Cerebrospinal fluid HIV-1 escape according to different thresholds and underlying comorbidities: is it time to assess the definitions?

Tiltle: Cerebrospinal Fluid HIV-1 Escape According to Different Thresholds and Underlying Comorbidities: Is It Time to Assess the Definitions?

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Summary:

No consensus has been reached on how to define cerebrospinal fluid HIV-1 escape (CSF-E). We describe its prevalence in 1095 paired CSF-plasma HIV-RNA measurements from antiretroviral-treated patients according to several definitions and neurological affections. CSF-E prevalence varied substantially (9.0-38.9%) and was higher in patients with cerebrovascular disorders, HIV-associated dementia and white matter abnormalities. Considering the variability in HIV-RNA quantification assays, the biological relevance of viral escape at different thresholds needs to be accurately assessed.

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BACKGROUND

In the combination antiretroviral treatment (cART) era, HIV-related comorbidities and eradication of the virus from reservoirs represent major challenges. In view of this, the study of selective HIV replication observed in the cerebrospinal fluid (CSF) of treated and suppressed patients plays an eminent role. This phenomenon, called CSF viral escape (CSF-E), has in fact been associated erratically with central nervous system (CNS) complications [1-4] and represents a rare opportunity to understand the establishment and persistence of the virus in compartmentalised reservoirs [3]. CSF-E prevalence varies widely from 0,7% to 42,3%, as recently reviewed [3,5]; this difference may be explained by variability in methods and clinical features of patients' samples and by the surprisingly large range of adopted CSF-E definitions [3-5]. An International Consortium has been recently founded: [3] one of its aims is to find consensus on CSF-E through the identification of clinically meaningful CSF HIV-RNA thresholds and level of discordance with plasma HIV-RNA [3]. To date, without a shared CSF-E definition both clinical management and research opportunities are debatable. Aiming at unravelling these issues, we described and compared the prevalence of CSF-E in treated patients according to different escape definitions and several underlying CNS conditions.

METHODS

We conducted a retrospective (1993-2018) multi-centric observational study (Rome, Milan, Brescia, Turin). We included data on HIV-positive adult patients on cART with available paired plasma and CSF HIV-RNA measurements on samples collected maximum 14 days apart. Asymptomatic patients/participants enrolled in research studies (A/R) and those with lymphomas without CNS involvement (LYM) were included for comparisons. The ethics

approval was obtained at each Institutions in the context of other ongoing studies**.** The used CSF-E definitions included:

A. Detectable CSF HIV-RNA with undetectable plasma HIV-RNA or CSF HIV-RNA 1 Log_{10} higher than plasma if the latter was detectable;
B. Detectable CSF HIV-RNA with undetectable plasma HIV-RNA or CSF HIV-RNA 0.5 Log_{10} higher than plasma if the latter was detectable;
C. Detectable CSF HIV-RNA with undetectable plasma HIV-RNA or CSF HIV-RNA higher than plasma if the latter was detectable;
D. Detectable CSF HIV-RNA with undetectable plasma HIV-RNA

RESULTS

We included 1095 paired measurements of CSF and plasma HIV-RNA from 774 patients. Median age was 47 years (40-53); 601 (77.6%) were male; 523 (47.7%) and 556 (50.8%) plasma and CSF HIV-RNA measurement were below the lower limit of quantification. CSF-E cases overall were: 164 (15.0%; A), 200 (18.3%; B), 275 (25.1%; C) and 108 (9.9%; D). The median (IQR) distance between plasma and CSF collection for HIV-RNA measurement was 0 (0-14) days, while CSF-plasma HIV-RNA difference among CSF-E cases were: 1.18 (0.37-1.68; A), 0.91 (0.48-1.59; B), 0.61 (0.30-1.35; C) and 0.55 (0.099-1.55; D) Log10 cp/mL. Similar CSF-E prevalence was observed according to calendar years. CSF-E prevalence according to the four definitions among A/R, LYM and those with HIV-associated neurocognitive disorders (HAND) is depicted in Fig.1, while according to all the different underlying clinical conditions is shown in Supplementary Figure.

CSF-E prevalence in cART-treated patients significantly varied according to escape definitions and CNS affections. Definition C, recently adopted by EACS guidelines [6], was associated with the highest rate of escape in any considered category (Fig.1 and Supplementary Figure). The lowest prevalence was described by definition D in any group. Asymptomatic CSF-E prevalence among A/R and LYM ranged significantly from 9.7 to 20.0% and from 9.0% to 25.1% (definition D-C), as well as for all other included clinical categories. The highest prevalence was observed in patients with cerebrovascular diseases (9.1-36.4%), white matter abnormalities (19-38.1%) and HIV-Associated Dementia (HAD, 11.1-38.9%).

DISCUSSION

In a large sample size we observed that a significant proportion of patients presented with CSF-E: its prevalence varied largely according to the applied definitions and underlying CNS

conditions. We observed that HAD, white matter abnormalities and cerebrovascular disorders were the top three categories identified as having the highest CSF-E prevalence by all the definitions, except for definition D; these categories were also those featured by the highest relative prevalence variability. Interestingly, we observed a CSF-E trend among HAND described by an increasing prevalence along with the worsening of neurocognitive impairment severity according to all definitions, but D. Our sample included few patients with inflammatory conditions and very heterogeneous CNS infections potentially representing secondary CSF-E, having no possibility to differentiate and better explore this issue.

A patient presenting with plasma HIV-RNA below 20 copies/mL and a CSF HIV-RNA of 21 copies/mL would be categorized, by all definitions, as a CSF-E. What may be the clinical and scientific consequences of adopting such a low-threshold definition? A CSF-E diagnosis triggers a remarkable time and resource-consuming clinical workout, a tailored follow-up and primarily it should prompt to reassess and eventually modify the ongoing cART, aiming at a better CNS-targeted combination in terms of penetration and efficacy [7]. According to definition C, we observed a substantially high prevalence of primary asymptomatic CSF-E; current knowledge suggests no relevant worsening over time but a significantly higher immune activation; [8] however, additional biomarkers able to exclude associated CNS damages may be helpful in avoiding unnecessary treatment changes. Still, overestimation may occur and be detrimental. HIV-RNA quantification uncertainty increases at low-level/residual viremia and the estimates precision is limited by biological and analytical variability [9,10]. The lower the viremia, the higher are the coefficients of variation of modern real-time PCR assays, widely ranging (26.7%-83.1%) [9] and presenting total intra-assay variations of up to 0.26 Log10 RNA cp/mL [10], which would account for 57 (20.7%) of our CSF-E cases by definition C. Furthermore, viral blips have been shown to be analytically reproducible in less than a fifth of the samples and this may potentially be applied also to CSF HIV-RNA measurements [9].

Limitations of the present study are mainly represented by the retrospective design, its wide time-frame and the lack of data on antiretroviral treatment duration and CSF-E persistence, so that we may have also included slow CSF suppressors and CSF viral blips.

In conclusion taking into account current laboratory limits, the very high prevalence and the few prospective studies, we believe that CSF escape definitions need to be accurately determined according to meaningful clinical conditions, viral compartmentalization and replication capacity or neurological outcomes: in this framework definition B (CSF HIV-RNA 0.5 Log_{10} higher than plasma HIV-RNA) might better represent underlying virological process.

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Figures:

Figure 1. Prevalence of Cerebrospinal Fluid HIV-1 Escape according to different Definitions and Index clinical conditions among on cART-treated patients

Legend: A/R Asymptomatic patients/participants enrolled in Research studies; LYM Lymphomas without central nervous system involvement; ANI, Asymptomatic Neurocognitive Impairment; MND, Mild Neurocognitive Disorders; HAD, HIV-Associated Dementia; HAND HIV-Associated Neurocognitive disorders

Supplementary Figure. Prevalence of Cerebrospinal Fluid HIV-1 Escape according to different Definitions and Central Nervous System Affections among on cART-treated patients

Legend: A/R Asymptomatic patients/participants enrolled in Research studies; LYM Lymphomas without central nervous system involvement; ANI, Asymptomatic Neurocognitive Impairment; MND, Mild Neurocognitive Disorders; HAD, HIV-Associated Dementia; HAND HIV-Associated Neurocognitive disorders; CNS C, Central Nervous System Cancers (lymphomas and other cancers involving central nervous system); CNS OI, Central Nervous System Opportunistic Infections; Other CNSI, Other Central Nervous System Infections (non-opportunistic bacterial and viral meningitis and encephalitis); CNS Syph, Central Nervous System Syphilis; Syph, Syphilis not involving central nervous system; All CNSI, Central Nervous System opportunistic and non-opportunistic infections, including central nervous system syphilis; CNS ID, Central Nervous System Inflammatory disorders (multiple sclerosis); CVD, Cerebrovascular disorders (stroke and vascular dementia); WMA, White Matter Abnormalities; Other includes neurocognitive deficits not fulfilling Frascati Criteria, Headache, Hepatic encephalopathy, Differential diagnosis in patients without CNS infections, Seizures, Psychiatric symptoms, Neuropathy, Polyneuropathy, Myelitis, Drug-drug Interactions and Paraneoplastic encephalopathy.