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Antiretroviral therapy affects the z-score index of deviant cortical EEG rhythms in naïve HIV individuals

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ABSTRACT

Objective: Here we tested the effect of combined antiretroviral therapy (cART) on deviant electroencephalographic (EEG) source activity in treatment-naïve HIV individuals.
Methods: Resting state eyes-closed EEG data were recorded before and after 5 months of cART in 48 male HIV subjects, who were naïve at the study start. The EEG data were also recorded in 59 age- and sex-matched healthy subjects as a control group. Frequency bands of interest included delta, theta, alpha1, alpha2 and alpha3, based on alpha frequency peak specific to each individual. They also included beta1 (13–20 Hz) and beta2 (20–30 Hz). Low-resolution brain electromagnetic tomography (LORETA) estimated EEG cortical source activity in frontal, central, temporal, parietal, and occipital regions.
Results: Before the therapy, the HIV group showed greater parietal delta source activity and lower spatially diffuse alpha source activity compared to the control group. Thus, the ratio of parietal delta and alpha3 source activity served as an EEG marker. The z-score showed a statistically deviant EEG marker (EEG+) in 50% of the HIV individuals before therapy ($p < 0.05$). After 5 months of cART, delta source activity decreased, and alpha3 source activity increased in the HIV subjects with EEG+ (about 50% of them showed a normalized EEG marker).
Conclusions: This procedure detected a deviant EEG marker before therapy and its post-therapy normalization in naïve HIV single individuals.
Significance: The parietal delta/alpha3 EEG marker may be used to monitor cART effects on brain function in such individuals.
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1. Introduction

Infection with human immunodeficiency virus (HIV) causes neuropathological changes as well as neurological and neuropsychological symptoms in most (i.e. >50%) subjects over time (McArthur, 1987; Chavanet et al., 1988; Gastaut et al., 1989; Chalmers et al., 1990; Fauci and Lane, 1998; Williams et al., 2002, 2012; Selnes, 2005; Anthony and Bell, 2008; Antinori et al., 2007; Roberts et al., 2010). Fortunately, combined antiretroviral therapy (cART) mitigates these effects of HIV on brain function (Clifford, 2008), thanks to the attenuation of viral load (VL) and increased CD4 cell counts (Graham et al., 1992; Hammer et al., 1997; Hunt et al., 2003; Williams et al., 2012). However, the prevalence of HIV patients with neurological and neuropsychological symptoms remains relatively high (Cysique et al., 2009; Sevigny et al., 2004). One of the reasons for this situation is the lack of biomarkers suitable for monitoring the brain function effects of HIV and cART, especially for the personalized clinical management of HIV and therapy monitoring on an individual-, rather than population-level.

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Resting state electroencephalographic (EEG) rhythms are a readily observed and quantified functional feature of the brain, and represent promising candidates for disease biomarkers. EEG rhythms reflect neurophysiological mechanisms of cortical neural synchronization related to the fluctuation of cortical arousal during changes in states of vigilance (Babiloni et al., 2011a). It can be hypothesized that quantitative EEG biomarkers can contribute to a preliminary instrumental assessment of HIV subjects, as well as to understanding the neurophysiological mechanisms underlying HIV's effects on brain function. In this line, preceding studies have shown the topography of resting state eyes-closed EEG rhythms in groups of HIV individuals. Compared to control groups of healthy subjects, HIV groups exhibited decreased posterior alpha (8–13 Hz) power density (Gruzelier et al., 1996; Baldeweg and Gruzelier, 1997; Baldeweg et al., 1997). This effect preceded cognitive and neurological impairment, was associated with changes in psychiatric status, and was normalized by cART (Baldeweg and Gruzelier, 1997; Baldeweg et al., 1997). In addition, deviant theta (4–7 Hz) and alpha power density was associated with mood and immune status (i.e. CD4 counts) in a group of asymptomatic HIV subjects (Gruzelier et al., 1996).

To overcome partially head volume conduction effects blurring the localization of the cortical generators of resting state scalp EEG rhythms, our research group has used a popular freeware called low-resolution brain electromagnetic tomography (LORETA; Pascual-Marqui and Michel, 1994). Results were quite promising at the group level. Compared to healthy subjects, naïve HIV subjects were characterized by greater activity of central and parietal delta sources (<4 Hz), as well as lower alpha (8–12 Hz) sources in widespread cortical regions (Babiloni et al., 2012). These differences in alpha sources were less marked in HIV subjects experiencing cART for at least 12 months (Babiloni et al., 2014). Furthermore, they showed normal delta source activity (Babiloni et al., 2014). This beneficial effect of cART on brain function was confirmed by a longitudinal study in which EEG rhythms were recorded before and after 5 months of cART in naïve HIV subjects (Babiloni et al., 2015a).

More recently, we have tested a simple statistical procedure based on the computation of z-score ($p < 0.05$, one-tailed) to identify treatment-naïve HIV male individuals having deviant activity of cortical (LORETA) sources of resting state eyes-closed delta and alpha rhythms (Babiloni et al., 2015b). The ratio of the activity between parietal delta and high-frequency alpha sources served as an EEG marker of interest. The z-score of this EEG marker allowed the identification of a relatively high percentage (about 40–50%) of treatment-naïve HIV individuals with a statistical difference of EEG activity ($p < 0.05$, one-tailed). These results suggested that this EEG marker might enrich the instrumental assessment of HIV effect on brain function in treatment-naïve HIV male individuals. However, the study could not clarify the extent to which the mentioned z-score procedure is sensitive to the impact of cART on naïve HIV individuals (no follow-up). Therefore, we sought to address this unanswered question in the present study. We hypothesized that the naïve HIV individuals with z-scores indicating that normal EEG source activity before the therapy would show a global stability of this activity after 5 months of cART. In other words, we expected their EEG rhythms to remain relatively unchanged and within the previously determined levels of normalcy even after treatment. In contrast, those individuals with z-scores indicating deviant EEG source activity before treatment were expected to show beneficial effects of cART on this activity (i.e. a normalization of EEG source activity) at the follow-up.

2. Methods

2.1. Subjects

This study included EEG and clinical data of 48 naïve HIV male subjects (mean age 39.4 years ± 1.6 standard error, SE), recruited at University S. Andrea Hospital and Tor Vergata Hospital of Rome (Italy). It also included EEG and clinical data of an age-matched control group of 59 cognitively normal male subjects (Healthy; mean age 39 years ± 2.2 SE), selected from a university archive to obtain the best matching of age and gender between the two groups.

All experiments were performed with the written informed consent of each participant and with the approval by the local ethical committee, in line with the Code of Ethics of the World Medical Association (Declaration of Helsinki) and the standards established by the Author's Institutional Review Board.

2.2. Diagnostic criteria

All participants received laboratory exams for the confirmation of the HIV serostatus. These exams included HIV RNA viral load, Complete Blood Count (CBC), Treponema screening, CD4 lymphocyte count and percent, toxoplasmosis and cytomegalovirus antibody titers, HBV–HCV screening, serum protein, renal and liver function, and albumin. Toxicological analyses on urine samples controlled for the use of cocaine, opiates, amphetamine, and marijuana. No naïve HIV subjects showed CD4 counts compatible with a diagnosis of full-blown AIDS. A Computerized Diagnostic Interview Schedule interview (i.e. Version IV, CDIS-IV) controlled for DSM-IV Axis I and II disorders. The naïve HIV subjects completed questionnaires or brief interviews that assessed their medical history, medication use, parental psychopathology, psychiatric symptoms, demographics, drug and alcohol use, and cognitive status. Furthermore, neuropsychological tests assessed memory, language, executive function/attention, and visuo-construction abilities. Memory was evaluated by the Prose Memory Test delayed recall of a story (Spinnler and Tognoni, 1987). Language was assessed by 1-minute verbal fluency for letters (Novelli et al., 1986) and 1-minute verbal fluency for fruits, animals or car trades (Novelli et al., 1986). Executive function and attention were evaluated by the Trail Making Test part A and B (Reitan, 1958). Finally, Mini-Mental State Examination (MMSE) tested global basic cognitive functions (Folstein et al., 1975). Table 1 reports information about personal and clinical characteristics of the healthy and naïve HIV subjects of the present study. An independent t-test evaluated the presence or absence of statistically significant differences between the two groups (i.e. Healthy and naïve HIV) for age, education, and MMSE score ($p < 0.05$). Results showed a statistically significant difference in the MMSE score ($p = 0.0005$), which was higher in the Healthy group than in the naïve HIV group. Furthermore, there was a higher education in the Healthy group than in the naïve HIV group ($p = 0.01$). In contrast, no statistically significant difference was found for age ($p > 0.9$). Therefore, education was used as a covariate in the subsequent statistical comparisons between the Healthy and the naïve HIV groups.

Exclusion criteria included mental retardation, intelligent quotient (IQ) score lower than 70, seizures, acute illness, and neurosurgery. These criteria also included history of head injury with loss of consciousness (Bauer and Shanley, 2006) and major neurological and psychiatric disorders such as schizophrenia or bipolar disorder (Diagnostic and Statistical Manual of Mental Disorders fourth edition, DSM-IV). Naïve HIV subjects were excluded for medical disorders such as chronic obstructive pulmonary disease, hypertension, hepatic encephalopathy, ocular disorders, Type 1 diabetes, and cirrhosis. They were also excluded in

Table 1
Means (±standard error, SE) of the personal and/or clinical features of healthy and treatment-naïve HIV male individuals of the present study. The third column reports the results of t-tests ($p < 0.05$, one-tailed) comparing these mean values between the two groups (i.e. Healthy vs. treatment-naïve HIV). Legend: MMSE = Mini Mental State Examination; CD4 = CD4 lymphocyte count; VL = HIV-RNA viral load in the blood.

	Healthy	Naïve HIV	p values
N	59	48	
Age (years)	39.0 (±2.2 SE)	39.4 (±1.6 SE)	n.s.
Education (years)	14.8 (±0.5 SE)	13.2 (±0.4 SE)	$p = 0.01$
MMSE score	29.6 (±0.1 SE)	28.4 (±0.3 SE)	$p = 0.0005$
CD4 count (cells/μl)	–	374.2 (±33.3 SE)	
VL (copies/ml)	–	120,166.7 (±32,057.9 SE)	

case of a positive urine toxicology or breathalyzer tests. They were also excluded for a recent (past year) dependence upon alcohol, cocaine or opiates.

All healthy subjects underwent cognitive screening as well as physical and neurological examinations. They were neither affected by chronic systemic illnesses, major neurological or psychiatric diseases nor assumed psychoactive drugs. None of them had a major depression based on a clinical exam.

2.3. EEG recordings

Resting state eyes-closed EEG data were collected by 0.3–70 Hz bandpass, 19 electrodes placed according to 10–20 System (i.e. O1, O2, P3, Pz, P4, C3, Cz, C4, T3, T5, T4, T6, Fp1, Fp2, F7, F3, Fz, F4, and F8) and cephalic reference. To monitor eye movements, the horizontal and vertical electrooculograms (EOGs, 0.3–70 Hz bandpass) were also collected. EEG and EOG data were recorded for 5 min in continuous mode with 256 Hz sampling rate. In the naïve HIV subjects, these data were recorded at baseline (pre-treatment; T0) and after 5 months of cART (T5).

The recordings were carried out in the late morning. To control the state of vigilance, an expert continuously monitored subject's behavior and EEG–EOG signals. The subject was verbally alerted in the case of behavioral or EEG drowsiness. Specifically, the experimenter monitored the appearance of "tonic" theta rhythms, K-complexes and sleep spindles (behavior in the control subjects). Of note, a low percentage of subjects (much less than 10%) required such verbal prompt.

The 5 min duration of the EEG recording allowed the comparison of the present results with several previous EEG studies in neurological patients using recording periods shorter or equal to 5 min (Babiloni et al., 2004, 2006b, 2007, 2012, 2014, 2015a,b; Buchan et al., 1997; Pucci et al., 1999; Rodriguez et al., 2002; Szelies et al., 1999) or about 1 min (Dierks et al., 1993, 2000); longer EEG epochs would have reduced data variability but increased risks for dropping vigilance and arousal.

2.4. Preliminary EEG data analysis

The collected EEG data were off-line fragmented in consecutive periods (epochs) of 2 s. An automatic computerized procedure preliminarily identified the EEG epochs with ocular, muscular, and other types of artifacts (including verbal prompts). EEG epochs with sporadic blinking artifacts (threshold of ± 50 μV) were much less than 10% of the total. These epochs were corrected by an autoregressive method (Moretti et al., 2003). Afterwards, two independent experimenters (i.e. G.N and S.C.), blind to the diagnosis, manually confirmed the EEG epochs accepted for a further analysis. The EEG epochs with an incomplete removal of the blinking or other artifacts were manually rejected. Of note, a control analysis showed no difference ($p > 0.05$) in the percentage of the eyes blinks between the Healthy and the naïve HIV group (exact Fisher test, $p > 0.05$).

2.5. Spectral analysis of the EEG data

A spectrum analysis (FFT, Welch technique, Hanning windowing function with no phase shift) computed power density of EEG rhythms with frequency resolution of 0.5 Hz. Absolute EEG power density values were normalized by the following procedure: 1) mean EEG power density was computed for all frequency bins and all electrodes; and 2) absolute power density values for all frequency bins at each electrode were divided by the mean power density computed at 1). The EEG frequency bands of interest were determined based on the identification of EEG power density peak in the range of alpha rhythms (e.g. 7–13 Hz) in any subject, the so-called individual alpha frequency peak (IAF peak; Klimesch et al., 1998). In this line, the EEG frequency bands of interest were defined as follows: (i) delta, IAF −8 Hz to IAF −6 Hz; (ii) theta, IAF −6 Hz to IAF −4 Hz; (iii) alpha 1, IAF −4 Hz to IAF −2 Hz; (iv) alpha 2, IAF −2 Hz to IAF; and (v) alpha 3, IAF to IAF +2 Hz. Of note, the mean IAF peak was 10.1 Hz (± 0.1 SE) in naïve HIV group and 10.2 (± 0.1 SE) in the Healthy group. No statistical difference in the IAF peak was observed between the two groups (Student's t-test one-tailed, $p > 0.05$). For higher EEG band frequencies, standard fixed frequency bands were selected, as a beta power density peak was not clearly detectable in all subjects. Specifically, they were defined as beta 1 (13–20 Hz) and beta 2 (20–30 Hz).

2.6. Cortical (LORETA) source of EEG rhythms

The LORETA software (http://www.unizh.ch/keyinst/NewLORETA/LORETA01.htm) was used for the estimation of the activity (i.e. neural current density) of cortical sources of the EEG rhythms (Pascual-Marqui and Michel, 1994, Pascual-Marqui et al., 1999, 2002). LORETA is a functional imaging technique belonging to a family of linear inverse solution procedures like minimum norm solution, weighted resolution optimization or weighted minimum norm solution, which model 3D distributions of EEG sources (Valdès et al., 1998; Pascual-Marqui et al., 1999; Phillips et al., 2002; Yao and He, 2001). Of note, LORETA has been successfully used in recent EEG studies on neurological subjects by the present experimental set up of the present study (Dierks et al., 2000; Babiloni et al., 2004, 2006b, 2007, 2012, 2014, 2015a,b). Results of these studies suggest that LORETA provides reliable estimates of distributed cortical source activity of resting state EEG rhythms collected from 19 electrodes placed according to 10–20 montage (Anderer et al., 2000, 2003, 2004; Mulert et al., 2001; Winterer et al., 2001; Babiloni et al., 2004, 2006b, 2007, 2012, 2014, 2015a,b; Veiga et al., 2003). Indeed, these resting state EEG rhythms are generated by largely distributed cortical sources whose EEG activity can be recorded by 10–20 system without spatial aliasing. Overall, the present results suggest that topographically widespread cortical sources of resting state delta and alpha rhythms reflect neurophysiologic abnormalities in naïve HIV subjects, at least at a group level.

LORETA provides 3D solutions to the EEG inverse problem (i.e. LORETA solutions). The LORETA solutions estimate the neural current density into a head model formed by 3 spherical compartments representing the electrical properties of the scalp, skull, and brain. The brain compartment is co-registered to the Talairach probability brain atlas (Talairach and Tournoux, 1988). It includes cortical gray matter and hippocampus, digitated in the Brain Imaging Center of Montreal Neurological Institute. The brain compartment is constituted by 2394 voxels with 7 mm of spatial resolution. Any voxel contains an equivalent current dipole, which is fixed as position and orientation. The LORETA solutions estimate the current intensity of all equivalent current dipoles of the brain compartment to explain scalp EEG power density at scalp electrodes (Pascual-Marqui and Michel, 1994).

Solutions of the EEG inverse problem are under-determined and ill-conditioned, when the number of spatial samples (electrodes) is lower than that of the unknown samples (current density at each voxel). For this reason, the cortical LORETA solutions (i.e. estimates of dipole current density at any voxel) predicting scalp EEG spectral power density were regularized to estimate maximally-smoothed rather than circumscribed EEG source activity (Pascual-Marqui and Michel, 1994, Pascual-Marqui et al., 1999, 2002). Furthermore, these solutions were normalized averaging any estimated dipole current density at each voxel and frequency bin by the mean of the dipole current density computed across all frequencies (0.5–45 Hz) and voxels of the brain volume. This procedure of normalization typically fits EEG power density into a Gaussian distribution and reduces its inter-subject variability (Nuwer, 1988; Leuchter et al., 1993). After this normalization, the LORETA solutions lost the original physical dimension and were represented by arbitrary units. In this scale, the value "1" was equal to the mean of the dipole current density at all frequencies (0.5–45 Hz) and voxels of the brain volume.

In line with the intrinsic low-spatial resolution of LORETA solutions, we developed a MATLAB software to average all LORETA solutions of

Brodmann areas (BAs) belonging to a given cortical macro-region of interest (ROI). Specifically, we considered frontal, central, parietal, occipital, temporal, and limbic ROIs in the LORETA brain compartment. Table 2 reports the list of BAs relative to any ROI used in the present study.

Finally, a main advantage of LORETA source estimation is that the respective contribution of several cortical sources of scalp EEG rhythms can be approximately disentangled. For example, EEG rhythms recorded at scalp parietal electrodes are the summation of EEG activity generated by parietal, occipital, and temporal source activity. LORETA uses a mathematical model of head taking into account the volume conductor effects and may disentangle the contribution of the difference cortical sources of EEG rhythms recorded at scalp parietal electrodes.

2.7. Statistical analyses

Sixth main statistical sessions were performed by commercial tool STATISTICA 10 (StatSoft Inc., www.statsoft.com).

The first statistical session tested the hypothesis that the activity of EEG cortical (LORETA) sources did differ between the Healthy and naïve HIV groups. To this aim, an ANOVA used regional normalized LORETA solutions at T0 as a dependent variable ($p < 0.05$). The ANOVA factors were Group (Healthy, HIV; independent variable), Band (delta, theta, alpha 1, alpha 2, alpha3, beta 1, beta 2), and ROI (central, frontal, parietal, occipital, temporal, limbic). Mauchly's test evaluated the sphericity assumption. The degrees of freedom were corrected by Greenhouse–Geisser procedure when appropriate. The education was used as a covariate. Duncan test was used for post-hoc comparisons ($p < 0.05$ one-tailed). The hypothesis would be confirmed by the following two statistical results: (i) a statistical interaction effect including the factor Group ($p < 0.05$); and (ii) a post-hoc test indicating statistically significant differences between the normalized LORETA solutions with the pattern Healthy ≠ naïve HIV (Duncan test, $p < 0.05$, one-tailed).

The second statistical session tested the hypothesis that the z-score of the ratio between parietal delta and alpha 3 LORETA source activity (i.e. the "EEG marker") at T0 allows the identification of a relatively high percentage (about 40–50%) of naïve HIV individuals with statistical abnormality of EEG activity. To this aim, z-score was calculated for the EEG marker as follows:

$$z = \frac{x-\mu}{\sigma},$$

where "x" is the value of the EEG marker in a given naïve HIV subject; "μ" is the mean of the EEG marker in the reference Healthy group; and "σ" is the standard deviation of the EEG marker in that control group. Overall, z-score indicates the probability that any given naïve HIV subject belongs to the Healthy group in terms of the EEG marker. The statistical threshold was set at $p < 0.05$, one-tailed. This means that a given naïve HIV subject was considered as having a statistically deviant EEG marker ("EEG+") when z-score value of that EEG marker was higher than the threshold of $p < 0.05$ (one-tailed) with reference to the Healthy group. If this is not the case, naïve HIV individuals were denoted as "EEG−" (i.e. statistically normal EEG marker).

The third statistical session tested the hypothesis that EEG+ status is relevant for the assessment of naïve HIV subjects. To this aim, the MMSE score, CD4 count, and VL at T0 were compared between the sub-group of naïve HIV subjects with EEG+ and the sub-group of those with EEG− (Student test, $p < 0.05$ one-tailed). The working hypothesis would be confirmed by t-values indicating lower MMSE score, lower CD4 count, and higher VL in the sub-group of naïve HIV subjects with EEG+ than in the sub-group of those with EEG− ($p < 0.05$ one-tailed).

The fourth statistical session tested the hypothesis that 5 months of cART induces an improvement of global cognitive status and/or biological markers in the two sub-groups of naïve HIV subjects (i.e. EEG+ and EEG−). To this aim, MMSE score, CD4 count, and VL in the two HIV sub-groups were compared between T0 and T5 (Student test, $p < 0.05$ one-tailed). The hypothesis would be confirmed by t-values indicating higher MMSE score, lower CD count, and higher VL at T5 than at T0 ($p < 0.05$ one-tailed; nonparametric test for the MMSE score).

The fifth statistical session tested the hypothesis that 5 months of cART induce a recovery of EEG cortical (LORETA) source activity only in the sub-group of naïve HIV subjects with EEG+. To this aim, two ANOVAs (one for the sub-group of naïve HIV subjects with EEG+ and one for the sub-group of naïve HIV subjects with EEG−) were computed using regional normalized LORETA solutions as a dependent variable ($p < 0.05$). The ANOVA factors were Time (T0, T5), Band (delta, theta, alpha 1, alpha 2, alpha 3, beta 1, and beta 2), and ROI (central, frontal, parietal, occipital, temporal, and limbic). Mauchly's test evaluated the sphericity assumption. Correction of the degrees of freedom was made with the Greenhouse–Geisser procedure when appropriate. Duncan test was used for post-hoc comparisons ($p < 0.05$). The hypothesis would be confirmed by the following three results: (i) a statistical interaction effect including the factor Time ($p < 0.05$) for the sub-group of naïve HIV subjects with EEG+; (ii) a post-hoc test indicating statistically significant differences of the regional normalized LORETA solutions with the pattern T0 ≠ T5 (Duncan test, $p < 0.05$, one-tailed) for the sub-group of naïve HIV subjects with EEG+; and (iii) no statistical interaction effect including the factors Time for the sub-group of naïve HIV subjects with EEG−.

The sixth statistical session tested the hypothesis that the activity of EEG cortical (LORETA) sources is related to biological markers of the HIV. To address this issue, we performed a correlation analysis by Pearson test between normalized LORETA solutions and CD4 count in the two sub-groups of −naïve HIV subjects (i.e. EEG+ and EEG−; $p < 0.05$) at T0. The hypothesis would be confirmed by a statistical correlation ($p < 0.05$) in the sub-group of naïve HIV subjects with EEG+, and no statistical correlation ($p > 0.05$) in the sub-group of naïve HIV subjects with EEG−.

Table 2
Brodmann areas included in the cortical regions of interest (ROIs) of the present study.

LORETA Brodmann areas into the regions of interest (ROIs)	
Frontal	8, 9, 10, 11, 44, 45, 46, 47
Central	1, 2, 3, 4, 6
Parietal	5, 7, 30, 39, 40, 43
Temporal	20, 21, 22, 37, 38, 41, 42
Occipital	17, 18, 19
Limbic	31, 32, 33, 34, 35, 36

3. Results

3.1. Cortical (LORETA) sources of EEG rhythms estimated before the therapy (T0): the Healthy vs. the naïve HIV group

Fig. 1 maps the grand average of the normalized LORETA solutions (i.e., normalized dipole current density at cortical voxels) modeling the distributed EEG cortical sources at delta, theta, alpha 1, alpha 2, alpha 3, beta 1, and beta 2 bands in the Healthy group and in the naïve HIV group before. These data refer to the period before the beginning of the cART (T0). The Healthy group showed alpha 2 and alpha 3 sources with the highest activity in the occipital regions. Delta, theta, and alpha 1 sources pointed to moderate activity when compared to the alpha 2 and alpha 3 source activity. Finally, the beta 1 and beta 2 sources had the lowest activity. When compared to the Healthy group, the naïve HIV group was characterized by higher parietal delta source activity and lower posterior alpha 2 and alpha 3 source activity.

The ANOVA on the normalized LORETA solutions pointed to a statistically significant interaction ($F(30, 3120) = 14.649$, $p < 0.0001$) among the factors Group (HIV and Healthy; independent variable), Band (beta 1, beta 2, alpha 1, alpha 2, alpha 3, theta, and delta), and ROI (frontal, central, temporal, parietal, occipital, and limbic). Fig. 2 shows the

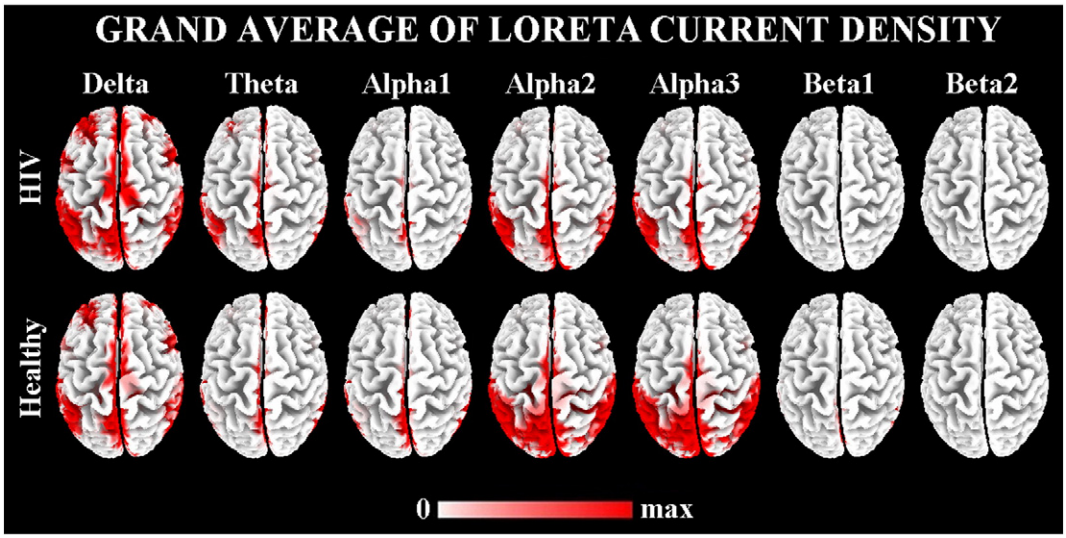

Fig. 1. Grand average of the normalized low-resolution brain electromagnetic tomography (LORETA) solutions (i.e. normalized dipole current density at the cortical voxels) modeling the activity of distributed electroencephalographic (EEG) cortical sources at delta, theta, alpha 1, alpha 2, alpha 3, beta 1, and beta 2 bands in the Healthy and naïve HIV groups before the therapy (T0). The normalization of the LORETA solutions was obtained by computing the ratio between the LORETA current density at each voxel with the mean of the current density values averaged across all frequencies (0.5–45 Hz) and 2394 voxels of the brain source space. The color scale ranges from 0 to the maximum value of the normalized current density estimated at alpha 2 frequency band.

normalized LORETA solutions for this statistical ANOVA interaction. These solutions modeled the activity of the cortical sources of EEG rhythms at the various frequency bands. In the figure, the source activity across the frequency bands has the shape of EEG relative power spectra. Notably, profile and magnitude of the source activity differed in the naïve HIV group compared to the Healthy group. The highest values of the source activity were observed in posterior cortical regions at high-frequency alpha sub-bands (i.e. alpha 2 and 3). Duncan planned post-hoc testing showed that the parietal delta source activity was higher in the naïve HIV group than in the control Healthy group ($p < 0.005$). Furthermore, compared to the control Healthy group, the naïve HIV group showed a lower alpha 1 source activity in the limbic region ($p < 0.05$), as well as lower alpha 2 and alpha 3 source activity in parietal, occipital, temporal, and limbic regions ($p < 0.00005$).

The present results confirm that the parietal cortical sources of resting state eyes-closed delta and high-frequency alpha rhythms are

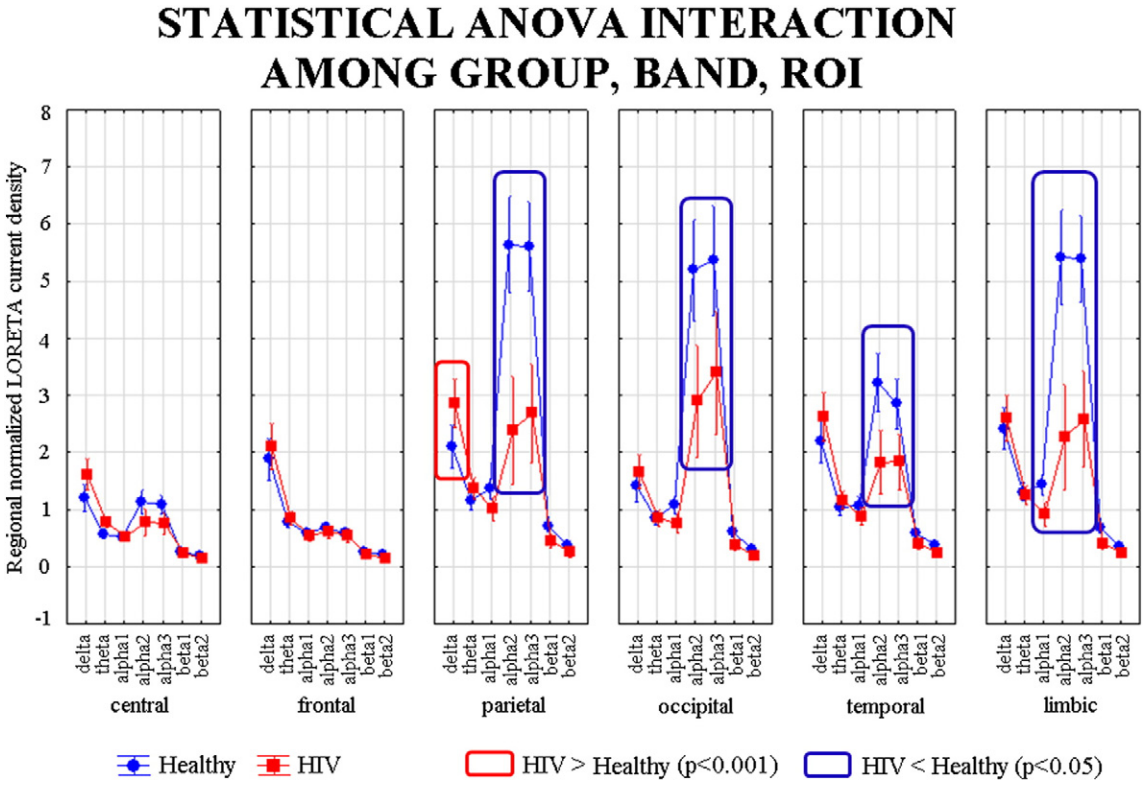

Fig. 2. Mean regional normalized LORETA solutions relative to a statistical ANOVA interaction ($F(30, 3120) = 14.649$, $p < 0.0001$) among the factors Group (HIV and Healthy; independent variable), Band (delta, theta, alpha 1, alpha 2, alpha 3, beta 1, beta 2), and ROI (central, frontal, parietal, occipital, temporal, limbic).

suitable candidates for the computation of a valid EEG marker for a neurophysiological assessment of naïve HIV patients before the beginning of the cART. Fig. 3A represents the individual values of the ratio between parietal delta and alpha-3 source activity (i.e. the EEG marker) in all healthy and naïve HIV subjects before the therapy (i.e. T0). As expected, the EEG marker showed higher values (as a trend towards EEG abnormalities) in the HIV group than the Healthy group.

Fig. 3B plots the z-score of the EEG marker (i.e. the ratio between parietal delta and alpha-3 source activity) in the naïve HIV subjects before the therapy (T0). In the figure, the dashed horizontal separation line indicates the threshold of the statistical difference ($p < 0.05$ one-tailed) that splits all naïve HIV individuals into the following two sub-groups: the sub-group of the naïve HIV subjects with a statistically deviant EEG marker (i.e. EEG+, above the horizontal separation line) and the sub-group of those with a normal EEG marker (i.e. EEG−, below the horizontal separation line). Interestingly, there was a relatively high percentage (i.e. 50%) of the naïve HIV subjects with z-score values indicating a statistically deviant EEG marker (i.e. EEG+).

Fig. 3C draws the scatterplot between the z-score at T0 and T5 for all naïve HIV individuals. In this figure, the diagonal separation line divided the naïve HIV individuals showing a reduction (above the diagonal line those with an improvement of the EEG marker at T5) vs. an increase (down the diagonal line those with a worsening of the EEG marker at T5) in the z-score after 5 months of cART. Note the symmetrical and homogenous distribution of the naïve HIV individuals around the diagonal line until z-score values of about 2.5, suggesting a Gaussian distribution of the cART effects on the EEG marker. Instead, there was a prevalence of the naïve HIV individuals down the diagonal line with the z-score values higher than about 2.5, pointing to a beneficial effect of the cART on the EEG marker in the majority of those naïve HIV individuals.

Table 3 reports the values of a statistical analysis ($p < 0.05$, one-tailed) computed to evaluate the differences between the two sub-groups of the naïve HIV subjects (i.e. EEG+ and EEG−) for each dependent variable of interest, namely the age, education, MMSE score, IAF, CD4 count, VL, Trial making test A, Trial making test B, Trial making test B–A, and Verbal fluency for letter and category. Mann–Whitney U test was used for the MMSE score, and t-test for the remaining variables ($p < 0.05$). Results showed that compared with the naïve HIV sub-group with EEG−, the naïve HIV sub-group with EEG+ exhibited a lower global cognitive status, as revealed by the MMSE score, and abnormal serological indexes such as lower CD4 count and higher VL ($p < 0.05$). The other variables showed no statistical difference ($p > 0.05$).

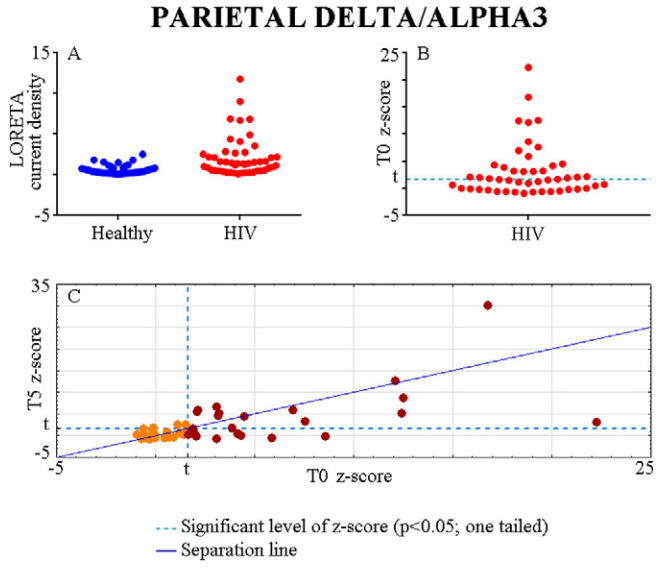

Fig. 3. A) Individual values of the EEG marker in all naïve HIV (before the therapy, T0) and healthy subjects. This EEG marker was defined as the ratio between parietal delta and alpha-3 source activity. The EEG source activity was estimated by the LORETA software. Specifically, this software estimated dipole current density in regions of interest of an average model of the human cerebral cortex (see the Methods). B) z-Score of the EEG marker in all naïve HIV individuals. The z-score values were computed using a control Healthy group as a normative reference. Dashed horizontal separation lines indicate the threshold of a statistical difference ($p < 0.05$, one-tailed) that splits all naïve HIV subjects in two sub-groups at T0: the sub-group of the naïve HIV individuals with a statistically deviant EEG marker (i.e. EEG+, above the line) and the sub-group of those with a normal EEG marker (i.e. EEG−, below the line). C) Scatterplot between the z-score at T0 and T5 in all naïve HIV subjects. In the scatterplot, the naïve HIV individuals with EEG− and EEG+ are represented as yellow and red circles, respectively.

Table 3
Means (±SE) of the demographic, clinical features, and neuropsychological data in a sub-group of naïve HIV individuals with a statistically deviant EEG marker (i.e. EEG+) and another sub-group of naïve HIV individuals with a normal EEG marker (i.e. EEG−). All data refer were collected before (T0) the beginning of a combined antiretroviral therapy (cART). The third column reports the results of a statistical analysis ($p < 0.05$, one-tailed) comparing the reported mean values between the two sub-groups (i.e. EEG− vs. EEG+).

	EEG−	EEG+	p values
N	24	24	
Age (years)	36.8 (±2.2 SE)	39.4 (±1.6 SE)	n.s.
Education (years)	13.6 (±0.4 SE)	12.8 (±0.8 SE)	n.s.
MMSE score	29.2 (±0.3 SE)	27.7 (±0.4 SE)	p = 0.01
IAF (Hz)	10.4 (±0.2 SE)	9.9 (±0.2 SE)	p = 0.02
CD4 count (cells/μl)	443.2 (±44.1 SE)	305.2 (±46.5 SE)	p = 0.01
VL (copies/ml)	65,030 (±313,807.7 SE)	175,303.5 (±61,198.2 SE)	p = 0.04
Trial making test A	37.2 (±2.8 SE)	37.5 (±2.8 SE)	n.s.
Trial making test B	109.4 (±9.4 SE)	93.5 (±9.0 SE)	n.s.
Trial making test B-A	71.6 (±7.8 SE)	56.6 (±7.3 SE)	n.s.
Verbal fluency for letter	34.1 (±2.4 SE)	33.0 (±2.2 SE)	n.s.
Verbal fluency for category	44.2 (±2.1 SE)	41.4 (±1.8 SE)	n.s.

3.2. Effects of cART on clinical markers and cortical (LORETA) sources of EEG rhythms

Table 4 reports the values of t-tests comparing the IAF, CD4 count, and VL between T0 (i.e. before the therapy) and T5 (i.e. after 5 months of therapy) in the naïve HIV sub-group with EEG+ and that with EEG−. For the MMSE score, that table reports the results of the Wilcoxon test ($p < 0.05$). From T0 to T5, the two HIV sub-groups showed an improvement in the serological indexes. Namely, there was a higher CD4 count and a lower VL ($p < 0.05$) at T5 than T0.

Table 4
Means (±SE) of the clinical features of the two sub-groups of treatment-naïve HIV subjects (i.e. EEG+ and EEG−) before (T0) and after 5 months of cART (T5). The third column reports the results of t-tests ($p < 0.05$, one-tailed) comparing these mean values between the pre-treatment and follow up (i.e. T0 vs. T5).

	T0	T5	p values
EEG−			
MMSE score	29.2 (±0.3 SE)	29.0 (±0.4 SE)	n.s.
IAF (Hz)	10.4 (±0.2 SE)	10.1 (±0.2 SE)	n.s.
CD4 count (cells/μl)	443.2 (±44.1 SE)	561.9 (±57.8 SE)	p = 0.0004
VL (copies/ml)	65,030 (±313,807.7 SE)	23.6 (±7.3 SE)	p = 0.00005
EEG+			
MMSE score	27.7 (±0.4 SE)	27.3 (±0.4 SE)	n.s.
IAF (Hz)	9.9 (±0.2 SE)	10.1 (±0.2 SE)	n.s.
CD4 count (cells/μl)	305.2 (±46.5 SE)	438.9 (±47.4 SE)	p = 0.0001
VL (copies/ml)	175,303.5 (±61,198.2 SE)	41.7 (±11.9 SE)	p = 0.004

Fig. 4 illustrates the grand average of the normalized LORETA solutions (i.e., normalized dipole current density at cortical voxels) modeling the EEG source activity at delta, theta, alpha 1, alpha 2, alpha 3, beta 1, and beta 2 bands in the two HIV sub-groups (i.e. EEG+ and EEG−). These LORETA solutions referred to both T0 and T5. The HIV sub-group with EEG+ was characterized by a decrement of the delta source activity from T0 to T5 in widespread cortical regions. In parallel, there was a slight increase of alpha source activity from T0 to T5 in posterior regions. On the contrary, the HIV sub-group with EEG− showed similar EEG source activity at T0 and T5. This effect was true at all frequency bands. Of note, the alpha source activity was quite higher in the HIV sub-group with EEG− than in that with EEG+. This difference was expected based on the criterion (the z-score of the EEG marker) used for the formation of the two sub-groups of HIV subjects. It is irrelevant for the following statistical comparisons due to the "within" design of the planned ANOVAs in the two sub-groups of HIV subjects.

In the sub-group with EEG+ before the therapy ($N = 24$), the ANOVA on the regional normalized LORETA solutions showed a statistically significant interaction ($F(30, 690) = 3.187$, $p < 0.0001$; see Fig. 5 bottom) among the factors Time (T0, T5), Band (delta, theta, alpha 1, alpha 2, alpha 3, beta 1, beta 2), and ROI (frontal, central, temporal, limbic, parietal, occipital). Duncan planned post-hoc testing showed that compared with T0, T5 was associated with increased delta source activity in central, frontal, parietal, occipital, temporal, and limbic regions ($p < 0.005$). It was also related to increased limbic alpha 1 source activity ($p < 0.05$). Furthermore, there was an increase of source activity in parietal, occipital, and limbic alpha 2 and alpha 3 regions ($p < 0.05$). The ANOVA also showed a statistically significant interaction ($F(6, 138) = 4.1882$, $p < 0.0005$) between the factors Time and Band. Duncan planned post-hoc testing showed that compared to T0, T5 was associated with an increased delta activity in widespread sources (i.e. regardless the factor ROI) in the HIV subgroup with EEG+ ($p < 0.005$).

An exploratory correlation analysis (Pearson test, $p < 0.05$) was performed between the some EEG variables of interest (i.e. global delta, posterior alpha-3 source activity, and parietal delta/alpha-3 source activity) and the serological variables (i.e. viral load and CD4 count) before the therapy, when there was no interference of the cART. Results showed a statistically significant correlation only between the global delta source activity and the CD4 count ($r = -0.51$; $p = 0.01$), possibly due to a high inter-subject variability in the alpha source activity in the HIV subjects (Fig. 6).

In the sub-group with EEG− ($N = 24$), the ANOVA on the regional normalized LORETA solutions showed no statistically significant effect of the cART ($p > 0.05$; see Fig. 5 top). To further control about the lack of a relationship between EEG source activity and HIV in these subjects, an exploratory correlation analysis (Pearson test, $p < 0.05$) was performed between some EEG variables of interest (i.e. global delta, posterior high-frequency alpha, and parietal delta/alpha-3 source activity) and the serological variables (i.e. viral load and CD4 count) before the therapy. Results showed no statistically significant correlation between these variables ($p > 0.05$).

In Fig. 7, some diagrams plot the z-score of the EEG marker (i.e. parietal delta/alpha-3 source activity) in the naïve HIV individuals (circles) of the two EEG+ and EEG− sub-groups before (T0) and after 5 months of cART (T5). Overall, most naïve HIV individuals with EEG− showed a quite stable z-score from T0 to T5 (Fig. 7A) while several naïve HIV people with EEG+ unveiled a reduced z-score from T0 to T5 (Fig. 7B). In Fig. 7, other diagrams draw the scatterplots between the z-score at T0 and T5 in the naïve HIV individuals with EEG+ and EEG−. The naïve HIV persons with EEG− showed a symmetrical distribution around the diagonal separation line (Fig. 7C), indicating a Gaussian impact of the cART on the z-score. In contrast, most of the naïve HIV individuals with EEG+ were located down the diagonal separation line (Fig. 7D), pointing to an apparent reduction in the z-score at T5 (i.e. trend towards the normalization of the EEG marker). Other diagrams of the Fig. 7 illustrate the scatterplots between the z-score at T0 and the difference in the z-score between T5 and T0 (i.e. T5 minus T0) as an index of the change due to the therapy. The horizontal separation lines divide the naïve HIV

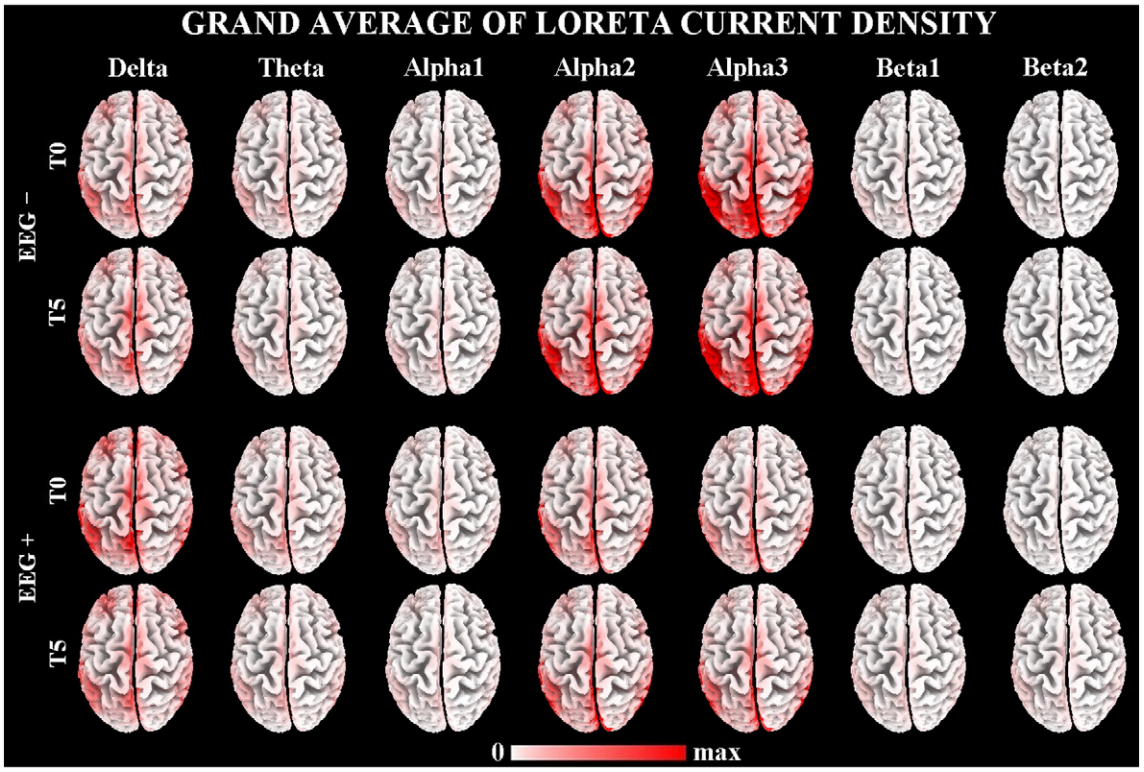

Fig. 4. Grand average of the normalized LORETA solutions modeling the distributed EEG cortical sources at delta, theta, alpha 1, alpha 2, alpha 3, beta 1, and beta 2 bands in the two sub-groups of treatment-naïve HIV individuals (i.e. EEG+ and EEG−) before (T0) and after 5 months of cART (T5). Color scale ranges from 0 to the maximum value of the normalized current density estimated at alpha 3 frequency band.

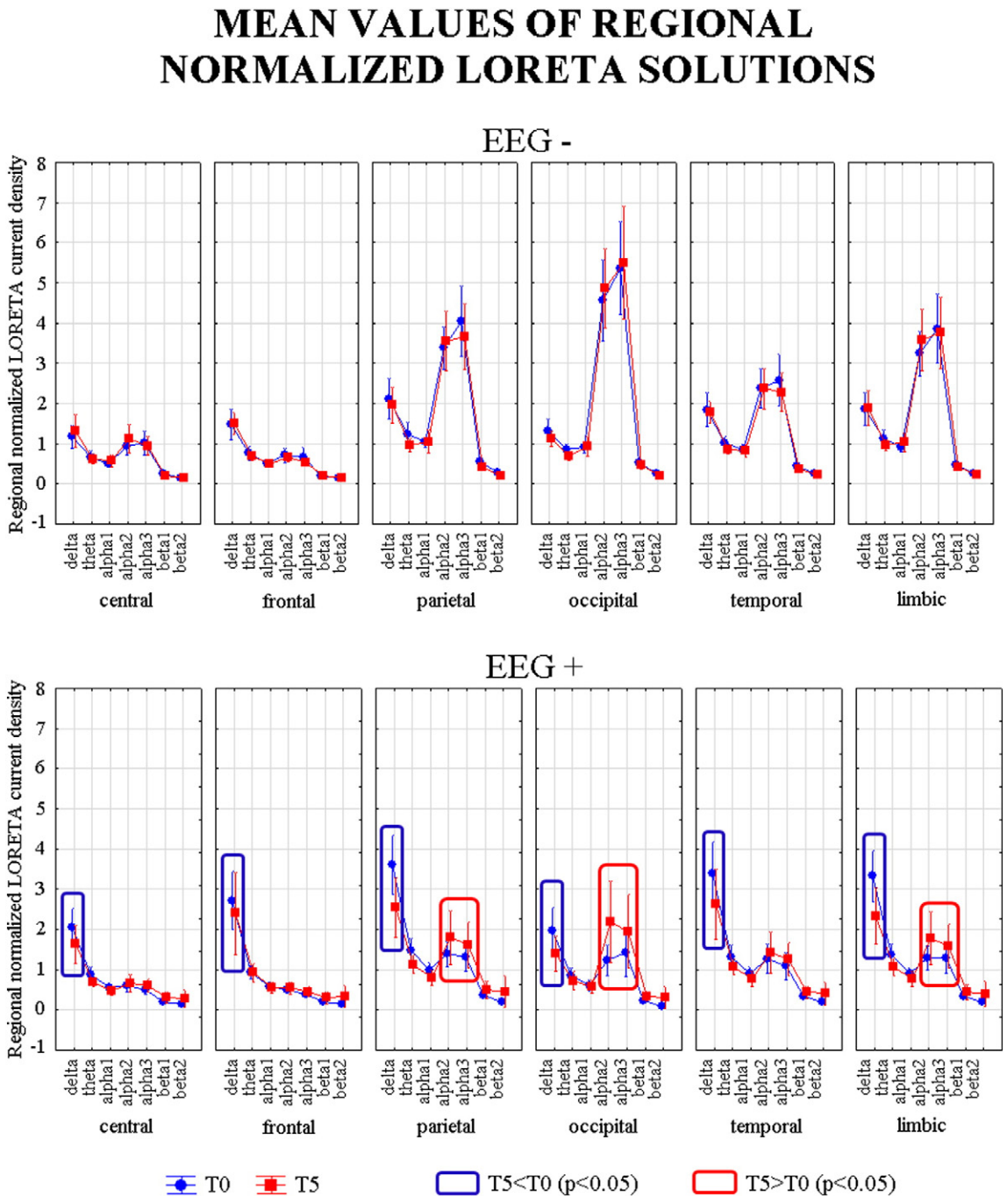

Fig. 5. Top: Mean regional normalized LORETA solutions relative to a statistically non-significant ANOVA interaction ($p > 0.9$) among the factors Time (T0, T5), Band (delta, theta, alpha 1, alpha 2, alpha 3, beta 1, beta 2), and ROI (central, frontal, parietal, occipital, temporal, limbic) for the sub-group of treatment-naïve HIV subjects with a normal EEG marker (i.e. EEG−). Bottom: Mean regional normalized LORETA solutions relative to a statistically significant ANOVA interaction ($F(30, 3120) = 14.649$, $p < 0.0001$) among the factors Time, Band, and ROI for the sub-group of treatment-naïve HIV subjects with a statistically deviant EEG marker (i.e. EEG+).

individuals showing a reduction (down the horizontal separation line) vs. an increase (above the horizontal line) in the z-score after 5 months of cART. Note that the naïve HIV individuals with EEG− (Fig. 7E) were symmetrically and homogenously distributed around the horizontal separation line, indicating a Gaussian distribution of the cART effect on the z-score. In contrast, most of the naïve HIV individuals with EEG+ (Fig. 7F) were located down the horizontal separation line, pointing to a prominent reduction in the z-score at T5 (i.e. trend towards the normalization of the EEG marker).

The statistical analyses confirmed the above readouts. A Fisher-test showed that the T5 minus T0 variance was greater in the naïve HIV individuals with EEG+ than those with EEG+ were ($p < 0.0001$). Furthermore, a t-test unveiled that the T5 minus T0 values were more negative (greater z-score reduction) in the former than the latter HIV group ($p < 0.05$). Moreover, 49% ($N = 11$) of the naïve HIV individuals with EEG+ ($N = 24$) changed to EEG− at T5 (Fig. 3C bottom-right quadrant) while only 8% ($N = 2$) of those with EEG− ($N = 24$) did (Fig. 3C top-left quadrant). These converging findings suggest that 5 months of cART reduced the z-score (normalized the EEG marker) much more in the naïve EEG individuals with EEG+ than those with EEG− did. Indeed, the naïve EEG individuals with EEG+ but not EEG− were supposed to be sensitive to the therapy.

The above results showed that there were two naïve HIV sub-groups of particular interest, namely the EEG+ subjects who recovered to

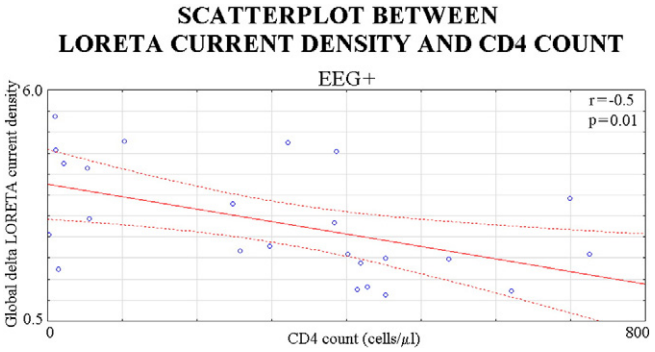

Fig. 6. Scatterplot between the global delta LORETA current density and the CD4 count before the therapy in the naïve HIV subjects with a statistically deviant EEG marker (i.e. EEG+).

EEG− (N = 11 out of 24) and those who remained EEG+ (N = 13 out of 24). How did these HIV sub-groups contribute to the difference T5 minus T0 in the CD4 count and VL in the global HIV group (Table 4)? To address this issue, two control t-tests were performed ($p < 0.05$). The respective dependent variables were the difference T5 minus T0 in the CD4 count and VL. For the VL, there was a greater difference T5 minus T0 (i.e. decrease) in the EEG+ recovered to EEG− than the EEG+ stable ($p = 0.01$). This finding suggests that most of the VL reduction in the whole HIV group was driven by the EEG+ subjects recovered to EEG−. For the CD4 count, no statistical effect was observed ($p = 0.3$), indicating that the recovered CD4 count in the whole HIV group was driven by both HIV sub-groups, namely the subjects with EEG+ recovered to EEG− and those with a stable EEG+. This statistical design was not used for the naïve EEG− subjects as practically all remained EEG− at T5 (i.e. N = 22 out of 24).

3.3. Control analyses

To cross-validate LORETA cortical source estimates, we performed a control analysis at T0 by computing topographic maps of EEG power density for the most representative bands of interest (i.e. delta, alpha 2 and alpha 3) for the Healthy and the naïve HIV groups, separately (Fig. 8). In line with normalization procedure used for LORETA source estimation, EEG power density at each scalp electrode was normalized to EEG power density averaged across all frequencies (0.5–45 Hz) and across all electrodes (N = 19). Fig. 9 plots the statistical t maps of scalp EEG power density for delta, alpha 2 and alpha 3, respectively. These maps showed the statistical differences of the EEG power density between the Healthy and the naïve HIV group. Although these results globally confirmed the differences between the two groups based on the comparison of the corresponding LORETA cortical source estimates, the spatial resolution of EEG power density mapped at scalp level appeared to be lower than that at source level.

4. Discussion

Previous evidence indicated that HIV patients showed a beneficial cART effect on brain function (Clifford, 2008) as reduction in the VL and an increase in the CD4 cell count (Graham et al., 1992; Hammer et al., 1997; Hunt et al., 2003; Williams et al., 2012; Babiloni et al., 2014). Furthermore, we demonstrated that before the cART, about

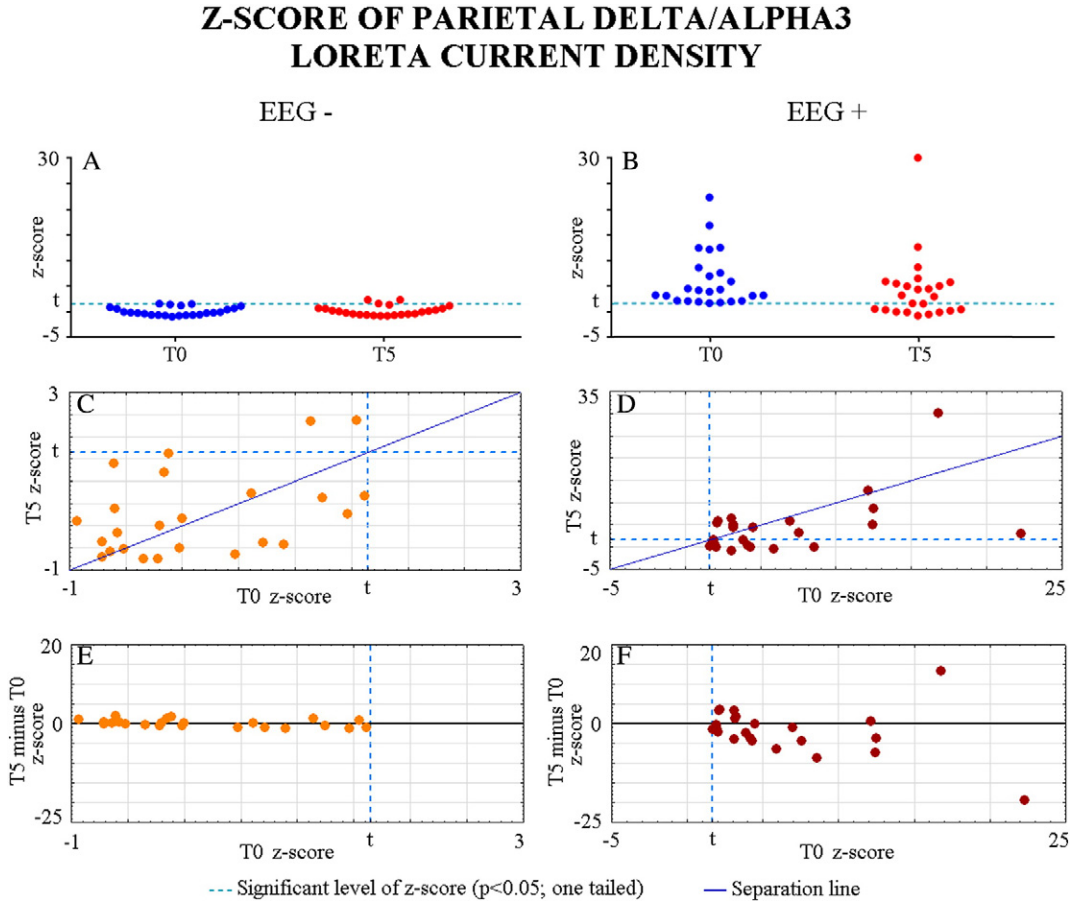

Fig. 7. A) z-Score of the EEG marker in all naïve HIV subjects with EEG− at baseline (T0) and after 5 months of cART (T5). B) z-Score of the EEG marker in all naïve HIV subjects with EEG+ at T0 and T5. C) Scatterplot of the z-score at T0 and T5 in all naïve HIV subjects with EEG−. D) Scatterplot of the z-score at T0 and T5 in all naïve HIV subjects with EEG+. E) Scatterplot of the z-score at T0 and T5 minus T0 (T5–T0) in all naïve HIV subjects with EEG−. F) Scatterplot of the z-score at T0 and T5 minus T0 (T5–T0) in all naïve HIV subjects with EEG+.

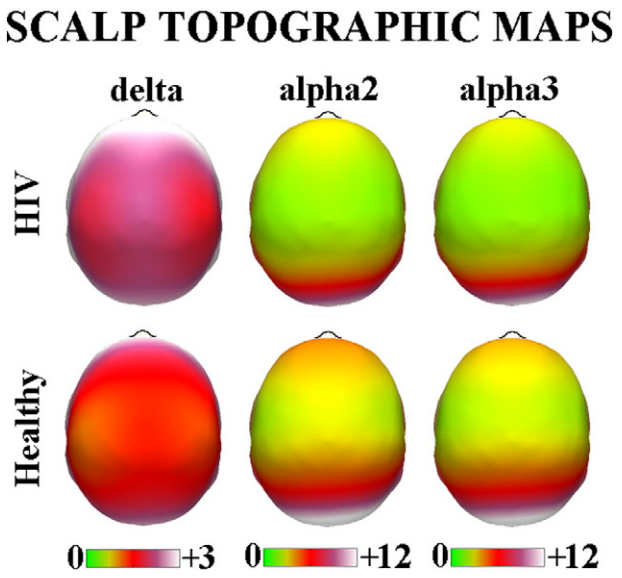

Fig. 8. Spatial distribution of EEG power density for most representative EEG bands (i.e. delta, alpha 2 and alpha 3) at baseline (T0) and for the Healthy and the naïve HIV.

50% of naïve HIV male single individuals were characterized by an abnormal activity of parietal cortical sources of resting state delta and high-frequency alpha rhythms (Babiloni et al., 2015b). Such abnormality was defined as the z-score of the ratio between the parietal delta and high-frequency alpha source activity in any individual naïve HIV male with respect to a reference group of healthy subjects. This ratio, then, was proposed as an EEG marker for the neurophysiological assessment of naïve HIV male subjects before the therapy (Babiloni et al., 2015b).

Results of the present study confirmed those reported in a reference previous research in HIV individuals (Babiloni et al., 2015b). Compared with the Healthy group, the naïve HIV group pointed to a decrease in the low- and high-frequency (i.e. alpha 2 and alpha 3) source activity in several cortical regions (i.e. parietal, occipital, temporal and limbic regions). In contrast, the increase in the delta (i.e. parietal) and the decrease in the low-frequency alpha (i.e. limbic) source activity were regionally circumscribed. Of note 50% (N = 24) of the present naïve HIV subjects pointed to a z-score indicating a deviant EEG marker (i.e. EEG+) before the therapy.

At this early stage of the research, we can just speculate about the clinical neurophysiological meaning of these effects. Resting state eyes-closed alpha rhythms in healthy humans are prominent in widespread posterior cortical regions, especially in visuospatial and somatomotor areas (Pfurtscheller and Lopez da Silva, 1999). These

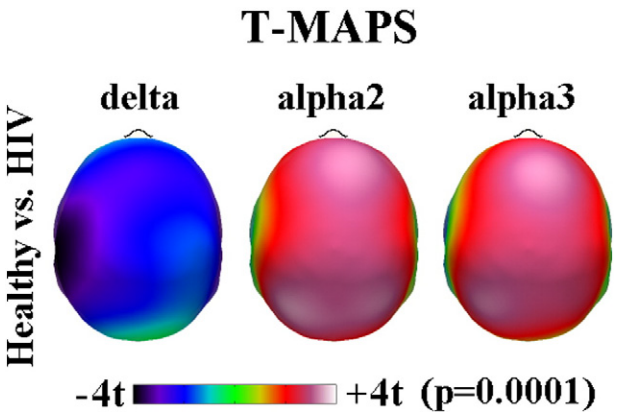

Fig. 9. Topographical maps of statistical values at electrode (sensor) level for frequency bands that presented at baseline most statistical significant differences between Healthy and naïve HIV groups (i.e. delta, alpha 2 and alpha 3).

rhythms are generated because of the cortical neural synchronization in distributed pyramidal neurons, which regulates low brain arousal and vigilance. This effect of synchronization actively inhibits the cerebral cortex, and may depend on the efficiency of the signal transmission and functional connectivity within cerebral neural networks involving the basal forebrain, ascending neurotransmitter systems, thalamus, and cerebral cortex (Pfurtscheller and Lopez da Silva, 1999). The functional meaning of low- and high-frequency alpha rhythms is supposed to be different. Low-frequency alpha rhythms (i.e. alpha 1 and alpha 2) may subserve subject's global arousal and alertness while high-frequency alpha rhythms (i.e. alpha 3) may reflect the event-related oscillation within peculiar neural systems engaged in the elaboration of semantic or sensorimotor information (Klimesch et al., 1998; Klimesch, 1999). Keeping in mind this theoretical premise, the present results hint that the naïve HIV subjects with EEG+ were characterized by a widespread alteration in the neural systems oscillating in broad posterior cortical regions that underpin several cognitive functions such as attention, sensorimotor, and semantic information processing. Instead, a prominent decrease in the lower-frequency alpha rhythm (i.e. alpha 1) was localized in the limbic region, and it might be associated with the regulation of global brain arousal and alertness. Limbic region includes anterior and posterior cingulate cortices and medial prefrontal cortex (BA 31, 32 and 33), which are part of the so-called default mode network (DMN). Previous evidence has shown that DMN operates to maintain a cortical idling and plays a causal role in the enhancement of dominant low-frequency alpha rhythms in quiet wakefulness (Capotosto et al., 2014).

As mentioned above, the present finding showed a noticeable increase in the delta source activity localized in the parietal region. This effect might be associated with vascular lesions in the periventricular white matter, which were found to affect neurotransmission from subcortical to parietal regions in patients with cerebrovascular lesions and cognitive deficits (Leuchter et al., 1994). Future research should evaluate if naïve HIV subjects with EEG+ suffer from this kind of vascular lesions. In previous studies, HIV patients, especially those with cognitive deficits, were characterized by a white matter hyperintensity in MRIs as a reflection of similar cerebrovascular lesions (Thurnher and Donovan Post, 2008).

As a novelty of the present study, here we tested the hypothesis that naïve HIV individuals with z-score indicating a deviant EEG marker (i.e. EEG+), but not HIV patients with normal EEG marker (EEG+), would show a normalization of the EEG source activity after 5 months of cART. This novel hypothesis was grounded on the concept that z-scores of an adequate EEG marker would be sensitive not only to HIV's detrimental effects on brain function, but also to the beneficial effects of cART. Indeed, the present naïve HIV subjects exhibited beneficial effects after 5 months of cART in both delta and high-frequency alpha source activity, although the major effect was observed on the former. Namely, the delta source activity decreased, while the high-frequency alpha source activity increased.

Importantly, as a further novelty, we report that almost half (49%) of these HIV individuals with EEG+ showed a statistical normalization of the proposed EEG marker (z-score) after 5 months of cART. Notably, no statistical difference was found before and after the therapy in the HIV patients with EEG−. Overall, these findings extend previous evidence suggesting that the beneficial cART effects on brain function are significant only in HIV patients with deviant EEG marker before the therapy.

An interesting result of the present study was the cART pronounced effect on delta source activity in the naïve HIV individuals with EEG+ status prior to therapy. A tentative explanation can be based on the physiological meaning of resting state delta rhythms in human subjects. These rhythms are negligible in the physiological condition of relaxed wakefulness, in contrast to dominant amplitude of posterior cortical alpha rhythms (Babiloni et al., 2006b; Klimesch, 1996, 1999; Klimesch et al., 1997, 1998; Pfurtscheller and Lopez da Silva, 1999). Nevertheless, it has been proposed that cortical association areas physiologically

generate delta rhythms as a correlate of integrative information processing across widely spatially distributed neural assemblies (Nuñez, 1995). In this line, delta rhythms have been correlated positively with cortical metabolism in normal subjects, as revealed by fluorodeoxyglucose positron emission tomography (Sadato et al., 1998; Alper et al., 2006). On the other hand, cortical delta rhythms have shown an abnormal high power in patients with cognitive impairment such as Alzheimer's disease, cerebrovascular disease, and Parkinson's disease (Babiloni et al., 2004, 2006b, 2011b). In those with mild cognitive impairment and Alzheimer's disease, these abnormal delta rhythms were associated with white matter vascular lesions (Babiloni et al., 2011c), atrophy of hippocampus and cortical gray matter (Babiloni et al., 2010, 2013), and cortical hypometabolism (Rodriguez et al., 2004). Taking together these studies and the present data, we posit that such deviant activity of widespread delta cortical sources reflects both the HIV impact on brain function and the beneficial effects of cART. The underlying clinical neurophysiological mechanisms have yet to be fully elucidated. Neuronal-glia and inflammatory markers due to HIV might be associated with cerebrovascular, metabolic, and structural changes, as recently shown in vivo in humans by fluorodeoxyglucose positron emission tomography (Sathekge et al., 2014) as well as spectroscopic (Sailasuta et al., 2015), structural (Li et al., 2014), and functional (Chang et al., 2013) magnetic resonance imaging (MRI). In this line of reasoning, these changes might partially derail neural synchronization mechanisms and induce a partial "disconnection mode" of the cerebral cortex (Babiloni et al., 2014).

Keeping in mind the above data and considerations, one might argue that brain atrophy may have an unpredictable impact on the EEG cortical source estimation with a normal average brain model as that implemented in the LORETA freeware. Indeed, a fine EEG cortical source estimation should use a brain source model adapted to the individual cerebral atrophy. Furthermore, it should use a high spatial sampling of the potential distribution over the scalp (i.e. 64–128 electrodes). However, we think that the present methodological approach was adequate for an exploratory study for the following reasons. In the naïve HIV subjects of this study, the magnitude of the brain atrophy is expected to be low, although it can derange the cortical neural synchronization mechanisms generating rsEEG rhythms. In this line, the global cognitive performance, as revealed by the MMSE score, was significantly lower in the HIV group than the matched control group. However, the HIV group exhibited a global cognitive performance within the inferior normal limit, on average. This quasi-normal cognitive status in the HIV group may be associated with an initial phase of the HIV neuroinvasion. At that initial phase, the effects of the HIV neuroinvasion may be not so disruptive to produce marked white matter vascular lesions and grey matter atrophy (Thurnher and Donovan Post, 2008; Peluso et al., 2012; Gottumukkala et al., 2014) to alter significantly the macroscopic volume of the cortical sources generating rsEEG rhythms. The impact of the brain atrophy may be furtherly mitigated from the expected widespread distribution of the rsEEG potential distributions. Indeed, several independent research groups have shown that rsEEG rhythms are generated by largely distributed cortical sources whose EEG activity can be recorded by 10–20 montage system without spatial aliasing. For this reason, several independent research groups used LORETA cortical source estimation from EEG data collected by a relatively low spatial sampling with 19 electrodes of 10–20 system (Anderer et al., 2000, 2003, 2004; Babiloni et al., 2004, 2006a, 2006b, 2006c, 2006d, 2006e; Laufer and Pratt, 2003a, 2003b; Mulert et al., 2001; Veiga et al., 2003; Winterer et al., 2001). According to the theory of the signal sampling, a temporal or spatial sampling should be higher than the spatial and temporal (frequency) information content of the signal to be recorded, to avoid the so-called aliasing (i.e. a distortion in the signal reconstruction). The present methodology may fit this theory. It can be assumed that rsEEG rhythms have a low spatial information content due to a widespread cortical neural synchronization underpinning a low brain arousal and vigilance in a condition of quiet wakefulness. In the present study, we corroborated this assumption with the results of a control analysis performed to cross-validate the LORETA cortical source estimation. In that analysis, the spectral variables were computed from the rsEEG rhythms at the sensor (electrode) level. The results confirmed those obtained at the cortical source level. Overall, topographically widespread cortical sources of delta and alpha rhythms reflected neurophysiologic abnormalities in HIV patients, at least at the group level. It was concluded that the use of a normal brain source model and a traditional 10–20 electrode montage was adequate for this exploratory study, due to the general low spatial information content in the rsEEG rhythms and the expected low brain atrophy in the present naïve HIV subjects. However, future confirmatory studies should use MRI-based realistic head models in naïve HIV subjects for an EEG cortical source estimation performed from rsEEG rhythms recorded using 64–128 scalp electrodes.

4.1. Specificity and clinical significance of the present results

An important remaining issue at present regards the specificity of cART impact on the parietal delta/alpha 3 EEG marker. The current study design prevents a definitive conclusion, as we did not have access to repeated EEG recordings in HIV subjects not receiving cART and control healthy subjects. However, two findings support a preliminary positive answer to this question. Indeed, the naïve HIV individuals showing a normal EEG marker before the therapy (i.e. EEG −) did not exhibit altered EEG source activity after 5 months of cART. Furthermore, such EEG marker was not correlated with the CD4 count. These findings suggest that the z-score procedure can detect not only the cases in which HIV produces deviant EEG source activity but also the cases in which HIV does not produce these effects. These findings support the idea that the present procedure can unveil the pathological effects of HIV on the neural synchronization mechanisms of cerebral cortex generating resting state EEG rhythms.

Another issue of the present results is about the clinical significance of the cART impact on the parietal delta/alpha 3 EEG marker. The present results suggest that the z-score of the EEG marker may enrich the instrumental multifaceted assessment of naïve HIV individuals. This marker should not be considered surrogate index of cognitive impairment in naïve HIV individuals, as the neuropsychological and neurophysiological dimensions of the subject's assessment are complementary rather than overlapping and redundant. These two dimensions explore different levels of brain functioning, namely neurophysiological mechanisms and cognitive performance. In this theoretical framework, the z-score of the EEG marker estimated before and after the cART may be useful to monitor the effect of the therapy on brain function in naïve HIV individuals. This neurophysiological assessment can provide additional useful information for the personalized clinical management of the patient. Let us consider the following example with two naïve HIV subjects after 5 months of cART: Both subjects were EEG + before the therapy and had similar values in MMSE, autonomy in the daily life, and cognitive reserve as indexed by education years and intellectual occupations across the lifespan. One subject's EEG marker status changed from EEG + to EEG − at the follow-up, while the other subject remained EEG + at follow-up. The evaluation of the EEG marker would suggest that, compared to the HIV subject who recovered to EEG − status after therapy, the HIV subject who remained in the EEG + status should receive more neuroprotective therapeutic resources and clinical attention to address his cART-resistant neurophysiological "frailty".

Finally, other issues were the recording of the EEG rhythms at only two time-points (pre-treatment and follow-up), with a relatively short follow-up (5 months), and an undetermined lengthiness of the HIV infection at the date of the pre-treatment EEG recording. It would be important to demonstrate a fine-grain time-course of the EEG marker in association with clinical evolution of the infection and cognitive status, especially in the naïve HIV subjects with pre-treatment EEG +. In these individuals, it can be hypothesized that the HIV infection was

longer when compared to those with pre-treatment EEG −. Unfortunately, we could not test that hypothesis in the present study. We just knew the time of the serological diagnosis of HIV, which does not reliably reflect the time of the HIV infection. In several cases, that diagnosis is incidental. Furthermore, the event of the HIV contagion could not necessarily correspond to the episode told by the subject.

Keeping in mind the above considerations, future research should aim at testing the EEG marker with serial follow-ups across years, especially in the naïve HIV subjects with pre-treatment EEG +. Furthermore, we encourage the quest for the individual clinical cases in whom the event of the HIV contagion can be ascertained for the application of the present methodological approach.

5. Conclusions

In the present study, we computed the z-score ($p < 0.05$, one-tailed) of an EEG marker to monitor the effects of 5 months of cART on brain function in naïve HIV (male) individuals with respect to a reference group of healthy subjects. The EEG marker was defined as the ratio between parietal delta and high-frequency alpha source activity. Before the therapy, the z-score of the EEG marker was statistically deviant (EEG +) in 50% of the naive HIV individuals. In this HIV sub-group with EEG +, delta source activity decreased, while high-frequency alpha source activity increased after 5 months of therapy. Furthermore, >40% of them showed a normalized EEG marker (EEG −) at that follow-up. These results suggest that this statistical procedure was able to identify treatment-naïve HIV single individuals with a deviant EEG source activity before the cART. Furthermore, it was able to monitor the normalization of this EEG source activity after 5 months of therapy in many of them.

Conflict of interest statement

None of the authors has potential conflicts of interest to be disclosed.

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References

Alper, K.R., John, E.R., Brodie, J., Günther, W., Daruwala, R., Prichep, L.S., 2006. Correlation of PET and qEEG in normal subjects. Psychiatry Res. 146, 271–282.
Anderer, P., Saletu, B., Pascual-Marqui, R.D., 2000. Effect of the 5-HT(1A) partial agonist buspirone on regional brain electrical activity in man: a functional neuroimaging study using low-resolution electromagnetic tomography (LORETA). Psychiatry Res. 100, 81–96.
Anderer, P., Saletu, B., Semlitsch, H.V., Pascual-Marqui, R.D., 2003. Non-invasive localization of P300 sources in normal aging and age-associated memory impairment. Neurobiol. Aging 24, 463–479.
Anderer, P., Saletu, B., Saletu-Zyhlarz, G., Gruber, D., Metka, M., Huber, J., Pascual-Marqui, R.D., 2004. Brain regions activated during an auditory discrimination task in insomniac postmenopausal patients before and after hormone replacement therapy: low-resolution brain electromagnetic tomography applied to event-related potentials. Neuropsychobiology 49, 134–153.
Anthony, I.C., Bell, J.E., 2008. The Neuropathology of HIV/AIDS. Int. Rev. Psychiatry 20, 15–24.
Antinori, A., Trotta, M.P., Lorenzini, P., Torti, C., Gianotti, N., Maggiolo, F., et al., 2007. Virological response to salvage therapy in HIV-infected persons carrying the reverse transcriptase K65R mutation. Antivir. Ther. 12, 1175–1183.
Babiloni, C., Binetti, G., Cassetta, E., Cerboneschi, D., Dal Forno, G., Del Percio, C., et al., 2004. Mapping distributed sources of cortical rhythms in mild Alzheimers disease: A multi-centric EEG study. NeuroImage 22, 57–67.
Babiloni, C., Binetti, G., Cassarino, A., Dal Forno, G., Del Percio, C., Ferreri, F., et al., 2006a. Sources of cortical rhythms in adults during physiological aging: a multi-centric EEG study. Hum. Brain Mapp. 27, 162–172.
Babiloni, C., Binetti, G., Cassetta, E., Dal Forno, G., Del Percio, C., Ferreri, F., et al., 2006b. Sources of cortical rhythms change as a function of cognitive impairment in pathological aging: a multi-centric study. Clin. Neurophysiol. 117, 252–268.
Babiloni, C., Benussi, L., Binetti, G., Bosco, P., Busonero, G., Cesaretti, S., et al., 2006c. Genotype (cystatin C) and EEG phenotype in Alzheimer disease and mild cognitive impairment: a multicentric study. NeuroImage 29, 948–964.
Babiloni, C., Benussi, L., Binetti, G., Cassetta, E., Dal Forno, G., Del Percio, C., et al., 2006d. Apolipoprotein E and alpha brain rhythms in mild cognitive impairment: A multicentric EEG study. Ann. Neurol. 59, 323–334.
Babiloni, C., Frisoni, G., Steriade, M., Bresciani, L., Binetti, G., Del Percio, C., et al., 2006e. Frontal White Matter Volume and Delta EEG Sources Negatively Correlate In Awake Subjects With Mild Cognitive Impairment and Alzheimer's Disease. Clin. Neurophysiol. 117, 1113–1129.
Babiloni, C., Cassetta, E., Binetti, G., Tombini, M., Del Percio, C., Ferreri, F., et al., 2007. Resting EEG sources correlate with attentional span in mild cognitive impairment and Alzheimer's disease. Eur. J. Neurosci. 25, 3742–3757.
Babiloni, C., Frisoni, G.B., Vecchio, F., Pievani, M., Geroldi, C., De Carli, C., et al., 2010. Global functional coupling of resting EEG rhythms is related to white-matter lesions along the cholinergic tracts in subjects with amnesic mild cognitive impairment. J. Alzheimers Dis. 19, 859–871.
Babiloni, C., Vecchio, F., Lizio, R., Ferri, R., Rodriguez, G., Marzano, N., et al., 2011a. Resting state cortical rhythms in mild cognitive impairment and Alzheimer's disease: electroencephalographic evidence. J. Alzheimers Dis. 26 (Suppl. 3), 201–214.
Babiloni, C., De Pandis, M.F., Vecchio, F., Buffo, P., Sorpresi, F., Frisoni, G.B., et al., 2011b. Cortical sources of resting state electroencephalographic rhythms in Parkinson's disease related dementia and Alzheimer's disease. Clin. Neurophysiol. 122, 2355–2364.
Babiloni, C., Lizio, R., Carducci, F., Vecchio, F., Redolfi, A., Marino, S., et al., 2011c. Resting state cortical electroencephalographic rhythms and white matter vascular lesions in subjects with Alzheimer's disease: an Italian multicenter study. J. Alzheimers Dis. 26, 331–346.
Babiloni, C., Vecchio, F., Buffo, P., Onorati, P., Muratori, C., Ferracuti, S., et al., 2012. Cortical sources of resting-state EEG rhythms are abnormal in naïve HIV subjects. Clin. Neurophysiol. 123, 2163–2171.
Babiloni, C., Carducci, F., Lizio, R., Vecchio, F., Baglieri, A., Bernardini, S., et al., 2013. Resting state cortical electroencephalographic rhythms are related to gray matter volume in subjects with mild cognitive impairment and Alzheimer's disease. Hum. Brain Mapp. 34, 1427–1446.
Babiloni, C., Buffo, P., Vecchio, F., Onorati, P., Muratori, C., Ferracuti, S., et al., 2014. Cortical sources of resting state EEG rhythms in "experienced" HIV subjects under antiretroviral therapy. Clin. Neurophysiol. 125, 1792–1802.
Babiloni, C., Pennica, A., Vecchio, F., Onorati, P., Muratori, C., Ferracuti, S., et al., 2015a. Antiretroviral therapy effects on sources of cortical rhythms in HIV subjects: Responders vs. Mild Responders. Clin. Neurophysiol. 126, 68–81.
Babiloni, C., Pennica, A., Del Percio, C., Noce, G., Cordone, S., Muratori, C., et al., 2015b. Abnormal cortical sources of resting state electroencephalographic rhythms in single treatment-naïve HIV individuals: a statistical z-score index. Clin. Neurophysiol. 126, 68–81.
Baldeweg, T., Gruzelier, J.H., 1997. Alpha EEG activity and subcortical pathology in HIV infection. Int. J. Psychophysiol. 26, 431–442.
Baldeweg, T., Catalan, J., Pugh, K., Gruzelier, J., Lovett, E., Scurlock, H., et al., 1997. Neurophysiological changes associated with psychiatric symptoms in HIV-infected individuals without AIDS. Biol. Psychiatry 41, 474–487.
Bauer, L.O., Shanley, J.D., 2006. ASPD blunts the effects of HIV and antiretroviral treatment on event-related brain potentials. Neuropsychobiology 53, 17–25.
Buchan, R.J., Nagata, K., Yokoyama, E., Langman, P., Yuya, H., Hirata, Y., et al., 1997. Regional correlations between the EEG and oxygen metabolism in dementia of Alzheimer's type. Electroencephalogr. Clin. Neurophysiol. 103, 409–417.
Capotosto, P., Babiloni, C., Romani, G.L., Corbetta, M., 2014. Resting-state modulation of α rhythms by interference with angular gyrus activity. J. Cogn. Neurosci. 26, 107–119.
Chalmers, A.C., Aprill, B.S., Shepherd, H., 1990. CSF and HIV. Arch. Neurol. 150, 1538–1540.
Chang, L., Holt, J.L., Yakupov, R., Jiang, C.S., Ernst, T., 2013 Aprr. Lower cognitive reserve in the aging human immunodeficiency virus-infected brain. Neurobiol. Aging 34 (4), 1240–1253.
Chavanet, P.U., Giroud, B.S., Lancon, J.P., Borsotti, J.P., Waldner-Combernoux, A.C., Pillon, D., et al., 1988. Altered peripheral nerve conduction in HIV patients. Cancer Detect. Prev. 12, 249–257.
Clifford, D.B., 2008. HIV-associated neurocognitive disease continues in the antiretroviral era. Top. HIV Med. 16, 94–98.
Cysique, L.A., Vaida, F., Letendre, S., Gibson, S., Cherner, M., Woods, S.P., et al., 2009. Dynamics of cognitive change in impaired HIV-positive patients initiating antiretroviral therapy. Neurology 73, 342–348.
Dierks, T., Ihl, R., Frolich, L., Maurer, K., 1993. Dementia of the Alzheimer type: effects on the spontaneous EEG described by dipole sources. Psychiatry Res. 50, 51–162.
Dierks, T., Jelic, V., Pascual-Marqui, R.D., Wahlund, L.O., Julin, P., Linden, D.E.J., et al., 2000. Spatial pattern of cerebral glucose metabolism (PET) correlates with localization of intracerebral EEG-generators in Alzheimer's disease. Clin. Neurophysiol. 111, 1817–1824.
Fauci, S.A.S., Lane, H.C., 1998. HIV disease - AIDS and related disorders. In: Fauci, S.A.S., Braunwald, E., Esselbacher, R.T., et al. (Eds.), Harrison's principle of internal medicine, 14th ed. McGraw Hill Co, USA, pp. 1791–1855.
Folstein, M.F., Folstein, S.E., McHugh, P.R., 1975. "Mini-mental state". A practical method for grading the cognitive state of patients for the clinician. J. Psychiatr. Res. 12, 189–198.
Gastaut, J.L., Gastaut, J.A., Pellissier, J.F., Tapco, J.B., Weill, O., 1989. Peripheral neuropathies in HIV infections - a prospective study of 56 cases. Rev. Neurol. 145, 451–459.
Gottumukkala, R.V., Romero, J.M., Riascos, R.F., Rojas, R., Glikstein, R.S., 2014. Imaging of the brain in patients with human immunodeficiency virus infection. Top. Magn. Reson. Imaging 23, 275–291.
Graham, B.S., Belshe, R.B., Clements, M.L., Dolin, R., Corey, L., Wright, P.F., et al., 1992. Vaccination of vaccinia-naïve adults with human immunodeficiency virus type 1

gp160 recombinant vaccinia virus in a blinded, controlled, randomized clinical trial. The AIDS Vaccine Clinical Trials Network. J. Infect. Dis. 166, 244–252.
Gruzelier, J., Burgess, A., Baldeweg, T., Riccio, M., Hawkins, D., Stygall, J., et al., 1996. Prospective associations between lateralised brain function and immune status in HIV infection: analysis of EEG, cognition and mood over 30 months. Int. J. Psychophysiol. 23, 215–224.
Hammer, S.M., Squires, K.E., Hughes, M.D., Grimes, J.M., Demeter, L.M., Currier, J.S., et al., 1997. A controlled trial of two nucleoside analogues plus indinavir in persons with human immunodeficiency virus infection and CD4 cell counts of 200 per cubic millimeter or less. AIDS Clinical Trials Group 320 Study Team. N. Engl. J. Med. 337, 725–733.
Hunt, P.W., Deeks, S.G., Rodriguez, B., Valdez, H., Shade, S.B., Abrams, D.I., et al., 2003. Continued CD4 cell count increases in HIV-infected adults experiencing 4 years of viral suppression on antiretroviral therapy. AIDS 17, 1907–1915.
Klimesch, W., 1996. Memory processes, brain oscillations and EEG synchronization. Int. J. Psychophysiol. 24, 61–100.
Klimesch, W., 1999. EEG alpha and theta oscillations reflect cognitive and memory performance: a review and analysis. Brain Res. Brain Res. Rev. 29, 169–195.
Klimesch, W., Doppelmayr, M., Pachinger, T., Russegger, H., 1997. Event-related desynchronization in the alpha band and the processing of semantic information. Brain Res. Cogn. Brain Res. 6, 83–94.
Klimesch, W., Doppelmayr, M., Russegger, H., Pachinger, T., Schwaiger, J., 1998. Induced alpha band power changes in the human EEG and attention. Neurosci. Lett. 244, 73–76.
Laufer, I., Pratt, H., 2003a. Evoked potentials to auditory movement sensation in duplex perception. Clin. Neurophysiol. 114, 1316–1331.
Laufer, I., Pratt, H., 2003b. The electrophysiological net response ('F-complex') to spatial fusion of speech elements forming an auditory object. Clin. Neurophysiol. 114, 818–834.
Leuchter, A.F., Cook, I.A., Newton, T.F., Dunkin, J., Walter, D.O., Rosenberg Tompson, S., et al., 1993. Regional differences in brain electrical activity in dementia: use of spectral power and spectral ratio measures. Electroencephalogr. Clin. Neurophysiol. 87, 385–393.
Leuchter, A.F., Dunkin, J.J., Lufkin, R.B., Anzai, Y., Cook, I.A., Newton, T.F., 1994. Effect of white matter disease on functional connections in the aging brain. J. Neurol. Neurosurg. Psychiatry 57, 1347–1354.
Li, Y., Li, H., Gao, Q., Yuan, D., Zhao, J., 2014. Structural gray matter change early in male patients with HIV. Int. J. Clin. Exp. Med. 7, 3362–3369.
McArthur, J.C., 1987. Neurological manifestations of AIDS. Medicine 26, 601–611.
Moretti, D.V., Babiloni, F., Carducci, F., Cincotti, F., Remondini, E., Rossini, P.M., et al., 2003. Computerized processing of EEG-EOG-EMG artifacts for multicentirc studies in EEG oscillations and event-related potentials. Int. J. Psychophysiol. 47, 199–216.
Mulert, C., Gallinat, J., Pascual-Marqui, R., Dorn, H., Frick, K., Schlattmann, P., et al., 2001. Reduced event-related current density in the anterior cingulate cortex in schizophrenia. NeuroImage 13, 589–600.
Novelli, G., Papagno, C., Capitani, E., Laiacona, N., Vallar, G., Cappa, S.F., 1986. Tre test clinici di ricerca e produzione lessicale. Taratura su soggetti normali. Arch. Psicol. Neurol. Psichiatr. 47, 477–506.
Nuñez, P.L., 1995. Experimental connections between EEG data and the global wave theory. In: Nuñez, P.L. (Ed.), Neocortical Dynamics and Human EEG Rhythms. Oxford University Press, London, pp. 534–590.
Nuwer, M.R., 1988. Quantitative EEG: I. tecniques and problems of frequency analysis and topographic mapping. J. Clin. Neurophysiol. 5, 1–43.
Pascual-Marqui, R.D., Michel, C.M., 1994. LORETA (low resolution brain electromagnetic tomography): new authentic 3D functional images of the brain. ISBET Newsl. ISSN 5, 4–8.
Pascual-Marqui, R.D., Lehmann, D., Koenig, T., Kochi, K., Merlo, M.C., Hell, D., et al., 1999. Low resolution brain electromagnetic tomography (LORETA) functional imaging in acute, neuroleptic-naïve, first-episode, productive schizophrenia. Psychiatry Res. 90, 169–179.
Pascual-Marqui, R.D., Esslen, M., Kochi, K., Lehmann, D., 2002. Functional imaging with low resolution brain electromagnetic tomography (LORETA): a review. Methods Find. Exp. Clin. Pharmacol. 24, 91–95.
Peluso, M.J., Ferretti, F., Peterson, J., Lee, E., Fuchs, D., Boschini, A., et al., 2012. Cerebrospinal fluid HIV escape associated with progressive neurologic dysfunction in patients on antiretroviral therapy with well controlled plasma viral load. AIDS 26, 1765–1774.
Pfurtscheller, G., Lopez da Silva, F., 1999. Event-related EEG/MEG synchronization and desynchronization: basic principles. Clin. Neurophysiol. 110, 1842–1857.
Phillips, C., Rugg, M.D., Friston, K.J., 2002. Systemic regularization of linear inverse solutions of the EEG source localization problem. NeuroImage 17, 287–301.
Pucci, E., Belardinelli, N., Cacchio, G., Signorino, M., Angeleri, F., 1999. EEG power spectrum differences in early and late onset forms of Alzheimer's disease. Clin. Neurophysiol. 110, 621–631.
Reitan, R., 1958. Validity of the Trail Making Test as an indication of organic brain damage. Percept. Mot. Skills 8, 271–276.
Roberts, T.K., Buckner, C.M., Berman, J.W., 2010. Leukocyte transmigration across the blood-brain barrier: perspectives on neuroAIDS. Front. Biosci. 15, 478–536.
Rodriguez, G., Vitali, P., De Leo, C., De Carli, F., Girtler, N., Nobili, F., 2002. Quantitative EEG changes in Alzheimer patients during long-term donepezil therapy. Neuropsychobiology 46, 49–56.
Rodriguez, G., Vitali, P., Canfora, M., Calvini, P., Girtler, N., De Leo, C., et al., 2004. Quantitative EEG and perfusional single photon emission computed tomography correlation during long-term donepezil therapy in Alzheimer's disease. Clin. Neurophysiol. 115, 39–49.
Sadato, N., Nakamura, S., Oohashi, T., Nishina, E., Fuwamoto, Y., Waki, A., 1998. Neural networks for generation and suppression of alpha rhythm: a PET study. NeuroReport 9, 893–897.
Sailasuta, N., Ananworanich, J., Lerdlum, S., Sithinamsuwan, P., Fletcher, J.L., Tipsuk, S., Pothisri, M., Jadwattanakul, T., Jirajariyavej, S., Chalermchai, T., Catella, S., Busovaca, E., Desai, A., Paul, R., Valcour, V., 2015. Neuronal-glia markers by Magnetic Resonance Spectroscopy in HIV Before and After Combination Antiretroviral Therapy. J. Acquir. Immune Defic. Syndr. (Aug 6).
Sathekge, M., McFarren, A., Dadachova, E., 2014. Role of nuclear medicine in neuroHIV: PET, SPECT, and beyond. Nucl. Med. Commun. 35 (8), 792–796 (Aug).
Selnes, O.A., 2005. Memory loss in persons with HIV/AIDS: assessment and strategies for coping. AIDS Read. 15, 289–292,294.
Sevigny, J.J., Albert, S.M., McDermott, M.P., McArthur, J.C., Sacktor, N., Conant, K., et al., 2004. Evaluation of HIV RNA and markers of immune activation as predictors of HIV-associated dementia. Neurology 63, 2084–2090.
Spinnler, H., Tognoni, G., 1987. Standardizzazione e taratura italiana di test neuropsicologici. Ital. J. Neurol. Sci. 8, 1–120.
Szelies, B., Mielke, R., Kessler, J., Heiss, W.D., 1999. EEG power changes are related to regional cerebral glucose metabolism in vascular dementia. Clin. Neurophysiol. 110, 615–620.
Talairach, J., Tournoux, P., 1988. Co-Planar Stereotaxic Atlas of the Human Brain. Thieme, Stuttgart.
Thurnher, M.M., Donovan Post, M.J., 2008. Neuroimaging in the brain in HIV-1-infected patients. Neuroimaging Clin. N. Am. 18, 93–117.
Valdès, P., Picton, T.W., Trujillo, N., Bosch, J., Aubert, E., Riera, J., 1998. Constraining EEG-MEG source imaging with statistical neuroanatomy. NeuroImage 4, 635.
Veiga, H., Deslandes, A., Cagy, M., Fiszman, A., Piedade, R.A., Ribeiro, P., 2003. Neurocortical electrical activity tomography in chronic schizophrenics. Arq. Neuropsiquiatr. 61, 712–717.
Williams, M.A., Trout, R., Spector, S.A., 2002. HIV-1 gp120 modulates the immunological function and expression of accessory and co-stimulatory molecules of monocyte-derived dendritic cells. J. Hematother. Stem Cell Res. 11, 829–847.
Williams, D.W., Eugenin, E.A., Calderon, T.M., Berman, J.W., 2012. Monocyte maturation, HIV susceptibility, and transmigration across the blood brain barrier are critical in HIV neuropathogenesis. J. Leukoc. Biol. 91, 401–415.
Winterer, G., Mulert, C., Mientus, S., Gallinat, J., Schlattmann, P., Dorn, H., Herrmann, W.M., 2001. P300 and LORETA: comparison of normal subjects and schizophrenic patients. Brain Topogr. 13, 299–313.
Yao, D., He, B., 2001. A self-coherence enhancement algorithm and its application to enhancing three-dimensional source estimation from EEGs. Ann. Biomed. Eng. 29, 1019–1027.