Neurocognitive impairment in well controlled HIV-infected patients. A cross sectional study

Running Head: *Neurocognitive impairment in HIV-patients on ART*

Keywords: Neurocognitive impairment; HIV; Interleukin-6; Bacterial translocation; Subclinical atherosclerosis; Delayed recall.

Authors: Irene Portilla^{1,2,3}, Sergio Reus^{1,2,4}, Rafael León¹, Carlos van-der Hofstad^{2,5,6}, José Sánchez^{2,7}, Nicolás López^{2,8}, Vicente Boix^{1,2,4}, Esperanza Merino^{1,2}, Joaquín Portilla^{1,2,3}

Affiliations:

¹ Department of Infectious Diseases, General University Hospital of Alicante, Alicante, Spain

² Alicante Institute for Health and Biomedical Research (ISABIAL – FISABIO Foundation), Spain

³ Department of Health Psychology, Alicante University, Spain

⁴ Department of Clinical Medicine, Miguel Hernández University, Alicante, Spain

⁵ Department of Clinical Psychology, General University Hospital of Alicante, Alicante, Spain

⁶ Department of Health Psychology, Miguel Hernández University, Alicante, Spain

⁷ Department of Epidemiology and Public Health, General University Hospital of Alicante, Alicante, Spain

⁸ Department of Neurology, General University Hospital of Alicante, Alicante, Spain

Corresponding author:

Joaquín Portilla

Unit of Infectious Diseases

Department of Internal Medicine

Hospital General Universitario de Alicante

Pintor Baeza s/n

Alicante, 03010

España

Phone number: 34-659477426

Fax number: +34-965913565

E-mail: portilla_joa@gva.es

ABSTRACT

Background. The reported prevalence of HIV-associated neurocognitive disorders in HIV-people depends on the population studied and the methodology used. We analyze the prevalence of **neurocognitive impairment (NCI)** and associated factors in patients on successful antiretroviral therapy (ART), without comorbidities.

Material and methods. Cross-sectional observational study in HIV-subjects, ≥ 18 years-old, on stable ART and HIV viral load < 50 copies/mL. Patients with medical or psychiatric comorbidities and substance abuse were excluded. **NCI** was diagnosed using Frascati criteria, examining seven neurocognitive domains. We analyzed the association between **NCI** and: HIV-related clinical variables, carotid intima media thickness, bacterial translocation, and plasma inflammatory biomarkers (soluble CD14, Interleukin-6 [IL-6], and tumor necrosis factor- α). The prevalence of **NCI** was calculated with a 95%CI. We fitted a logistic regression model to assess the strength of the associations.

Results. Eighty-four patients were included with an observed **NCI** prevalence of 29.8% (95%CI: 21.0-40.2): 19% had asymptomatic neurocognitive impairment; 8.3%, mild neurocognitive disorder; 2.4%, HIV-associated dementia. Delayed recall was the most commonly affected neurocognitive domain (27.4%). People diagnosed at least 10 years ago (OR:6.5, 95%CI: 1.6-21.7) and those with IL-6 levels above 1.8 pg/mL (OR:6.0, 95%CI 1.1-31.3.3) showed higher odds of **NCI** in adjusted analyses. Participants with carotid plaques had lower scores for delayed recall: -0.9 ± 1.1 versus -0.2 ± 1.1 ($p=0.04$).

Conclusions. Prevalence of **NCI** is high in otherwise healthy adults with HIV-infection. In this population, more than 10 years since HIV diagnosis and high IL-6 levels are associated with **NCI**. Delayed recall neurocognitive domain is worse in patients with subclinical atherosclerosis.

Introduction

HIV-related neurocognitive disorder (HAND) is associated with poor adherence to antiretroviral therapy (ART), difficulties in performing activities of daily living, lower quality of life, and higher mortality in people living with HIV (PLWHIV)^{1,2}.

Reports of **neurocognitive impairment (NCI)** prevalence in PLWHIV are discordant³⁻¹⁰. Most studies have been done on heterogeneous populations, with different **level** of HIV infection control or with comorbidities that may increase the prevalence of neurocognitive disorder⁸⁻¹⁰. NCI prevalence variability also depends on the demographic characteristics and the geographical region where is analyzed^{11,12}.

In developed countries with easy access to ART, most PLWHIV with good adherence to ART maintain long-term suppression of HIV-viremia and high CD4+ lymphocyte counts, enjoying a life expectancy that almost equals that of the general population^{13,14}. Few studies have analyzed **NCI** prevalence in PLWHIV with a good response to ART, and even fewer have included participants without co-morbidities or behavioral factors that could act as confounders in HAND diagnosis⁵⁻⁷.

Reported risk factors for NCI in PLWHIV include previous AIDS diagnosis, low CD4+ nadir, and treatment with antiretroviral drugs with poor penetration in the central nervous system (CNS)¹⁵⁻¹⁶. In recent years, some authors have suggested that systemic inflammation¹⁷⁻²¹, bacterial translocation (BT)^{22,23} and cardiovascular diseases^{24,25} are associated to worse neurocognitive function and may contribute to **NCI**. In PLWHIV receiving successful ART, the predominant hypothesis of ongoing CNS dysfunction is the presence of persistent inflammation³. Bacterial translocation has been proposed as a cause of monocyte activation and CNS inflammation in PLWHIV¹⁷.

The aims of our study were to describe **NCI** prevalence in otherwise healthy PLWHIV **who are receiving long-term effective ART and maintain a long-term HIV viral suppression. Secondly**, to study the potential association between neurocognitive disorders and systemic inflammation biomarkers, BT and subclinical atherosclerosis.

Patients and methods

Design

Cross-sectional observational study.

Participants

Inclusion criteria were: HIV infection, ≥ 18 years-old, unchanged ART for at least one year and HIV-VL < 50 copies/mL over the previous 12 months. Exclusion criteria were: self-reported poor adherence to ART ($< 95\%$ of antiretroviral drugs doses during previous month), smoking (≥ 10 cigarette/day) or substance abuse (alcohol, methadone, psychotropic and recreational drugs) **during last 6 months**; traumatic brain injury **in the last 6 months**, and **presence of comorbidities**: dementia or other CNS diseases, psychiatric illness, viral chronic hepatitis, active cancer or infection, diabetes mellitus, high blood pressure, cardiovascular disease, hypothyroidism or malnutrition). **Finally**, mental or physical impairment that could hinder participants ability to complete or understand the study questionnaires.

After inform consent was signed, a clinician and a neuropsychologist reviewed all electronic clinical reports shared with psychiatrists and family doctors and performed a structured clinical **interview to ensure the fidelity to inclusion/exclusion criteria prior to the administration of neuropsychological tests**. In addition, participants self-completed the Hospital Anxiety and Depression Scale (HADS) questionnaire. We excluded all patients with previous psychiatric illness or a HADS score of more than 10 points.

In a second visit, participants completed a comprehensive neurocognitive test battery, covering seven cognitive domains which is recommended in international consensus, using two tests for each ND: attention/working memory (WAIS-IV Digit Span and D2, Test of Attention); executive function (Trail making Test Form A, Five Digit Test and The Tower of London Test); verbal fluency (PMR and animals); speed of information processing (Trail making Test Form A and WAIS-IV Digit Symbol); learning (TAVEC - Consistent Long-Term Retrieval- and Brief Visuospatial Memory Test-Revised-Total Learning); delayed recall (TAVEC- delayed recall and Brief Visuospatial Memory Test-

Revised (Delayed Recall); and motor function (Grooved Pegboard Test and The MacQuarrie Test)²⁶. All tests were used in their Spanish versions. Raw scores were converted into a Z-score that was demographically corrected for age, education level and sex. To convert raw scores in Z-score we used Spanish normative data. The same neuropsychologist conducted the battery of tests in all patients.

The study was carried on in the Infectious Diseases Unit of the General University Hospital of Alicante, which attends approximately 1250 PLWHIV. All patients in usual care between October 2014 and February 2015 and meeting inclusion criteria were invited to participate in the study. Only 8 patients, who complied inclusion criteria, refused to perform the neuropsychological tests, mainly because of lack of time.

Variables

Outcome variable

Neurocognitive impairment was diagnosed using the Frascati criteria, and participants were classified as having asymptomatic neurocognitive impairment (ANI), minor neurocognitive disorder (MND) or HIV-associated dementia (HAD)²⁷.

We defined ANI as a neuropsychological test score of less than 1.0 standard deviation (SD) below the demographically corrected mean on at least two ability domains, together with no interference in daily functioning. We considered MND if test scores of less than 1.0 SD below the mean in at least two disability domains, plus at least mild interference with daily functioning. HAD was defined by a score of ≥ 2 SDs below the mean on at least two domains, along with marked interference with daily function.

To assess functional impairment and interference in daily life, we asked patients or close people if cognitive impairment interfered, at least mildly, in one of the following functions: mental acuity, work, household tasks or social activities²⁷. We also analyzed each neurocognitive domain (ND) separately and independently of NCI diagnosis or not. When the score of the cognitive domain was less than 1.0 standard deviation (SD) below the demographically corrected mean, it was defined as cognitive domain impairment.

Explanatory variables

Demographic variables were sex, age, education level, route of HIV transmission, and smoking. HIV-related variables included HIV clinical stage, time since HIV diagnosis, time receiving ART, current and nadir CD4+ lymphocyte count, and HIV-VL. Inflammatory biomarkers were plasma levels of soluble CD14 (sCD14), interleukin-6 (IL-6) and tumor necrosis factor alpha (TNF- α); these were determined by ELISA following the manufacturer's instructions (R&D Systems). Bacterial DNA (rDNA) was amplified by PCR of the 16S ribosomal gene²⁸. Carotid ultrasound was performed by a single sonographer, blind to clinical and laboratory data, using a CV 50 PHILIPS ultrasound device with a high frequency linear probe (L12-3). Carotid intima media thickness (CIMT) was measured twice in both sides, gathered on the same day, in the carotid (common and internal) and carotid bulb, and was reported as the average of mean CIMT values. Subclinical atherosclerosis was diagnosed if common-CIMT was greater than 0.8 mm or by the presence of a carotid plaque, defined as: focal wall thickening of 50% or greater than the thickness of the surrounding wall or penetrating at least 0.5 mm into the artery lumen, or the presence of a localized area with CIMT >1.5 mm that entered the lumen and was clearly distinguished from the surrounding area.

Data collection

We collected sociodemographic and clinical variables from the medical records. A trained psychologist conducted a personal interview with each participant, lasting approximately two hours and consisting of a psychological evaluation and a battery of neurocognitive tests.

Ethical standards

The study was approved by the Ethics Committee of the General University Hospital of Alicante. We carried out the project in accordance with good clinical practice standards and international ethical principles applicable to medical research in humans. All participants provided written informed consent.

Data analysis

To describe qualitative variables, we used absolute frequencies and percentages. With regard to quantitative variables, we performed the Kolmogorov-Smirnov test to determine the normality of the distribution and then expressed parametric variables as means $\pm$ SD and nonparametric variables as medians and interquartile ranges. **NCI** prevalence and **ND** impairment were calculated with a 95% confidence interval (CI). We compared baseline characteristics by means of the student's T-test or Mann-Whitney U test.

We dichotomized quantitative variables using clinical criteria (for instance CD4+ cells count) or variable distribution (age, time since HIV diagnosis or time with ART). In the case of variables without established thresholds (IL-6, sCD4 or TNF- α levels), we used the best level that discriminated between the groups. We used the Chi-squared test to assess associations and calculated the crude odds ratio (OR) and 95%CI. Variables showing a significant ($p<0.05$) association in bivariable analysis were fitted in a logistic regression model to calculate the adjusted OR.

We analyzed the relationships among impaired or not impaired neurocognitive domains and the previously dichotomized explanatory variables (CD4+, years since diagnosis, etc.), by the Chi-squared test and OR (95%CI). Variables showing a significant ($p<0.05$) association in bivariable analyses were fitted in a logistic regression model to calculate the adjusted OR. Also, we analyzed neurocognitive domains as continuous variables to study the relationship between their scores and the dichotomized explanatory variables (CD4+, years since diagnosis, etc.) by the student's T-test or Mann-Whitney U test as appropriate.

The level of statistical significance in the hypothesis testing was $p<0.05$. For data analysis, we used the software package IBM-SPSS Statistics 22.0

Results

We included 84 patients with a mean age of 46.02 ± 7.5 years, 77.4% were men. Table 1 shows HIV-related clinical variables and inflammatory biomarkers values. BT was present in 23.6% of participants.

We found a **NCI** prevalence of 29.8% (95% CI 21.0-40.2) and subsequently categorized as ANI (n=16; 19%), MND (n=7; 8.3%), or HAD: (n=2; 2.4%) (Table 2).

Table 3 shows the associations between explanatory variables and **NCI**. Lower education level and smoking were associated with higher prevalence of **NCI** in crude analyses. We observed higher prevalence in patients over 45 years, but the difference was not statistically significant. When we analyzed age as a continuous variable, we observed no differences between **NCI** (46.2±7.2 years) and **non-NCI** patients (45.9±7.6 years). HIV-related variables showing crude associations with **NCI** were previous AIDS diagnosis, ≥10 years since HIV diagnosis, and CD4+ lymphocyte count nadir ≤350 cells/μL. After adjusting for other variables with an ORc **with p≤0.05**, the only variable showing an independent association with **NCI** was time (≥10 years) since HIV diagnosis (OR:6.5, 95%CI: 1.6-26.7). With regard to inflammatory biomarkers, adjusted analyses showed a significant association only with IL-6 levels ≥1.8 pg/mL (OR:6.0, 95%CI: 1.1-31.3). When we compared these same variables quantitatively, we found significant differences only in the CD4+ count nadir (**NCI**: 188.6±114.6 cells/μL versus **non-NCI**: 270.1±158.0 cells/μL; p = 0.02). Participants with **NCI** had a higher frequency of BT, but the difference was not statistically significant. Carotid atherosclerosis was not associated with **NCI**.

We also analyzed the association between explanatory variables **and each ND**, regardless of **NCI** diagnosis. In the multivariable analysis, previous AIDS diagnosis, ≥10 years since HIV diagnosis, and baseline CD4+ lymphocyte count <500 cells/μL were significantly associated with DR impairment. When we analyzed DR as a continuous variable, participants with ≥10 years since HIV diagnosis (−0.2±0.8 versus 0.4±0.6; p<0.001), CD4+ nadir ≤350 cells/μL (−0.1±0.8 versus 0.5±0.6; p<0.01) and carotid plaques (−0.9±1.1 versus −0.2±1.1; p=0.04) scored lower in this domain (Fig 1).

The impairment of the neurocognitive domains executive and motor functions were not associated with any of the dichotomous explanatory variables analysed. However, when we analyzed them as quantitative variables, participants with ≥10 years since HIV diagnosis had worse executive function performance than those with more recent diagnoses (−0.5±0.9 versus −0.1±0.6; p=0.02). Similarly, participants with IL-6 levels ≥1.8pg/mL

scored lower than those with <1.8 pg/mL (-0.5 ± 0.9 versus -0.1 ± 0.7 ; $p=0.04$). Analyzing the motor function domain, we found worse results in patients with ≥ 10 years since HIV diagnosis (-0.4 ± 0.7 versus 0.1 ± 0.5 ; $p=0.004$) and in those with a CD4+ count nadir ≤ 350 cells/ μ L (-0.3 ± 0.7 versus 0.1 ± 0.7 ; $p=0.03$). **We did not find any association between attention/working memory, verbal fluency, speed of information processing and learning domains with explanatory variables analysed in our study.**

Discussion

Neurocognitive impairment prevalence in our participants was high, 29%. According to the Frascati criteria, 19% of the sample had ANI; 8.3%, MND; and 2.4%, HAD. The most affected neurocognitive domain was DR (27.4%).

Previous studies on **NCI** prevalence have reported vastly different findings as a result of the different populations analyzed and the methodologies used. The CHARTER group reported a 45% prevalence of milder forms of HAND (ANI: 33%, MND: 12%) in 1555 HIV-infected adults without medical or psychiatric comorbidities, in the era of highly activity antiretroviral therapy (HAART)⁴. In a Swiss study in 200 people with long-term suppression of HIV viremia⁵, the authors reported an HAND prevalence of 84% in patients with neuropsychological complaints versus 64% in those without; this study did not excluded people with hepatitis C or on methadone treatment. In a German study²⁹, using the Frascati criteria and the International HIV Dementia Scale, the authors reported 43% HAND prevalence in 489 PLWHIV, but not all patients were on ART. Finally, González-Baeza⁶ analyzed the potential differences in HAND prevalence between patients receiving ART and those receiving monotherapy with boosted protease inhibitors, reporting prevalence of 31.6% and 22.9%, respectively. Results are quite similar to those observed in our study, probably because authors used the Frascati criteria, and included non-viremic HIV patients without psychiatric disorders or substance abuse.

In the neurocognitive evaluation used in our study, we made normative adjustments by age, sex, and education level. We found that low education level and active smoking were associated with **NCI** diagnosis, although the statistical significance disappeared in the multivariable analysis. Aging and low education level are frequently reported as factors

associated with low performance on neurocognitive tests; however, other studies do not report these associations⁶.

In our study, after adjusting for other variable, only ≥ 10 years since HIV diagnosis and IL-6 levels or ≥ 1.8 pg/mL were independently associated with **NCI**. It is important to note that people may have been infected for a long time before HIV diagnosis, especially those who acquired the infection more than 10 years ago. Furthermore, up to five years ago, ART was not recommended until CD4+ lymphocyte count fell below 350-500 cells/ μ L. So, the actual association between length of HIV infection and neurocognitive impairment may be stronger than suggested by our results.

Other studies have reported associations between high levels of IL-6 and **NCI** in **PLWHIV**^{19,30}, and also in the general population^{31,32}. Concerning HIV-people, two postulated sources of this immune activation are intracellular HIV DNA in tissues (gut), lymph nodes and monocytes²⁰. It has been suggested that in patients with viral suppression, HAND may be driven by viral components, such as TAT and Nef, whereas in viremic patients, HAND may be fueled by whole virus, in particular envelop proteins³³. Regarding to BT, some studies suggest that chronic or intermittent systemic immune activation secondary to microbial translocation from the gut could play a role in the pathogenesis of HAND^{22,23}. Using ribosomal bacterial DNA in plasma to determine BT, a more sensitive biomarker of BT²⁸, we found no association with **NCI** in the population studied. Our group has demonstrated that BT determined by rDNA raises IL-6 levels, and higher IL-6 levels are associated with subclinical atherosclerosis³⁴. In the present study, subclinical atherosclerosis was not associated with **NCI**; however, participants with subclinical atherosclerosis showed significantly lower capacity for DR. This study and others^{35,36} suggest that CMI thickness could be associated with neurocognitive impairment in PLWHIV.

Finally, we analyzed the association between biological and inflammatory variables **with impaired neurocognitive domains** in our sample. Participants diagnosed with HIV for more than 10 years had lower DR, executive function and motor function scores, and those with higher IL-6 levels had a lower executive function score. In addition, participants with a

previous AIDS diagnosis and baseline CD4+ lymphocyte <500 cells/ μ L had worse DR score. **Memory, learning and recall, are neurocognitive domains analyzed in all patients when NCI is suspected. In contrast with subcortical pattern described initially in patients with HAND, cortical and subcortical functions are frequently affected in HIV people who receive HAART^{37,38}. Postmortem neuropathologic studies performed in PLWHIV with HAND have confirmed cortical and subcortical degeneration in patients receiving HAART³⁹. Aging, subclinical atherosclerosis in carotid arteria, CNS penetration effectiveness (CPE) score of antiretroviral drugs and long-term suppressed HIV-viremia could explain changes observed in the NCI pattern observed in our study and described by other authors during HAART-era.**

Our study has some limitations. Current guidelines suggest performing cerebral magnetic resonance scanning and lumbar puncture in HIV-people with HAND to exclude other possibilities⁴⁰. In our study we performed only neuropsychological tests, however, the absence of other comorbidities or confounding factors, suggests that the neurocognitive impairment observed is likely produced by HIV-associated factors. The cross-sectional design of our study likewise permits only an analysis of association, not causation. Inclusion criteria were very strict, including only well-controlled patients and without comorbidities. Unfortunately, not all PLWHIV meet these criteria, so our results cannot be extrapolated to the whole HIV-infected population. We were unable to evaluate the CPE of ART. Ten people were included in double-blind clinical trials, and some patients were receiving dolutegravir, rilpivirine or tenofovir alafenamide, which are not well-defined in the CPE score. Finally, we observed several trends that did not reach statistical significance, probably because of the sample size.

On the other hand, our study has several methodological strengths over other studies, and this allows us to draw conclusions. Although our strict inclusion criteria limit generalizability, they also constitute the greatest strength of our study: by recruiting only “healthy” HIV-infected people, we eliminated confounding variables that could result in an overestimation of the diagnosis. Moreover, a single, experienced psychologist used the same neuropsychological test battery and followed the same procedures for all participants, ensuring maximum reliability to the protocol.

Results of our study suggest that **NCI** prevalence is high in PLWHIV on stable ART who have undetectable HIV-VL and no diagnosed comorbidities that could be related to neurocognitive impairment. In this population, the two factors associated with NCI are high IL-6 levels (a marker of systemic inflammation) and ≥ 10 years since HIV diagnosis. PLWHIV without comorbidities, but with subclinical atherosclerosis, ranges lower scores in DR.

Our study highlights the importance of chronic HIV infection as a risk factor for developing **NCI** even in well-controlled PLWHIV. Future research **should** investigate whether early diagnosis and immediate ART could reduce the prevalence of NCI in PLWHIV. At the same time, clinicians should seriously consider the possibility of HIV-associated neurocognitive disorder in HIV-positive people complaining of difficulties with delayed recall.

Acknowledgements

The authors would like to thank all study participants without whom this research would not have been possible.

Conflicts of interest.

The authors declare that they have no competing interest.

Funding.

The authors declare that they have not received any financial support to carry out the study.

References

1. Mind Exchange Working Group. Assessment, diagnosis, and treatment of HIV-associated neurocognitive disorder: a consensus report of the mind exchange program. *Clin Infect Dis* 2013;56:1004-17.
2. Sevigny JJ, Albert SM, McDermott MP, et al. An evaluation of neurocognitive status and markers of immune activation as predictors of time to death in advanced HIV infection. *Arch Neurol* 2007;64:97-102.
3. Clifford DB. HIV-associated neurocognitive disorder. *Curr Opin Infect Dis* 2017; 30:117-122.
4. Heaton RK, Clifford DB, Franklin DR Jr, et al. HIV-associated neurocognitive disorders persist in the era of potent antiretroviral therapy: CHARTER Study. *Neurology* 2010; 75:2087-96.
5. Simioni S, Cavassini M, Annoni JM, et al. Cognitive dysfunction in HIV patients despite long-standing suppression of viremia. *AIDS* 2010;24:1243-50.
6. González-Baeza A, Carvajal F, Bayón C, et al. Pattern of neurocognitive function in patients receiving boosted protease inhibitor monotherapy: a detailed neuropsychological study. *J Neurovirol* 2014;20:362-70.
7. Winston A, Arenas-Pinto A, Stöhr W, et al. Neurocognitive function in HIV infected patients on antiretroviral therapy. *PLoS One* 2013;8:e61949.
8. Muñoz-Moreno JA, Prats A, Pérez-Álvarez N, et al. A brief and feasible paper-based method to screen for neurocognitive impairment in HIV-infected patients: the NEU screen. *J Acquir Immune Defic Syndr* 2013;63:585-92.
9. Robertson K, Bayon C, Molina JM, et al. Screening for neurocognitive impairment, depression, and anxiety in HIV-infected patients in Western Europe and Canada. *AIDS Care* 2014;26:1555-61
10. Sacktor N, Skolasky RL, Seaberg E, et al. Prevalence of HIV-associated neurocognitive disorders in the Multicenter AIDS Cohort Study. *Neurology* 2016;86:334-40.
11. Nightingale S, Winston A, Letendre S, et al. Controversies in HIV-associated neurocognitive disorders. *Lancet Neurol* 2014;13:1139-1151

12. Patel PB, Spudich SS. Global Health Neurology: HIV/AIDS. *Semin Neurol* 2018;38:238-246.
13. Gueler A, Moser A, Calmy A, et al. Life expectancy in HIV-positive persons in Switzerland: matched comparison with general population. *AIDS* 2017;31:427-436.
14. Antiretroviral Therapy Cohort Collaboration. Survival of HIV-positive patients starting antiretroviral therapy between 1996 and 2013: a collaborative analysis of cohort studies. *Lancet HIV* 2017;4:e349-e356.
15. Robertson KR, Smurzynski M, Parsons TD, et al. The prevalence and incidence of neurocognitive impairment in the HAART era. *AIDS* 2007;21:1915-21.
16. Hammond ER, Crum RM, Treisman GJ, et al. The cerebrospinal fluid HIV risk score for assessing central nervous system activity in persons with HIV. *Am J Epidemiol* 2014;180:297-307.
17. Jespersen S, Pedersen KK, Anesten B, et al. Soluble CD14 in cerebrospinal fluid is associated with markers of inflammation and axonal damage in untreated HIV-infected patients: a retrospective cross-sectional study. *BMC Infect Dis* 2016; 16:176.
18. Vera JH, Guo Q, Cole JH, et al. Neuroinflammation in treated HIV-positive individuals: A TSPO PET study. *Neurology* 2016;86:1425-32.
19. Lake JE, Vo QT, Jacobson LP, et al. Adiponectin and interleukin-6, but not adipose tissue, are associated with worse neurocognitive function in HIV-infected men. *Antivir Ther* 2015;20:235-44.
20. Imp BM, Rubin LH, Tien PC, et al. Monocyte Activation Is Associated With Worse Cognitive Performance in HIV-Infected Women With Virologic Suppression. *J Infect Dis* 2017;215:114-121.
21. Eggers C, Arendt G, Hahn K, et al. HIV-1-associated neurocognitive disorder: epidemiology, pathogenesis, diagnosis, and treatment. *J Neurol* 2017;264:1715-1727.
22. Ancuta P, Kamat A, Kunstman KJ, et al. Microbial translocation is associated with increased monocyte activation and dementia in AIDS patients. *PLoS One* 2008;3:e2516.
23. Hoenigl M, de Oliveira MF, Pérez-Santiago J, et al. (1→3)- β -D-Glucan Levels Correlate With Neurocognitive Functioning in HIV-Infected Persons on Suppressive Antiretroviral Therapy: A Cohort Study. *Medicine (Baltimore)* 2016; 95:e3162

24. Becker JT, Kingsley L, Mullen J, et al. Vascular risk factors, HIV serostatus, and cognitive dysfunction in gay and bisexual men. *Neurology* 2009;73:1292-9.
25. Wright EJ, Grund B, Robertson K, et al ; INSIGHT SMART Study Group. Cardiovascular risk factors associated with lower baseline cognitive performance in HIV-positive persons *Neurology* 2010;75:864-73
26. Podzamczak Palter D, Muñoz-Moreno JA, Alcolea, et al. Grupo de expertos del Grupo de Estudio de Sida (GeSIDA). Consensus statement on the clinical management of human immunodeficiency virus-associated neurocognitive disorders. *Enferm Infecc Microbiol Clin* 2014;32:37-47.
27. Antinori A, Arendt G, Becker JT, et al. Updated research nosology for HIV-associated neurocognitive disorders. *Neurology*. 2007;69:1789-99.
28. Such J, Francés R, Muñoz C, et al. Detection and identification of bacterial DNA in patients with cirrhosis and culture-negative, nonneutrocytic ascites. *Hepatology* 2002;36:135-41.
29. Marin-Webb V, Jessen H, Kopp U, Jessen AB, Hahn K. Validation of the International HIV Dementia Scale as a Screening Tool for HIV-Associated Neurocognitive Disorders in a German-Speaking HIV Outpatient Clinic. *PLoS One* 2016;11:e0168225.
30. Sattler FR, He J, Letendre S, et al. Abdominal obesity contributes to neurocognitive impairment in HIV-infected patients with increased inflammation and immune activation. *J Acquir Immune Defic Syndr* 2015; 68:281-8.
31. Wright CB, Sacco RL, Rundek T, Delman J, Rabbani L, Elkind M. Interleukin-6 is associated with cognitive function: The Northern Manhattan Study. *J Stroke Cerebrovasc Dis* 2006;15:34-8.
32. Yaffe K, Lindquist K, Penninx BW, et al. Inflammatory markers and cognition in well-functioning African-American and white elders. *Neurology* 2003; 61:76-80.
33. Carroll A, Brew B. HIV-associated neurocognitive disorders: recent advances in pathogenesis, biomarkers, and treatment. *F1000Res* 2017; 6:312.
34. León R, Reus S, López N, et al. Subclinical atherosclerosis in low Framingham risk HIV patients. *Eur J Clin Invest* 2017;47:591-599.

35. Wendell CR, Waldstein SR, Evans MK, Zonderman AB. Subclinical carotid atherosclerosis and neurocognitive function in an urban population. *Atherosclerosis* 2016; 249:125-31.
36. Fabbiani M, Ciccarelli N, Tana M, et al. Cardiovascular risk factors and carotid intima-media thickness are associated with lower cognitive performance in HIV-infected patients. *HIV Med.* 2013;14:136-44.
37. Heaton RK, Franklin DR, Ellis RJ, et al. HIV-associated neurocognitive disorders before and during the era of combination antiretroviral therapy: differences in rates, nature, and predictors. *J Neurovirol* 2011;17:3-16.
38. Chan LG, Kandiah N, Chua A. HIV-associated neurocognitive disorders (HAND) in a South Asian population - contextual application of the 2007 criteria. *BMJ Open.* 2012 Feb 13;2(1):e000662
39. Moore DJ, Masliah E, Rippeth JD, et al. Cortical and subcortical neurodegeneration is associated with HIV neurocognitive impairment. *AIDS.* 2006;20:879-87
40. Underwood J, Winston A. Guidelines for Evaluation and Management of Cognitive Disorders in HIV-Positive Individuals. *Curr HIV/AIDS Rep* 2016;13:235-40.

Table 1. Participant characteristics (N=84)

Demographic variables	
Men, n (%)	65 (77.4)
Age (years), mean±SD	46.0±7.5
Aged ≥ 45 years, n (%)	42 (50.0)
Education, n (%)	
- Primary or no education	30 (35.7)
- Secondary	24 (28.6)
- University	30 (35.7)
Smokers (<10 cigarettes/day), n (%)	13 (15.5)
HIV-related variables	
HIV transmission mechanism, n (%)	
• Ex-IDU	6 (7.1)
• Sexual:	78 (92.9)
- MSM	49 (58.3)
- HTX	29 (34.5)
Previous AIDS diagnosis, n (%)	18 (21.4)
Time since HIV diagnosis (years), mean±SD	12.5±7.1
≥ 10 years since HIV diagnosis, n (%)	47 (56.0)
Nadir CD4+ lymphocyte (cells/μL), mean±SD	245.8±150.5
Nadir CD4+ count ≤ 350 cells/μL, n (%)	64 (76.2)
Current CD4+ lymphocytes (cells/μL), mean±SD	652.5±218.8
Current CD4+ count < 500 cells/μL, n (%)	20 (23.8)
Time receiving ART (years), mean±SD	9.8±6.4
≥ 7 years with ART, n (%)	25 (29.7)
Inflammatory biomarkers and bacterial translocation	
rDNA, n (%)	20 (23.8)
sCD14 (ng/mL), median (IQR)	12.1(10.8-14.3)
sCD14 ≥ 10.8 ng/mL, n (%)	63 (75.0)

IL-6, pg/mL, median (IQR)	3.1 (1.8-24.1)
IL-6 $\geq$ 1.8 pg/mL, n (%)	65 (77.6)
TNF α , pg/mL, median (IQR)	11.4 (6.6-28.1)
TNF- α $\geq$ 6.6 pg/mL, n (%)	63 (75.0)
Subclinical atherosclerosis, n (%) (N=82)	19 (23.1)

HIV: human immunodeficiency virus; IDU: previous intravenous drug users; IQR: interquartile range; MSM: men who have sex with men; HTX: heterosexual relations; rDNA: ribosomal deoxyribonucleic acid; sCD14: soluble CD14 receptor; IL-6: interleukin-6; TNF α : tumor necrosis factor alpha.

Table 2. Prevalence of neurocognitive impairment and neurocognitive domains impaired, according to the Frascati criteria (N=84)

	Prevalence (95% CI)	N
HIV-associated neurocognitive disorder	29.8 (21.0-40.2)	25
- Asymptomatic neurocognitive impairment	19.0 (12.4-28.7)	16
- Mild neurocognitive disorder	8.3 (4.1-16.2)	7
- HIV associated dementia	2.4 (0.2-8.3)	2
Neurocognitive domains impaired (N=84)	Prevalence (95% CI)	
- Delayed recall	27.4 (17.2-37.5)	23
- Executive function	20.2 (11.0-29.4)	17
- Motor function (skills)	16.7 (8.1-25.2)	14
- Speed of information processing	15.5 (7.2-23.8)	13
- Verbal fluency	15.5 (8.1-25.2)	13
- Attention/working memory	13.1 (5.3-20.9)	11
- Learning	5.9 (2.0-13.3)	5

Table 3. Association between explanatory variables and neurocognitive impairment diagnosis (N=84)

Variables	NCI diagnosis		ORc (95% CI)	P _c	ORa (95% CI)	P _a
	%	(n/N)				
Gender						
Male	26.2	(17/65)	0.5 (0.2-1.5)	0.183	-	-
Female	42.1	(8/19)	1			
Age						
≥ 45 years	33.3	(14/42)	1.4 (0.5-3.6)	0.474	-	-
< 45 years	26.2	(11/42)	1			
Education level						
Non/primary	43.3	(13/30)	2.7 (1.0-7.0)	0.043	1.5 (0.5-4.8)	0.573
Secondary/tertiary	22.2	(12/54)	1			
Smokers (<10 cigarettes/day)						
Yes	50.0	(7/18)	3.1 (1.1- 9.2)	0.034	2.0 (0.6-6.8)	0.347
No	24.2	(10/66)	1			
HIV transmission						
Ex-IDU	33.3	(2/6)	1.2 (0.2-7.0)	0.843	-	-
Non IDU	29.5	(23/78)	1			
Previous AIDS diagnosis						
Yes	50.0	(9/18)	3.1 (1.1-9.2)	0.034	1.8 (0.5-6.4)	0.394
No	24.2	(16/66)	1			
Time since HIV diagnosis						
≥ 10 years	41.5	(22/53)	6.6 (1.8-24.6)	0.002	6.5 (1.6-26.7)	0.013
< 10 years	9.7	(3/31)	1			
Nadir CD4+ (cells./μL)						
≤ 350/μL	35.9	(23/64)	5.0 (1.1-23.7)	0.027	2.0 (0.3-11.5)	0.491
> 350/μL	10.0	(2/20)	1			
Current CD4+ (cells./μL)						

< 500/ μ L	25.0	(5/20)	0.7 (0.2-2.3)	0.594	-	-
$\geq$ 500/ μ L	31.3	(20/64)	1			
Time with ART						
$\geq$ 7 years	64.0	(16/46)	1.7 (0.7-4.5)	0.628	-	-
< 7 years	36.0	(9/38)	1			
rDNA						
Positive	40.0	(8/20)	1.8 (0.6-5.3)	0.251	-	-
Negative	26.6	(17/64)	1			
sCD14 (ng/mL)						
$\geq$ 10.8	33.3	(21/63)	2.1 (0.6-7.1)	0.215	-	-
<10.8	19.0	(4/21)	1			
IL-6 (pg/mL)						
$\geq$ 1.8	36.5	(23/65)	5.5 (1.2-25.6)	0.019	6.0 (1.1-31.3)	0.033
<1.8	9.5	(2/21)	1			
TNF α (pg/mL)						
$\geq$ 6.6	28.6	(18/63)	0.8 (0.3-2.3)	0.679	-	-
<6.6	33.3	(7/21)	1			
Subclinical atherosclerosis						
Positive	36.8	(7/19)	1.6 (0.5-4.7)	0.408	-	-
Negative	27.0	(17/63)	1			

NCI: neurocognitive impairment; HIV: human immunodeficiency virus; Ex-IDU: intravenous drug users; rDNA: ribosomal deoxyribonucleic acid; sCD14: soluble CD14 receptor; IL-6: interleukin-6; TNF α : tumor necrosis factor alpha. ORc: crude odds ratio; CI: confidence interval; Pc: crude statistical significance; ORa: adjusted odds ratio; Pa: adjusted statistical significance.

Fig 1. Differences in “delayed recall” Z-score between people diagnosed ≥ 10 versus < 10 years ago (A), CD4+ lymphocytes nadir ≤ 350 versus > 350 CD4+ cells/ μ L (B), and presence versus absence of subclinical atherosclerosis (C).