Four metabolic comorbidities that predispose individuals to cardiovascular disease proved highly prevalent in a large contemporary cohort of people in care for HIV infection in Washington, DC.⁶ The comorbidities—hypertension, type 2 diabetes, dyslipidemia, and obesity—were especially prevalent in older cohort members, and high proportions of people with these conditions had no record of treatment at their HIV care site.

Researchers from George Washington University and other centers noted that people with HIV face a 1.5- to 2-fold higher risk of cardiovascular disease than the general population. Understanding population-level prevalence of metabolic comorbidities that may lead to cardiovascular disease, the researchers said, can characterize the epidemiology of these conditions and guide promotion of appropriate lifestyle and pharmacologic interventions for people with HIV.

To address those needs, the investigators analyzed comorbidity data gathered in the central database of the DC Cohort study, a prospective observational cohort involving patients from 13 centers across the city. They focused on people at least 18 years old and enrolled in the cohort during the period 2011-2015. The researchers established diagnostic definitions for hypertension, type 2 diabetes, dyslipidemia, and obesity by using ICD-9 codes, documented treatment for these conditions, or measures of blood pressure, fasting or nonfasting glucose or HbA1c, fasting or nonfasting total cholesterol or high-density lipoprotein (HDL) cholesterol, or body mass index.

The study cohort included 7018 HIV-positive adults with a median age of 50 years (IQR 39 to 57) and with 18% aged 60 or older. While 73% of participants were male at birth, 77% were non-Hispanic black. Almost everyone, 97%, took antiretrovirals at some point, and 63% smoked currently or in the past.

During the 2011-2015 study period, nearly half of cohort members had hypertension (49.8%) or dyslipidemia (48.0%), while 35.2% were obese and 12.9% had type 2 diabetes. Almost half of these people, 45.6%, had two or more comorbidities, including 26% with exactly 2, 15% with exactly 3, and 5% with all 4. Prevalence of the four comorbidities proved even higher in people 60 year old or older (Figure 3). More than three quarters of cohort members 70 years old or older (77.5%) had multimorbidity.

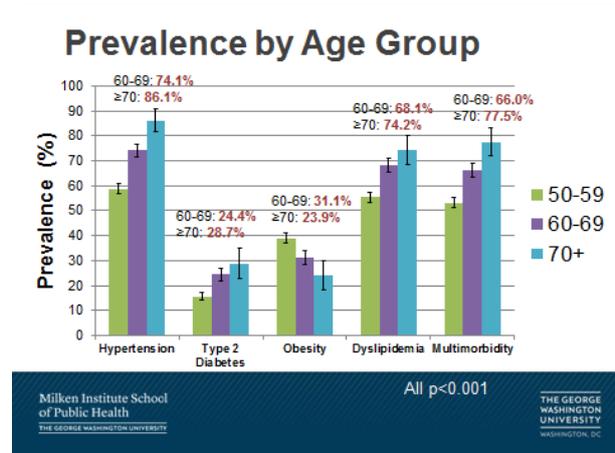


Figure 3. Prevalence of comorbidities that predispose to cardiovascular disease proved especially high in older members of a 7018-person HIV cohort in Washington, DC. (Source: Matthew Levy, George Washington University, and colleagues.⁶)

Obesity affected more than half of women versus fewer than 30% of men. Women had slightly higher rates of diabetes, while about 50% of men and 40% of women had dyslipidemia. Higher proportions of non-Hispanic blacks had hypertension, diabetes, and obesity, while non-Hispanic whites were more likely to have dyslipidemia. High proportions of participants with a comorbidity lacked evidence of treatment in the medical record at their HIV care site, including 38% with hypertension, 40% with diabetes, and 56% with dyslipidemia.

Multivariable logistic regression identified two independent correlates of hypertension (longer duration of any nonnucleoside regimen and longer duration of any other antiretroviral regimen), one independent correlate of type 2 diabetes (longer time since HIV diagnosis), four independent correlates of dyslipidemia (longer time since HIV diagnosis, longer duration of any protease inhibitor regimen, longer duration of any nonnucleoside regimen, and longer duration of any other antiretroviral regimen), and two independent correlates of obesity (shorter duration of any nonnucleoside regimen and shorter duration of any other antiretroviral regimen).

Given the high prevalence of metabolic comorbidities in this population and the apparent lack of treatment in many affected people, the researchers suggested that “models of HIV care that more centrally incorporate primary and secondary prevention of metabolic disease may be warranted.” They noted that diabetes prevalence was lower in their cohort than in other HIV cohorts, perhaps because they often had to rely on nonfasting glucose levels.

Up to 40% of MIs in HIV+ avoidable by preventing hypertension or smoking

Preventing hypertension or smoking would each lower rates of type 1 myocardial infarctions (MIs) by about 40% and rates of type 2 MIs by about 25%, according to calculation of population-attributable fractions (PAFs) in NA-ACCORD, a large North American HIV cohort.⁷ Avoiding low CD4 counts and high viral loads would also have an impact on both MI types.

The higher MI rate in people with HIV than in the general population can be traced to both traditional risk factors and HIV-specific factors. NA-ACCORD investigators who conducted this study noted that risk factors may differ for type 1 MIs (atherothrombotic coronary events resulting from plaque rupture) and type 2 MIs (mismatched oxygen supply and demand often related to sepsis or cocaine use).

To determine the relative contributions of traditional and HIV-related risk factors to each type of MI, the NA-ACCORD team estimated PAFs for an array of risk factors. PAF represents the proportion of a condition (in this case MI) that can be avoided if everyone in a population avoids the risk factor of interest.⁸

The analysis involved HIV-positive adults from 7 NA-ACCORD cohorts in the period from January 2000 to December 2009. The NA-ACCORD population is similar to the HIV population across the United States. The investigators counted MESA-validated incident type 1 or 2 MIs then explored the impact of traditional risk factors (smoking, statin use or total cholesterol above 240 mg/dL, hypertension, diabetes, stage 4 chronic kidney disease (CKD), and HCV infection) and HIV-related factors (CD4 count below 200 cells/mm³, viral load at or above 400 copies/mL, and a clinical AIDS diagnosis). They estimated PAF in analyses considering (1) prevalence of the risk factor and (2) hazard ratio for the factor adjusted for age, sex, race, and all modifiable risk factors considered.

The 29,100-person study population had 347 type 1 MIs and 275 type 2 MIs during a median follow-up of 3.5 years. Compared with participants who did not have a type 1 MI, those who did were older, more likely to be men, more likely to be white, and less likely to be antiretroviral-naïve at baseline. Those with versus without type 2 MIs were older, more likely to be women, and more likely to be black. Prevalence of 6 risk factors was higher in people with incident type 1 or 2 MIs: smoking, elevated total cholesterol, treated hypertension, low CD4 count, clinical AIDS diagnosis, and HCV. Additional risk factors specific to type 2 MIs were stage 4 CKD and detectable viral load, while diabetes prevalence was higher in participants with than without type 1 MI.

Adjusted hazard ratios indicated five independent predictors of type 1 MIs (smoking, treated hypertension, diabetes, stage 4 CKD, and CD4 count below 200 cells/mm³). Those five factors independently predicted type 2 MI, as did viral load at or above 400 copies/mL, a clinical AIDS diagnosis, and HCV.

PAF analysis determined that eliminating high total cholesterol would prevent 43% of type 1 MIs, eliminating hypertension would prevent 41%, and eliminating smoking would prevent 38% (**Figure 4**). Eliminating two of these risk factors would also have substantial impacts on preventing type 2 MIs: hypertension (PAF 26%) and ever smoking (PAF 22%). Three HIV-related factors had larger PAFs for type 2 MIs than type 1 MIs: detectable viral load (20% versus 6%), a clinical AIDS diagnosis

(12% versus 2%), and CD4 count below 200 cells/mm³ (11% versus 10%). Eliminating HCV would prevent 19% of type 2 MIs and 8% of type 1 MIs; respective PAFs for stage 4 CKD were 8% and 3%, and for diabetes 8% and 2%.

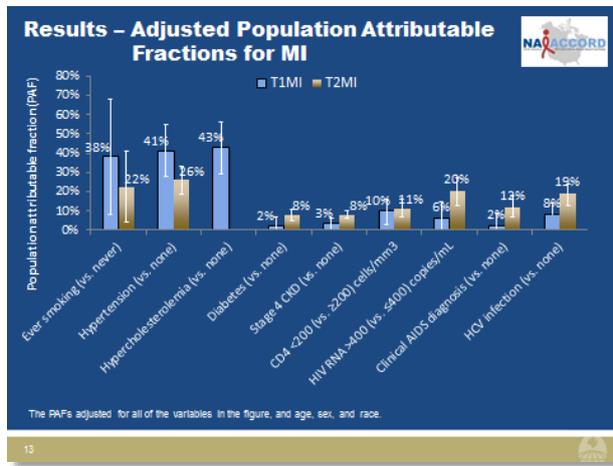


Figure 4. Population-attributable fractions for type 1 and type 2 MIs indicated that eliminating smoking and hypertension would have strong impacts in preventing both types; eliminating high cholesterol would have the strongest impact on type 1 MIs; and eliminating HCV and certain HIV-related factors would have stronger impacts on type 2 MIs than type 1 MIs. (Source: Keri Althoff, Johns Hopkins University, and colleagues.⁷)

The NA-ACCORD investigators proposed that interventions to prevent smoking, hypertension, high cholesterol, and HCV infection would reduce the MI burden in HIV populations like this one. Starting antiretroviral therapy early to preserve CD4 counts and sustain viral suppression could also have a considerable impact.

Resting metabolic rate higher in older men with than without HIV

Resting metabolic rate—a mortality predictor⁹—proved higher in older men with than without HIV, according to an analysis of men in the Baltimore group of the Multicenter AIDS Cohort Study (MACS).¹⁰ This association was exacerbated by metabolic comorbidities such as diabetes and impaired kidney function.

Resting metabolic rate—also called the energetic cost of living—slows with age, mainly because of changing body composition. Previous work by some of the investigators who conducted this MACS analysis linked “ideal” aging to a lower resting metabolic rate.¹¹ Previous work suggests resting metabolic rate is higher in people with uncontrolled HIV infection and in those with AIDS. Researchers conducted the new analysis to determine the impact of controlled HIV infection on the association between resting metabolic rate and age.

The analysis involved HIV-positive men who have sex with men (MSM) in the MACS Baltimore contingent and HIV-negative MSM from the same MACS group. All men were 40 years old or older and participating in MACS in 2015-2016. Researchers assessed resting metabolic rate by indirect calorimetry. To explore the association between HIV status and resting metabolic rate, they used linear regression models adjusted for age, DXA-determined body composition, and history of chronic conditions.

The 40 men with HIV averaged 58.5 years in age, compared with 64.5 years in the 35 HIV-negative men ($P < 0.01$), and a higher proportion of men with HIV were black (57.5% versus 22.9%, $P < 0.01$). Extremity fat mass tended to be lower in men with HIV (11.6 versus 13.3 kg, $P = 0.18$), but body mass index, trunk fat mass, and lean mass did not differ substantially between the two groups.

Despite their younger age and lower extremity fat mass, men with HIV tended to have a higher resting metabolic rate than HIV-negative men—1748 versus 1668 kcal/day ($P = 0.20$). Among men with HIV, the combined regression model identified four predictors of resting metabolic rate—lean mass (coefficient = 15.9, $P < 0.01$), fat mass (coefficient = 6.2, $P = 0.03$), estimated glomerular filtration rate (eGFR) (coefficient = 2.1, $P = 0.04$), and diabetes (coefficient = 105.3, $P = 0.08$). HIV itself did not independently predict resting metabolic rate (coefficient = 66.1, $P = 0.17$). In an analysis limited to HIV-positive men, lean mass (coefficient = 21.5, $P < 0.01$), eGFR (coefficient = 4.5, $P < 0.01$), and diabetes (coefficient = 225.4, $P < 0.01$) remained independent predictors of resting metabolic rate.

The researchers calculated that an HIV-positive 60-year-old man with 60 kg lean mass, 30 kg fat mass, eGFR 90 mL/min, and diabetes would have a 584-kcal/day higher resting metabolic rate than an HIV-negative diabetic man with the same lean mass, fat mass, and eGFR (2114 versus 1530 kcal/day) (**Table 1**). An HIV-positive man with the same lean mass, fat mass, and eGFR but *without* diabetes would have only a 187-kcal/day higher resting metabolic rate than an HIV-negative nondiabetic man with the same other numbers (1889 versus 1702 kcal/day). Those calculations led the investigators to suggest that the difference in resting metabolic rate between men with and without HIV may be driven in part by metabolic complications like diabetes.

Table 1. Metabolic complications like diabetes may drive the higher resting metabolic rate in men with versus without HIV

HIV-positive 60-year-old man With diabetes	HIV-negative 60-year-old man With diabetes	HIV-positive 60-year-old man Without diabetes	HIV-negative 60-year-old man Without diabetes
60 kg lean mass	60 kg lean mass	60 kg lean mass	60 kg lean mass
30 kg fat mass (33%)	30 kg fat mass (33%)	30 kg fat mass (33%)	30 kg fat mass (33%)
eGFR 90 mL/min	eGFR 90 mL/min	eGFR 90 mL/min	eGFR 90 mL/min
RMR 2114 kcal/day	RMR 1530 kcal/day	RMR 1889 kcal/day	RMR 1702 kcal/day
Difference = 584 kcal/day		Difference = 187 kcal/day	

RMR, resting metabolic rate.

Source: Jennifer Schrack, Johns Hopkins Bloomberg School of Public Health, and colleagues.¹⁰

Further analysis indicated that HIV-positive men with a higher than expected resting metabolic rate tended to have a lower nadir CD4 count (253 versus 344 cells/mm³, $P = 0.24$). But neither current nor cumulative viral load appeared to affect resting metabolic rate.

The researchers concluded that “HIV appears to attenuate the age-related decline in resting metabolic rate.” They suggested that metabolic comorbidities like diabetes and impaired kidney function may exacerbate this effect. But they noted that more in-depth analyses will require a bigger sample size.

Time told on epigenetic clock indicates HIV accelerates aging

HIV infection accelerates aging, according to time told on an epigenetic clock—a biological aging clock that uses DNA methylation levels as a marker of chronological age in years.¹² Steve Horvath of the University of California, Los Angeles, explained to HIV and Aging Workshop attendees that the DNA molecule is methylated at two center cytosines (**Figure 5**). DNA methylation plays a key role in disease development through epigenetic gene regulation. *Epigenetic* combines the prefix *epi* (meaning “besides,” as in *epiphenomenon*) with *genetic* to indicate nongenetic influences on gene expression—like methylation.

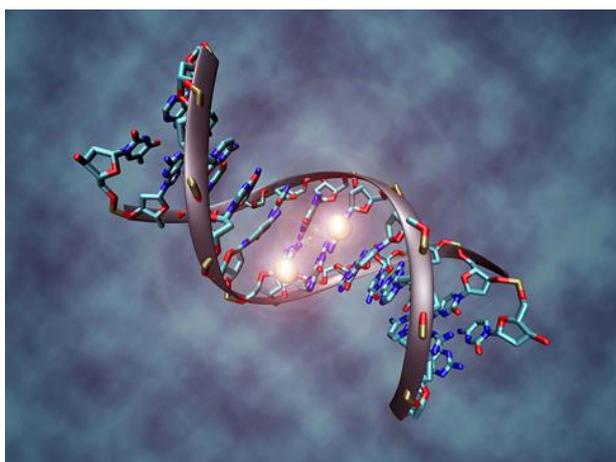


Figure 5. Measuring methylation of the two center cytosines in DNA provides an epigenetic clock that offers a molecular marker for chronological age. (Source: Christoph Bock, Research Center for Molecular Medicine of the Austrian Academy of Sciences.)

Research by Horvath shows that the epigenetic clock can tell time by measuring methylation in blood cells, neurons, glial cells, and many other cells and tissues¹³—but not in sperm. In the work describing this finding, Horvath proposed that “DNA methylation age measures the cumulative effect of an epigenetic maintenance system.”¹³ He explained that the epigenetic clock method involves three steps: (1) Measure DNA methylation levels of 353 CpGs. (CpG indicates DNA cytosine and guanine separated by one phosphate.) (2) Form a weighted average. (3) Transform the average into units of “years.” The result is an age estimate known as “epigenetic age” or “DNA methylation age.”

Phenotypes strongly linked to the epigenetic clock (so far) include body mass index, cancer, cognitive performance, Down syndrome,¹⁴ grip strength, Huntington disease,¹⁵ obesity, osteoarthritis, walking speed, and HIV infection.¹⁶ Assessing methylation in DNA from brain and blood, he determined that HIV increases epigenetic age in brain tissue by 7.4 years and in blood by 5.2 years. Horvath and colleague Andrew Levine believe their results “demonstrate that the epigenetic clock is a useful biomarker for detecting accelerated aging effects due to HIV infection.”¹⁶

Further work by Levine, Horvath, and others found evidence linking accelerated epigenetic aging in the brain to premortem HIV-related neurocognitive disorders.¹⁷ This analysis involved DNA from occipital cortex tissue bank specimens from 58 HIV-positive adults who underwent neurocognitive testing within 1 year of death. The researchers compared epigenetic clock-measured biological age with chronological age to gauge age acceleration. People with HIV-associated neurocognitive disorder (HAND) did not differ from neurocognitively normal people in demographic, histologic, neuropathologic, or virologic variables. Yet HAND was associated with a 3.5-year age acceleration compared with neurocognitively normal individuals.

The epigenetic clock also predicted all-cause mortality in work by Horvath and colleagues¹⁸ and by groups in Denmark¹⁹ and Germany.²⁰ A meta-analysis by Brian Chen, Horvath, and other researchers confirmed that a blood-based epigenetic clock predicts all-cause mortality independently of chronological age and traditional mortality risk factors.²¹ The analysis combined data on 13,089 individuals in 13 cohorts including non-Hispanic whites, Hispanics, and African Americans. All considered epigenetic age measures strongly predicted all-cause mortality ($P \leq 8.2 \times 10^{-9}$), and this association held true within each of the three racial/ethnic groups.

Horvath concluded that “the epigenetic clock is an attractive molecular biomarker of aging” that offers a highly robust measurement. The clock allows comparison of ages in different tissues, is strongly associated with many age-related conditions, and predicts mortality.

CD4 COUNT GAINS AND CD4s IN GALT

CD4 gains on ART differ by age in large, long-term African study

People 60 years old or older gained fewer CD4 cells than younger individuals in the 48 months after starting antiretroviral therapy (ART), according to an analysis of more than 158,000 people in four African countries.²² The difference between people 60 and older and people 20 to 29 years old appeared to be clinically relevant in those who started treatment with a CD4 count between 201 and 350 cells/mm³.

University of Maryland researchers who conducted the CD4-gain analysis noted that most studies of HIV in older adults involve US and European populations. Immunologic responses to ART may differ in sub-Saharan Africa, they suggested, because African populations have higher levels of immune activation reflecting nutritional deficiencies and endemic infections. And most studies of immune response in sub-Saharan individuals are limited to the 12 months after ART starts.

To get a better understanding of how age may affect immune reconstitution after ART begins, they conducted this retrospective analysis of adults who began ART from August 2004 through September 2012 in 157 PEPFAR-funded and AIDSRelief-supported facilities in four central-African countries: Kenya, Nigeria, Tanzania, and Uganda. The 158,160 study participants were at least 20 years old and had never taken an antiretroviral, not even single-dose nevirapine.

The investigators explored CD4 changes in 38,854 people 20 to 29 years old, 63,220 people 30 to 39 years old, 36,959 people 40 to 49 years old, 14,102 people 50 to 59 years old, and 5025 people 60 or older. Median pre-ART CD4 count lay just below 200 cells/mm³ in all five age groups; about one quarter of people in each group had a pretreatment CD4 count between 201 and 350 cells/mm³.

An analysis adjusted for sex, WHO HIV disease stage, functional status, TB, and other opportunistic infections determined that all five age groups starting ART with 200 or fewer CD4 cells/mm³ had steep and similar CD4 gains in the first 6 months of therapy. Then the curves describing further gains diverged: The 20-to-29 group clearly gained more CD4 cells on average than the three middle age groups, whose gains clustered around the same average. The 60-or-older group clearly gained fewer CD4 cells on average than the three middle groups. Compared with 20- to 29-year-olds, people 60 or older gained 22 fewer CD4 cells/mm³ through 12 months, 49 fewer through 24 months, and 63 fewer through 48 months.

The 48-month pattern in people starting ART with 201 to 350 cells/mm³ was similar to the pattern of people starting with a lower CD4 count: Average gains in the 20-to-29 group clearly lay above the average gains of the three middle age groups, which lay above the average of the 60-or-older group (**Figure 6**). Compared with the 20-to-29 group, people 60 or older gained an adjusted average 77 fewer CD4 cells/mm³ through 12 months of ART, 92 fewer through 24 months, and 76 fewer through 48 months.

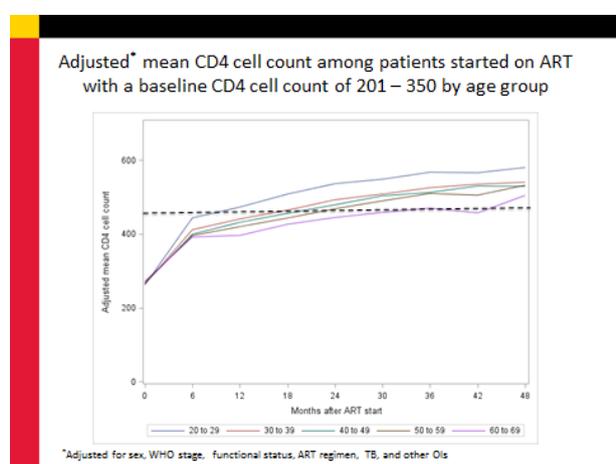


Figure 6. Sub-Saharan Africans 60 years old or older starting ART with 201 to 350 CD4 cells/mm³ gained substantially fewer CD4 cells through 48 months of treatment than four older groups. The dashed line indicates 450 cells/mm³, a proposed cutoff for TB reactivation. (Source: Kristen Stafford, University of Maryland, and colleagues.²²)

Among people starting ART with more than 350 CD4 cells/mm³, CD4 gains through 48 months again proved highest in the 20-to-29 group, with less distinct separation among the older four age groups. Compared with the 20-to-29 contingent, people 60 or older gained an adjusted average 85 fewer CD4 cells/mm³ after 12 months of ART, 154 fewer after 24 months, and 74 fewer after 48 months.

The University of Maryland team argued that age-related differences in CD4-cell gains among the oldest people starting ART with 201 to 350 cells/mm³ are clinically meaningful because delays in achieving the same gains as the youngest group are pronounced and long lasting. For example, on average the 60-or-older group did not reach the 450 cells/mm³ TB-reactivation cutoff until about 30 months after starting ART, whereas the 20-to-29 group reached that level in about 6 months (**Figure 6**). Although age-related differences in CD4 gains are smaller in people starting ART with 200 or fewer CD4 cells/mm³, the delay in achieving thresholds similar to younger patients may be clinically relevant and more important than looking at the mean difference at a particular time point.

The investigators noted that their analysis is limited because data were collected for routine care, not for research, and because they did not have data on duration of HIV infection or virologic response. Sensitivity analyses of excluded patients and missing data yielded results similar to the final models, and a mediation analysis showed adherence did not mediate the association between age and CD4-cell gain over time. The researchers proposed that stratifying the age groups into three pre-ART CD4 clusters provides a more accurate picture of the effect of age on CD4 response than controlling for baseline CD4 count in adjusted models since the mean difference over time differs by baseline CD4 cell count.

The researchers suggested that “the removal of CD4 thresholds for the initiation of ART in resource-limited settings has the potential to mitigate clinically meaningful age-related differences in CD4-cell reconstitution.”

Collagen marker correlates with GALT fibrosis in HIV group

Circulating levels of LOXL2, a marker that crosslinks collagen and elastin, correlated with gut-associated lymphoid tissue (GALT) fibrosis in an analysis of middle-aged people taking antiretroviral therapy (ART).²³ Researchers from the University of California, Los Angeles (UCLA) and colleagues from other centers suggested LOXL2 inhibition may be “a potential therapy for intestinal fibrosis and its sequelae in this population.”

GALT fibrosis occurs early in HIV infection, the UCLA team noted, and persists even in people who respond well to ART. Because the gut is a major T-cell reservoir, that fibrosis may contribute to the incomplete immune reconstitution in ART responders. Biopsy remains the gold standard for quantifying GALT fibrosis but cannot be routinely deployed in clinical practice. Thus these researchers sought correlations between circulating biomarkers of fibrosis and GALT fibrosis in people taking ART. Among these markers was LOXL2, lysyl oxidase-like homolog 2.²⁴ Other markers included hyaluronic acid, PAI-1, CXCL4, TIMP-1, CIC C1q, TGF-β₁, and TGF-β₂.

The researchers recruited antiretroviral-treated people with a viral load below 50 copies/mL and a rectosigmoid biopsy. They used Masson trichrome staining to quantify percent collagen deposition on GALT biopsy and used ELISA or multiplex assays to measure circulating markers of fibrosis. The researchers employed simple regression to analyze relationships between biomarker levels, clinical and demographic variables, lymphoid aggregate collagen deposition, and lymphoid aggregate CD4 density from intestinal biopsies.

The study involved 39 antiretroviral-treated adults in the San Francisco SCOPE cohort. Most were men (92%) and white (59%), and one third had chronic hepatitis virus infection. Median age stood at 48 years (IQR 45 to 55), median time since HIV diagnosis 17 years (IQR 15 to 21), nadir CD4 count 66 cells/mm³ (IQR 18 to 108), and current CD4 count 277 cells/mm³ (IQR 177 to 483), despite an undetectable viral load.

Only LOXL2 correlated with GALT lymphoid aggregate collagen deposition ($r = 0.44$, $P = 0.007$) (**Figure 7**). And only LOXL2 correlated inversely with CD4 count in lymphoid aggregate ($r = -0.32$, $P = 0.05$).

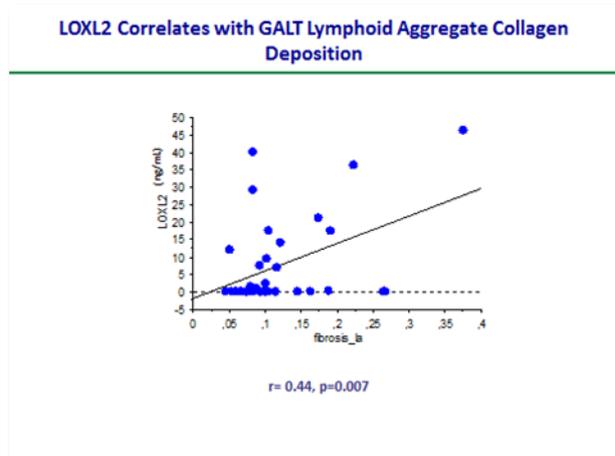


Figure 7. Among numerous fibrosis biomarkers, only LOXL2 correlated with GALT fibrosis in antiretroviral-treated adults with an undetectable viral load. (Source: Jordan Lake, University of California, Los Angeles, and colleagues.²³)

The researchers called for larger studies “to certify the utility of circulating LOXL2 levels as a noninvasive marker for fibrotic tissue burden in treated HIV infection.” Because LOXL2 becomes upregulated only in pathologic states, they suggested assessing LOXL2 inhibition to treat HIV-associated fibrotic disease.

DISABILITY, FALLS, AND FRAILTY

Neurocognitive impairment, lifestyle linked to disability in older HIV+ adults

Neurocognitive impairment plus lifestyle and socioeconomic factors were associated with impairment in Instrumental Activities of Daily Living (IADL) in a study of more than 1000 middle-aged and older US adults with HIV infection.²⁵ HIV-related factors and comorbidities other than neurocognitive impairment were not linked to disability in this analysis.

AIDS Clinical Trials Group (ACTG) investigators who conducted this study noted that an aging HIV population inevitably bears a growing burden of geriatric syndromes, including worsening IADL performance and classically defined frailty. They performed this analysis of ACTG protocol A5322 participants to learn more about the risk factors for these age-related deficits.

A5322 is a prospective observational study of people 40 years old or older who started antiretroviral therapy in an ACTG trial. When people enter A5322, researchers assess IADL, which include 8 categories: housekeeping, money management, cooking, transportation, telephone use, shopping, laundry, and medication management. A person who needs assistance in 1 or more categories is considered IADL-impaired. The investigators used logistic regression to identify factors associated with IADL disability. Frailty evaluation in this group relied on assessment of 5 items: 4-meter walk, grip strength, and three self-reported items: unintentional weight loss in the past year, exhaustion, and low activity. Meeting 3 or more of these criteria indicated frailty; meeting 1 or 2 indicated a prefrail state. The researchers used the ACTG neuroscreen (Trailmaking A and B, Digit Symbol) to evaluate neurocognitive impairment.

The analysis focused on 1015 A5322 participants, 19% of them women, 29% black, 20% Hispanic, 32% on Medicare or Medicaid, and 41% with private insurance. Median age stood at 51 years (standard deviation 7.5), and 15% of cohort members with 60 or older. Two thirds of participants had a CD4 count above 500 cells/mm³, and 94% had a viral load below 200 copies/mL.

IADL analysis indicated that 11% of these people had impairment in 1 IADL and 6% had impairment in 2 or more IADL. Participants needed assistance most often with housekeeping (48%), transportation (36%), shopping (28%), and laundry (20%). Fewer than 10% needed help managing medications. The overall 17% impairment rate in these people with HIV approximated the impairment rate in the general-population NHANES group older than 74. Among younger NHANES members, 6% or fewer needed IADL help.

Logistic regression identified four factors independently associated with higher odds of IADL disability and one factor independently associated with lower odds (**Table 2**).

Table 2. Variables independently associated with IADL impairment

Associated with greater odds of impairment:

- High school education or less: OR 2.22, 95% CI 1.43 to 3.47, $P < 0.001$
- Current or prior smoking: OR 1.55, 95% CI 0.99 to 2.41, $P = 0.05$
- Fewer than 3 days vigorous/moderate activities/week: OR 1.97, 95% CI 1.3 to 2.98, $P = 0.001$
- Neurocognitive impairment: OR 2.28, 95% CI 1.4 to 3.71, $P < 0.001$

Associated with lower odds of impairment:

- Private insurance versus Medicare/Medicaid: OR 0.42, 95% CI 0.25 to 0.7, $P < 0.001$

CI, confidence interval; OR, odds ratio.

Source: Kristine Erlandson, University of Colorado Denver, and ACTG colleagues.²⁵

While 560 A5322 participants (56%) were not frail, 377 (38%) were prefrail and 62 (6%) frail. The researchers found only limited overlap between frailty and IADL-defined disability (weighted kappa = 0.18, 95% CI 0.13 to 0.23).

The ACTG team concluded that middle-aged and older adults with HIV have 2 to 3 times higher rates of IADL disability than the general US population represented in NHANES. Neurocognitive impairment plus socioeconomic and lifestyle factors—but not HIV-related factors—were strongly associated with disability by this measure. The ACTG team suggested several reasons for IADL disability in older people with HIV:

1. Greater difficulty accessing typical resources for older adults, like churches and senior centers, because of stigma related to HIV, gender, or sexual identity, or because of lack of knowledge on how to access these resources.
2. Lack of adult children to assume a caregiver role.
3. Economic challenges resulting from long-term disability (for example, no retirement savings and limited Social Security benefits).

The researchers proposed that modifiable factors like smoking and low physical activity “may be targets for interventions to reduce IADL impairment.”

Cognitive complaints, comorbidities linked to falls in women with/without HIV

Subjective cognitive complaints more than doubled the odds of falls in HIV-positive and negative members of the Women’s Interagency HIV Study (WIHS).²⁶ Comorbid medical illnesses and substance use substantially mediated the impact of cognitive complaints on the odds of falling.

WIHS researchers who conducted this study noted that, compared with HIV-negative people, those with HIV have higher rates of conditions that predispose to falling, such as cognitive impairment, frailty, and polypharmacy. Prior study of the WIHS cohort found increased fracture incidence in middle-aged women with HIV than in HIV-negative women.²⁷ The high prevalence of low bone mineral density in people with HIV could make fractures more likely when they fall.

The WIHS team conducted this new study to determine the frequency of falls in women with and without HIV and to identify fall risk factors. WIHS is an ongoing observational study of HIV-positive women and demographically similar HIV-negative women at 9 sites across the United States. Women make twice-yearly visits for physical exams, interviews, testing, and sample collection.

Starting in 2014 WIHS women were asked to report any falls in the prior 6 months. This analysis focused on fall reports in 3 consecutive study visits. The researchers considered self-reported subjective cognitive impairment the primary predictor, defined as either (1) major problems with memory or concentration interfering with everyday activities and lasting more than 2 weeks, or (2)

confusion, getting lost in familiar places, or inability to perform routine mental tasks. Covariates included demographics, clinical comorbidities, substance use or use of central nervous system (CNS)-active medications, and HIV-specific variables. The WIHS investigators used hierarchical models to evaluate associations between subjective cognitive complaints and having at least 1 fall in the prior 6 months. They sequentially adjusted the analysis for each of the just-listed sets of covariates.

The analysis involved 1289 women with HIV and 587 without HIV. The HIV group was significantly older (median 49 versus 47 years, $P = 0.0002$) but had similar proportions of blacks (72% and 75%), women who completed high school or had more education (66% and 69%, $P = 0.17$), and women who had an annual income of at least \$12,000 (48% and 52%, $P = 0.16$). Women with and without HIV were similar in proportions who reported cognitive complaints (11% in each group) or had depressive symptoms (29% and 30%) but included a higher proportion with neuropathy (20% versus 16%, $P = 0.02$) and a lower proportion with obesity (48% versus 58%, $P < 0.0001$). Current smoking, use of cocaine or heroin, and alcohol use were less prevalent in women with HIV, while HIV-positive women used marijuana significantly more often (24% versus 19%, $P = 0.03$). Most women with HIV (88%) were taking antiretroviral therapy, median current CD4 count measured 586 cells/mm³ (IQR 382 to 777), and 63% had a viral load below 200 copies/mL.

About 18% of women with HIV or without HIV had 1 or more falls in the 6 months before each of the three study visits (**Figure 8**). The overall rate of any fall did not differ significantly between women with and without HIV (35.8% versus 36.8%, $P = 0.7$); nor did rates of falls with injury (13.2% and 15.5%, $P = 0.2$) or falls with fracture (1.9% and 2.2%, $P = 0.7$).

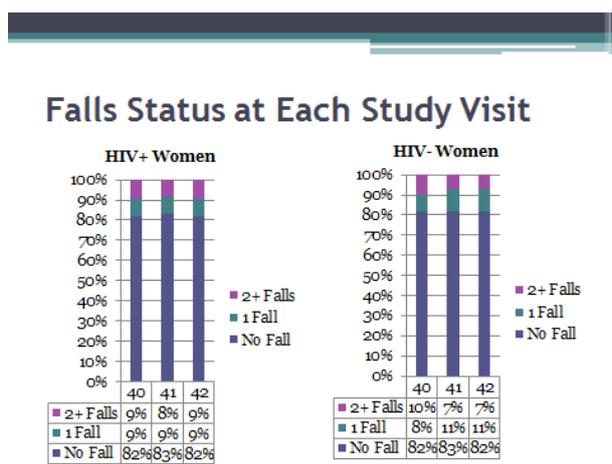


Figure 8. HIV-positive and negative women in the Women’s Interagency HIV Study (WIHS) did not differ in proportions with 1 fall or 2 or more falls in the 6 months before each of three consecutive semiannual visits. (Source: Anjali Sharma, Albert Einstein College of Medicine, New York, and WIHS colleagues.²⁶)

Unadjusted analysis identified several factors associated with any fall, including demographic factors (older age, lower income, earlier WIHS enrollment period, injecting drugs as HIV risk), comorbidities (subjective cognitive complaints, worse quality of life, neuropathy, obesity, depressive symptoms, diabetes, renal impairment, cancer, hypertension, HCV infection), substance use or CNS medications (former or current smoking, use of crack, cocaine, or heroin, former or current injection drug use, former or current marijuana use, heavy alcohol drinking, use of 1, 2 or 3 or more CNS-active medications). HIV infection was not associated with falling.

In the adjusted analysis, subjective cognitive complaints more than doubled the odds of any fall in the whole study group (adjusted odds ratio [aOR] 2.60, 95% CI 2.06 to 3.28, $P < 0.0001$) and in women with HIV alone (aOR 2.30, 95% CI 1.73 to 3.07, $P < 0.0001$). Further adjustment for demographics attenuated the association, which remained significant in the whole study group (aOR 2.30, 95% CI 1.78 to 2.96, $P < 0.0001$) and in women with HIV alone (aOR 2.03, 95% CI 1.49 to 2.76, $P < 0.0001$). After further adjustment for comorbidities, subjective cognitive complaints remained associated with any fall in the whole study group (aOR 1.46, 95% CI 1.12 to 1.91, $P = 0.006$) but not in the HIV group alone (aOR 1.23, 95% CI 0.89 to 1.71, $P = 0.20$). Additional adjustment for substance use or CNS medications further attenuated the association of subjective cognitive complaints and any fall, which remained significant in the whole study group (aOR 1.42, 95% CI 1.08 to 1.86, $P = 0.01$) but not in women with HIV alone (aOR 1.18, 95% CI 0.85 to 1.64, $P = 0.33$).

The WIHS team concluded that falls are common in middle-aged women with HIV, but not more common than in demographically similar HIV-negative women. Among women with HIV, they observed, “the association between cognitive complaints and falls appears to be mediated by comorbid medical illness.” They called for further study to determine which comorbid conditions have the greatest impact on fall risk and whether managing these conditions prevents falls in aging women with HIV.

Frailty and prefrailty linked to falls in aging HIV cohort

Test-determined frailty and prefrailty predicted falls in a US group of aging people with HIV.²⁸ Two individual components of the frailty test, slow walking and weak grip strength, also predicted falls.

Falls are common among aging adults with HIV (see preceding report²⁶) and may occur at younger ages in HIV patients than in the general population.²⁹ Abundant research links falling to greater morbidity and mortality. Frailty is also prevalent in aging HIV populations, but the link between frailty and falls in people with HIV remains incompletely understood.

To address these issues, ACTG investigators conducted this analysis of frailty and falls in the A5322 cohort. A5322 is a longitudinal study of 1035 people 40 years old or older who began antiretroviral therapy in an ACTG trial. A5322 participants have a falls interview every 6 months and complete other questionnaires and evaluations including a neurocognitive assessment once yearly. Every year they also complete a 5-component frailty assessment that includes a 4-meter walk, grip strength, and 3 self-reported items: unintentional weight loss of at least 10 pounds in the past year, exhaustion, and low physical activity level. Meeting 3 or more criteria indicates frailty, while 1 or 2 criteria indicate prefrailty. The ACTG team used multinomial logistic regression to explore associations between frailty and prefrailty (and, in separate models, weak grip strength and slow walk speed) at cohort entry and single or recurrent falls over the next year.

The analysis included 967 people, 19% of them women and 48% non-Hispanic white. The group had a median age of 51 years (IQR 46 to 56), 92% had a viral load below 50 copies/mL, and 68% had a CD4 count above 500 cells/mm³. Among these 967 individuals, 174 (18%) had 1 or more falls over 12 months and 68 (7%) had recurrent falls (2 or more) over 12 months. Among the 174 people with at least 1 fall, 36 (21%) sought medical attention for the fall and 9 (5%) had a fracture.

Fall prevalence rose with age, from 12.7% among 40- to 49-year-olds, to 20.7% among 50- to 59-year-olds, and to 25.8% among people 60 or older ($P = 0.0003$). Falls also were significantly more common in A5322 participants with prefrailty or frailty than in nonfrail individuals (**Figure 9**).

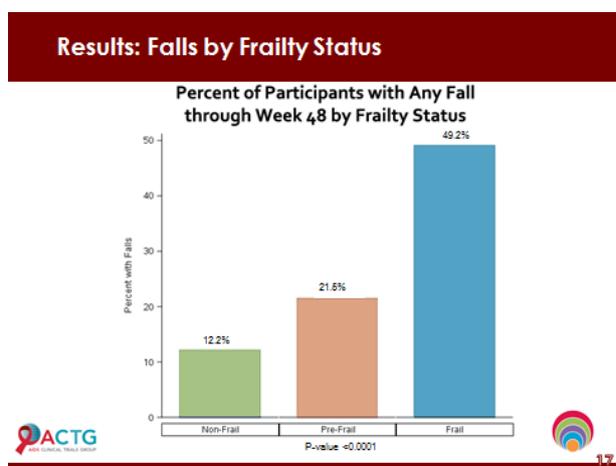


Figure 9. Over a 12-month period, falls were significantly more common in ACTG A5322 cohort members with prefrailty or frailty than in cohort members without frailty. (Source: Katherine Tassiopoulos, Harvard School of Public Health, and ACTG A5322 colleagues.²⁸)

Frailty and prefrailty were associated with recurrent falls (aOR 18.6, 95% CI 7.6 to 45.3, for frailty; aOR 3.78, 95% CI 1.86 to 7.69, for prefrailty) and marginally with single falls (aOR 2.38, 95% CI 0.89 to 6.36, for frailty; aOR 1.51, 95% CI 0.95 to 2.43, for prefrailty). Slow walk speed and weak grip

strength were associated with recurrent falls (aOR 2.9, 95% CI 1.57 to 5.36, for walk speed; aOR 3.86, 95% CI 2.25 to 6.63, for grip strength), but not with single falls.

The ACTG team proposed that frailty, prefrailty, and each of the two individual components of frailty (weak grip and slow walking) may be useful predictors of recurrent falls. They suggested that incorporating these frailty assessments into clinical practice “may help identify older HIV-infected individuals at increased risk for falls” who may “benefit from falls prevention interventions.”

T-cell reactivity to CMV correlates with inflammation in HIV+/HIV- MSM

T-cell responses to cytomegalovirus (CMV) were broad in HIV-positive and negative men who have sex with men (MSM) in the Multicenter AIDS Cohort Study (MACS).³⁰ Responses to CMV correlated with systemic inflammation and immune activation in certain subgroups defined by HIV serostatus and frailty.

MACS investigators who conducted this study observed that CMV is highly prevalent in older HIV-positive populations, and that CMV-specific T cells persist at high levels in HIV-positive individuals responding virologically to antiretroviral therapy.³¹ They added that CMV positivity correlates with frailty and mortality, and that both frailty and treated HIV infection are associated with ongoing immune activation. Prior work in the MACS found that frailty occurs at an earlier age in HIV-positive than HIV-negative MSM.³² But no research has explored potential correlations between total T-cell response to CMV and systemic immune activation.

A MACS team hypothesized that T-cell reactivity to CMV in virologically suppressed HIV-positive MSM and in HIV-negative MSM (1) contributes to systemic inflammation and immune activation and (2) is related to frailty. To test those hypotheses, they focused on 22 HIV-positive and 20 HIV-negative MSM in the Baltimore/DC MACS group. The investigators rated participants as frail if they met 3 of 5 criteria on two consecutive visits: slow 4-meter walk, weak grip strength, and self-reported weight loss, low energy, or low physical activity. Men with HIV had virologic control with antiretroviral therapy. Twenty-one men were frail and 21 nonfrail.

All study participants had detectable anti-CMV responses, and specific peptide pools eliciting responses varied widely from man to man. The study confirmed strong and significant correlations between anti-CMV CD4-cell responses and inflammation markers in HIV-positive and negative men without frailty. Strong correlations in HIV-positive men involved MCP-4 ($r = 0.63$), IL-6 ($r = 0.63$), C-reactive protein ($r = 0.60$), and activated CD4 cells ($r = 0.60$). HIV-positive frail men had strong correlations between anti-CMV responses and the sum of CD8-cell TNF-alpha responses ($r = 0.71$), the sum of CD8-cell IFN-gamma responses ($r = 0.61$), the sum of CD8 IL-2 responses ($r = 0.87$), and the sum of all CD8-cell responses ($r = 0.70$).

The MACS investigators proposed that their findings “are consistent with the hypothesis that T-cell responses to CMV could explain much of the immune activation in nonfrail treated HIV-positive and HIV-negative men.” They caution, though, that the correlations observed could result from immune activation due to other causes.

AGING AND COGNITION

Excess mitochondrial DNA damage in brain of adults with HIV

Postmortem analysis of brain from middle-aged adults with or without HIV infection verified reduced mitochondrial (mt)DNA content and increased mtDNA deletions in the HIV group.³³ Reduced mtDNA content was associated with markers of HIV disease activity.

Researchers from Newcastle University who conducted the mtDNA study noted that cognitive impairment persists in people with HIV despite viral suppression in blood. Reasons for this impairment remain poorly understood. HIV patients taking older nucleoside analogs had mtDNA depletion in blood, adipose tissue, and muscle cells, but few people use the implicated nucleosides today.

Changes in mtDNA content could reflect normal aging and age-related conditions. Work published in 2011 by the same investigators detected excess age-associated mtDNA mutations in skeletal muscle of antiretroviral-treated people.³⁴ Because mtDNA mutations have a similar natural history in muscle and brain, the Newcastle team hypothesized that people with HIV may have mtDNA changes in brain that could contribute to decreased cognitive function.

The researchers obtained frontal cortex samples from brain biobanks in the United States and the United Kingdom, including samples from 25 people with HIV and 10 without HIV. They excluded samples from anyone who had brain opportunistic infections. All individuals with brain samples had neurocognitive testing within approximately 6 months of death. The investigators used multiplex qPCR to determine mtDNA copy number and prevalence of the delta4977 mtDNA common deletion.

mtDNA copy number was significantly lower in people with HIV than in the HIV-negative group (mean 364 +/- 39 versus 706 +/- 68 copies/cell, $P < 0.001$) (**Figure 10**). The 25 people with HIV averaged 47.7 years of age at death, compared with 40.5 years among controls. mtDNA content was not associated with age at death in either HIV cases or HIV-negative controls, a finding indicating that age at death was not a confounder in this analysis. mtDNA copy number was not associated with use of d-nucleosides either at the time of death or in the past.

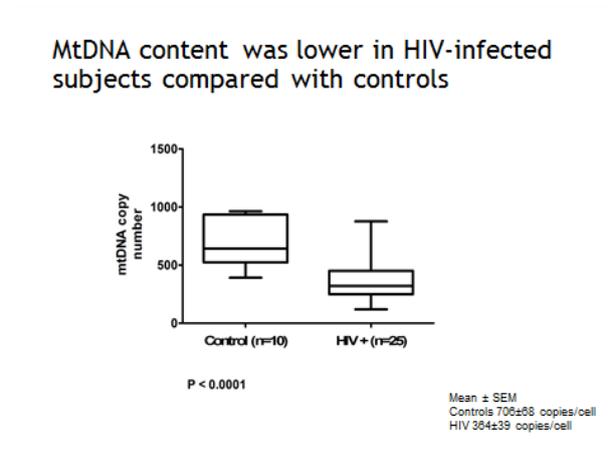


Figure 10. Mitochondrial DNA content proved significantly lower in postmortem brain samples of people with HIV than in an HIV-negative control group, a finding that may offer a clue to the mechanism of neurocognitive impairment in people with HIV. (Source: Carla Roca, Newcastle University, and colleagues.³³)

In the 25 people with HIV infection, mtDNA copy number correlated positively with CD4 count at the time of death—the higher the copy number the higher the CD4 count ($r = 0.65$, $P = 0.0007$). The 9 HIV-positive people with a plasma viral load below 1000 copies/mL had significantly higher mtDNA content than the 16 people with a higher viral load (482 +/- 86 versus 295 +/- 31 copies/cell, $P = 0.02$). Among people with HIV, mtDNA copy number at death did not vary significantly by neurological diagnosis at death: 20% were normal, 16% had subclinical neuropsychological impairment, 32% had mild cognitive disorder, and 32% had HIV-associated dementia.

The mtDNA common deletion mutation proved significantly more prevalent in brain of people with HIV than in the HIV-negative group ($P = 0.03$). Prevalence of the common deletion was nonsignificantly associated with older age at death in cases and controls (for the HIV group, $r = 0.37$, $P = 0.07$; for HIV-negative controls, $r = 0.11$, $P = 0.76$). mtDNA common deletion prevalence did not vary significantly between people who took a d-nucleoside and those who did not, and common deletion prevalence did not vary by degree of neurocognitive impairment.

The Newcastle investigators concluded that brain of people with HIV has excess mtDNA damage reflected in lower mtDNA content and higher prevalence of the mtDNA common deletion mutation. They proposed that HIV infection, and not d-nucleoside therapy, explains these findings because reduced mtDNA content in brain was associated with lower CD4 count and higher viral load. Because neuronal integrity depends on mitochondrial function, the researchers believe their findings are “consistent with virally mediated neuronal damage.”

High and low brain permeability predict neurocognitive impairment with HIV

Neurocognitive impairment in HIV-positive people taking antiretroviral therapy was associated with either high blood-brain barrier (BBB) permeability—or low permeability.³⁵ High BBB permeability could allow HIV and other neurotoxins to pass more readily into the central nervous system (CNS), leading to neurologic damage. But low permeability could have a negative impact by restricting penetration of antiretrovirals.

Because albumin typically cannot penetrate the CNS, cerebrospinal fluid (CSF)-to-serum albumin ratios (CSARs) offer one measure of BBB permeability: the higher the CSAR, the greater the permeability. Because few studies have used CSARs to compare disease-related outcomes in people with HIV infection, CHARTER Cohort investigators conducted this study.

CHARTER is an ongoing US observational study focused on neurocognition in people with HIV infection.³⁶ This analysis involved cohort members who had taken antiretroviral therapy for at least 1 month and did not have a comorbidity that would confound study findings. All participants underwent comprehensive neuromedical and neuropsychological assessments and gave samples for albumin measurement in blood plasma and CSF. The researchers defined neurocognitive impairment as a global clinical rating of 5 or greater.

Of the 169 study participants, 132 (78%) were men, 74 (44%) were non-Hispanic white, and median age stood at 44 years (IQR 39 to 48.5). Participants had taken antiretroviral therapy for a median of 8.8 months. Median current CD4 count was 410 cells/mm³ and median nadir 97 cells/mm³. HIV RNA was below 50 copies/mL in 61% of plasma samples and in 86% of CSF samples. Eighty-one participants (48%) had global neurocognitive impairment.

Median CSAR in the study group measured 4.0 (IQR 3.2 to 5.1) (**Figure 11**). An antiretroviral CNS penetration effectiveness (CPE) score below 7 was associated with CSF HIV RNA above 50 copies/mL only in people with lower CSARs, that is, below 3.75 (52.4% versus 10.0%, RR 5.23, $P = 0.0002$) but not in those with a CSAR of 3.75 or greater (9.4% versus 7.6%, $P > 0.20$). Recursive partitioning identified two CSAR threshold values that defined three CSAR groups: low (below 3.5), mid (3.5 to 6.1), and high (equal to or above 6.1). The low and high CSAR groups included significantly higher proportions of people with neurocognitive impairment (56% and 65%) than did the mid-CSAR group (38%) ($P < 0.05$) (**Figure 11**).

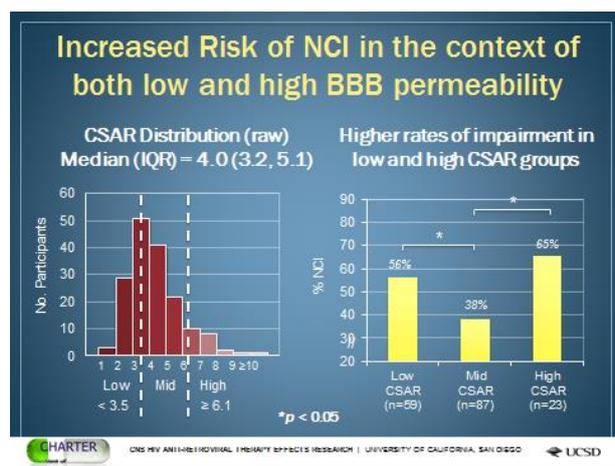


Figure 11. CHARTER participants with low or high blood-brain barrier (BBB) permeability, indicated by lower or higher CSF-to-serum albumin ratios (CSARs), had higher rates of neurocognitive impairment than participants with midrange CSARs. (Source: Jennifer Iudicello, University of California, San Diego, and CHARTER colleagues.³⁵)

In multivariable linear regression analysis, this tripartite CSAR variable independently predicted neurocognitive impairment ($P = 0.05$), as did older age, higher antiretroviral CPE score, higher creatinine, HCV positivity, and Hispanic ethnicity (all $P < 0.05$). When the researchers restricted the analysis to the low and high CSAR groups, neurocognitive impairment was associated with Hispanic ethnicity, higher systolic blood pressure, higher creatinine levels, higher CPE score, and HCV infection (all $P < 0.05$).

Univariable analyses within the low CSAR group revealed significant associations between neurocognitive impairment and Hispanic ethnicity, diabetes, HCV seropositivity, lower nadir CD4 count, and higher serum glucose levels (all $P < 0.05$). Neurocognitive impairment in the high CSAR group was significantly associated with a longer duration of current antiretroviral regimen, the presence of medical and psychiatric comorbidities, higher systolic blood pressure, and older age (all $P < 0.05$).

The researchers suggested these findings appear to indicate that risk factors for HIV-related neurocognitive impairment may vary depending on the permeability of the BBB. Neurocognitive impairment in the context of high BBB permeability may be related to factors such as vascular or metabolic variables (for example, high blood pressure, lipids, and creatinine), chronic inflammation, and possibly antiretroviral neurotoxicity. While there may also be a vascular contribution to impairment in the low CSAR context, in contrast, neurocognitive impairment in the context of low BBB permeability may be more closely linked to other factors, such as HCV infection, antiretroviral ineffectiveness, and/or HIV-associated damage before starting ART.

The researchers proposed that BBB biomarkers like the CSAR may “provide insight into the variability in patients’ response to ART and guidance for the development of individualized treatment regimens that may more effectively improve disease outcomes.”

Immune response to CMV tied to lower CD4 nadir, worse neurocognitive performance

A stronger immune response to CMV was associated with older age and lower nadir CD4 count in an analysis of 138 CHARTER Cohort members.³⁷ High anti-CMV IgG was also associated with worse neurocognitive performance in these individuals.

Nearly all adults with HIV infection also carry CMV, which has been linked to worse neurocognitive function in HIV-positive people older than 65.³⁸ Among 1204 HIV-negative adults older than 60 in the Sacramento Area Latino Study on Aging (SALSA), CMV was associated with neurocognitive decline over 4 years. And in people with HIV infection, CMV-specific T-cell activation has been linked to vascular disease.³⁹ To get a better understanding of potential associations between CMV and age, HIV load, and neurocognitive performance, CHARTER researchers conducted this study.

The study focused on 138 CHARTER members specifically selected to vary by nadir CD4 count, antiretroviral use, and neurocognitive impairment. No one had a severe neuropsychiatric comorbidity. All participants underwent a comprehensive neuropsychological battery and immunoassays for anti-CMV IgG and markers of immune activation (sCD14, sCD163, CCL2, CXCL10, TNF-alpha, IL-6, IFN-gamma).

Study participants had a median age of 43 years (IQR 39 to 49) and included 26 women (19%) and 64 whites (46%). Median CD4 nadir measured 256 cells/mm³ (IQR 81 to 365) and current CD4 count 436 cells/mm³ (IQR 270 to 634). Most participants taking ART had a viral load below 50 copies/mL in CSF (83%) but not in plasma (45%). While 42% of participants had moderate neuropsychiatric comorbidity, the rest had mild or no neuropsychiatric conditions.

Older age correlated with higher anti-CMV IgG ($r = 0.24$, $P = 0.005$), as did lower CD4 nadir ($r = -0.40$, $P < 0.0001$), an AIDS diagnosis, antiretroviral use, and higher serum globulins (**Figure 12**). Higher anti-CMV IgG in serum correlated with higher HIV RNA in CSF ($r = 0.29$, $P = 0.04$).

Older Age and Lower Nadir CD4+ T-Cell Counts Correlated with Higher Anti-CMV IgG Levels

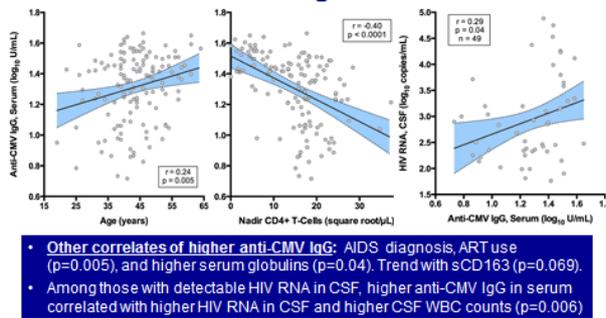


Figure 12. In a 138-person CHARTER Cohort group, older age and lower nadir CD4 count correlated with a stronger immune response to CMV. (Source: Scott Letendre, University of California, San Diego, and CHARTER colleagues.³⁷)

In multivariate analysis higher anti-CMV IgG was associated with worse neurocognitive performance (beta = -3.5). In the same analysis, significant associations with worse neurocognitive performance were neuropsychiatric comorbidity (beta = -1.2, $P = 0.017$), Hispanic or white ethnicity (beta = -1.7, $P = 0.0006$), longer HIV infection duration (beta = -2.7, $P = 0.003$), lower systolic blood pressure (beta = 0.09, $P = 0.006$), and the inflammation/activation marker CCL2 in CSF (beta = -7.9, $P = 0.02$).

This model included an interaction between anti-CMV IgG and CCL2 ($P = 0.02$): among adults with lower anti-CMV IgG levels ($< 1.60 \log_{10} \text{ U/mL}$), those with higher CCL2 levels in CSF ($\geq 3.04 \log_{10} \text{ pg/mL}$) had worse neurocognitive performance than those with lower CCL2 levels ($P = 0.004$). Participants who had anti-CMV IgG levels $\geq 1.60 \log_{10} \text{ U/mL}$ performed the worst, irrespective of CCL2 level. In an analysis limited to the 41 adults with undetectable HIV RNA in plasma, only higher anti-CMV IgG was associated with worse neurocognitive performance (beta = -12.8, $P = 0.0026$).

The CHARTER team concluded that “older age and more advanced past immune suppression are associated with a stronger immune response to CMV, which appears to influence HIV pathogenesis in the brain.” Because the association between immune response to CMV and neurocognitive performance was stronger in people with an undetectable HIV load in plasma, the researchers suggested these effects “may be particularly important when HIV-positive adults are taking suppressive ART.”

Advancing age, lower CD4s, HCV tied to neurologic impairment in ACTG group

Advancing age, lower CD4 count, less education, and HCV positivity were associated with higher odds of neurologic impairment in the 3300-person ACTG ALLRT cohort.⁴⁰ After adjustment for age alone, overall neurocognitive scores improved in the years after this relatively young cohort started antiretroviral therapy (ART).

Researchers who conducted this study observed that neurocognitive trajectories may decline in people otherwise responding well to ART. They undertook this analysis to explore associations between advancing age, other variables, and neurocognitive trajectory in middle-aged people starting ART in an ACTG trial.

The ACTG Longitudinal Linked Randomized Trials (ALLRT) was a prospective cohort study involving 3313 initially antiretroviral-naïve adults who started their first antiretroviral regimen in one of 7 ACTG trials. Participants underwent annual neurocognitive testing with four tests: Trail Making Tests A and B, the WAIS-R Digit Symbol subtest, and the Hopkins Verbal Learning Test. The investigators assessed overall performance by comparison with normative data from HIV-negative people summarized as a Z-score across the 4 tests (NPZ-4). They defined impairment as a score ≤ -2.0 standard deviation (SD) on one test or ≤ -1.0 SD on two tests. They considered test results collected 2 to 9 years after participants began ART. The investigators used multivariable regression models based on generalized estimating equations to identify predictors of NPZ-4 outcome.

Participants had a median baseline age of 38 years (IQR 31 to 45) and a median nadir CD4 count of

221 cells/mm³ (IQR 80 to 324). Most cohort members, 80%, were men, 36% black, and 22% Hispanic. At their first Hopkins Verbal Learning Test or 2 years after starting ART, 91% of participants had a viral load below 200 copies/mL, 21% had a CD4 count below 351 cells/mm³, 25% had a count between 351 and 500 cells/mm³, and 54% had a count above 500 cells/mm³. At that point the overall NPZ-4 score averaged -0.23 and 39% of participants had NPZ-4 impairment.

Through 768 weeks of follow-up, an analysis adjusted for age determined that neurocognitive performance improved from year to year (**Figure 13**). But in the completely adjusted analysis, every decade of age was associated with nearly 20% higher odds of neurocognitive impairment (aOR 1.18, 95% CI 1.1 to 1.25, $P < 0.001$). Other variables independently associated with greater odds of neurocognitive impairment were time-varying CD4 count of 0 to 350 cells/mm³ versus above 500 (aOR 1.21, 95% CI 1.05 to 1.39, $P = 0.009$), time-varying CD4 count of 351 to 500 cells/mm³ versus above 500 (aOR 1.12, 95% CI 1.01 to 1.24, $P = 0.03$), 12 or fewer years of education (aOR 1.36, 95% CI 1.19 to 1.56, $P < 0.001$), and HCV positivity (aOR 1.42, 95% CI 1.09 to 1.85, $P = 0.009$). This analysis found no association between injection drug use and neurocognitive impairment.

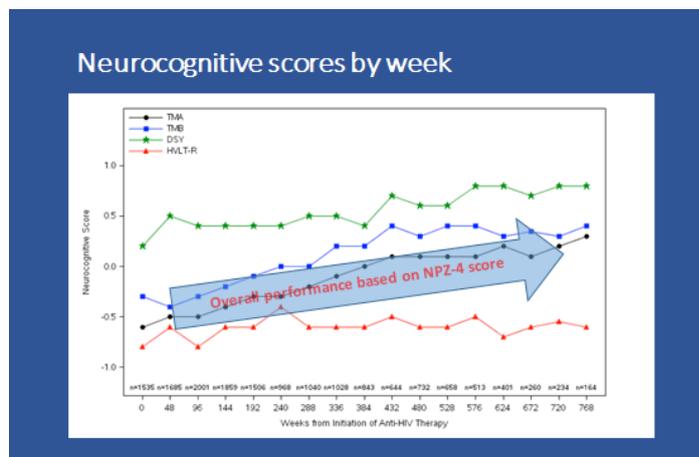


Figure 13. An age-adjusted analysis of 3313 antiretroviral-treated adults in the ALLRT cohort found an overall increase in neurocognitive scores through more than 700 weeks of follow-up. But in a fully adjusted analysis, advancing age, lower CD4 count, and other variables were associated with greater odds of neurocognitive impairment. (Source: Hamza Coban, University of California, San Diego, and colleagues.⁴⁰)

The researchers concluded that, “despite continued virologic suppression and neurocognitive improvement in the cohort as a whole, older individuals tended to have worse neurocognitive performance, after consideration of concurrent predictors, than younger individuals.” They suggested that future studies explore potential mediators of aging’s impact on neurocognitive performance, such as inflammation and vascular risk factors.

CSF iron status implicated in neurocognitive function of HIV+ adults

Higher cerebrospinal fluid (CSF) H-ferritin, an iron-storage protein, predicted better and improving neurocognitive function in HIV-positive people younger than 50 years old.⁴¹ CSF transferrin, the main iron transporter in the central nervous system, predicted better neurocognitive function over time in HIV-positive people aged 50 or older who carry the *APOE-ε4* genotype.

Abnormal iron transport in the brain is a well-appreciated consequence of aging and may contribute to neurocognitive disorders in HIV-positive people, noted Cleveland Clinic researchers and colleagues from other sites who conducted the new study. Research has linked the *APOE-ε4* allele, which confers risk for Alzheimer’s dementia, to greater neurocognitive impairment in older HIV-positive individuals. The investigators hypothesized that “CSF transferrin and H-ferritin are independently associated with changes in neurocognitive function over time in HIV-positive adults on ART” and that “these effects differ by age and *APOE-ε4* genotype.”

To test these hypotheses, the researchers measured iron, transferrin, and H-ferritin levels in baseline CSF from 403 participants in the observational CHARTER study. These CHARTER participants had comprehensive neurocognitive testing (including 15 measures and 7 ability domains) at baseline and 6-month intervals to yield a Global Deficit Score. The research team excluded people with severe medical conditions that would make it difficult to attribute neurocognitive impairment to HIV infection but included individuals with mild or moderate comorbidities. *APOE-ε4* genotype information, available in all participants, was also incorporated into analyses. Of the 403 participants, 157 completed follow-

up at 30 months, 131 of the 157 completed 36 months, and 110 of the 131 completed 42 months.

Among people who completed at least 30 months of follow-up, age averaged 45 years, CD4 count averaged 468 cells/mm³, 12% were women, and 74% were on ART. At baseline, 36 months, and 42 months, 22%, 32%, and 31% of participants were 50 years or older; 68%, 72%, and 67% had undetectable HIV RNA in CSF; and 32%, 33%, and 33% carried the *APOE-ε4* allele. At last follow-up, neurocognitive function had improved in 16%, remained stable in 64%, and declined in 20%.

At the baseline visit participants aged 50 or older had significantly higher CSF H-ferritin than younger people (3.1 versus 2.4 ng/mL, $P = 0.004$) and significantly higher transferrin (19.2 versus 16.8 ng/mL, $P = 0.041$) (**Figure 14**). CSF levels of H-ferritin and transferrin were also significantly higher in men than women. In an analysis adjusted for age, comorbidity, *APOE-ε4* allele, and zidovudine use (which affects iron transport), each unit higher baseline H-ferritin was associated with a 13% higher likelihood of neurocognitive improvement (versus stable status) at last follow-up (relative risk [RR] 1.13, 95% CI 1.01 to 1.26, $P = 0.03$) People with improving neurocognitive function also had nonsignificantly higher baseline H-ferritin than those with declining neurocognitive function (unadjusted $P = 0.17$).

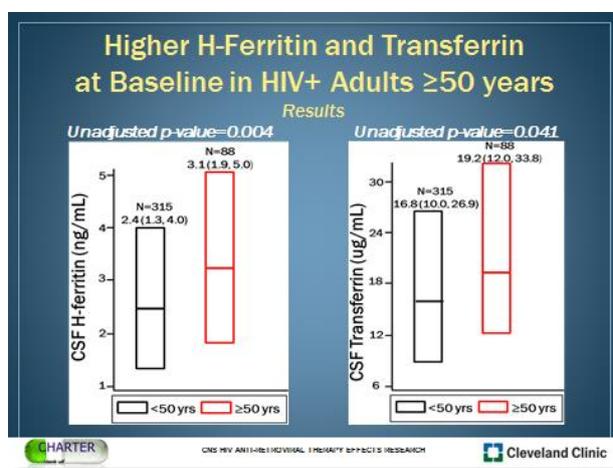


Figure 14. Levels of H-ferritin (an iron-storage protein that can also release iron) and transferrin (the principal iron transporter in the central nervous system) in cerebrospinal fluid were significantly higher in HIV-positive adults 50 years old or older than in younger people with HIV. (Source: Harpreet Kaur, Lerner Research Institute, Cleveland Clinic, and colleagues.⁴¹)

Baseline CSF transferrin and H-ferritin were associated with neurocognitive function measured by Global Deficit Score at 30, 36, and 42 months. Associations between transferrin and better function remained significant after adjustment for comorbidity ($P < 0.05$ at all time points).

Among participants younger than 50 years old, after a median follow-up of 36 months each unit higher baseline H-ferritin was associated with a 17% increased likelihood of improved versus stable status at the last follow-up visit (RR 1.17, 95% CI 1.03 to 1.33, $P = 0.01$). In this under-50 group, baseline H-ferritin was also significantly associated with improved neurocognitive function at 30, 36, and 42 months of follow-up, accounting for 0.7% to 1.3% of the variation in Global Deficit Score across all visits.

Among participants aged 50 years or older who were also *APOE-ε4* carriers, higher CSF transferrin at baseline predicted better neurocognitive function at 30 months even after adjustment for zidovudine use, explaining 11.6% of the variation in Global Deficit Score across visits.

The researchers concluded that higher baseline H-ferritin in CSF predicts better or improved neurocognitive function over 30 to 42 months of follow-up in HIV-positive people younger than 50 years old. They suggested that baseline CSF transferrin levels may predict improving neurocognitive function over time in HIV-positive people 50 or older who carry the *APOE-ε4* allele. The investigators noted that larger studies with more older people are needed “to better understand potential interactions between age, *APOE* genotype, comorbid conditions, and CSF iron status in promoting neurocognitive decline.”

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