The spectral slope as a marker of excitation/inhibition ratio and cognitive functioning in multiple sclerosis

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Highlights:

- As expected, the 1/f roll-off was steeper in people with MS (pwMS) treated with benzodiazepines as compared to people with MS not treated with benzodiazepines.
- We performed a dynamic moving time window approach and demonstrated that this difference in 1/f roll-off between pwMS with and without benzodiazepine treatment remains stable across different segments of the recording.
- In the sub-cohort of people with MS not being treated with benzodiazepines, the 1/f roll-off in the prefrontal and parietal cortex significantly correlates with verbal and spatial working memory functioning.
- People with multiple sclerosis with cognitive impairment demonstrate a steeper 1/f slope, suggesting a decreased E/I ratio, and a stronger inhibition/weaker excitation when compared with cognitively preserved pwMS and healthy subjects.

Abstract

Background: Multiple sclerosis (MS) is a neurodegenerative disease characterized by neuronal and synaptic loss, resulting in an imbalance of excitatory and inhibitory synaptic transmission and potentially cognitive impairment. Current methods for measuring the excitation/inhibition (E/I) ratio are mostly invasive, but recent research combining neurocomputational modeling with measurements of local field potentials has indicated that the slope with which the power spectrum of neuronal activity captured by electro- and/or magnetoencephalography rolls off, is a non-invasive biomarker of the excitation/inhibition (E/I) ratio. A steeper roll-off is associated with a stronger inhibition. This novel method can be applied to assess the E/I ratio in people with multiple sclerosis (pwMS), detect the effect of medication such as benzodiazepines, and explore its utility as a biomarker.

Methods: We recruited 44 healthy control subjects and 95 people with multiple sclerosis (pwMS) who underwent resting-state magnetoencephalographic recordings. The 1/f spectral slope of the neural power spectra was calculated for each subject and for each brain region.

Results: As expected, the spectral slope was significantly steeper in pwMS treated with benzodiazepines compared to pwMS not receiving this medication ($p = 0.01$). In the sub-cohort of pwMS not treated with benzodiazepines, we observed a steeper slope in cognitively impaired pwMS compared to cognitively preserved pwMS ($p = 0.01$) and healthy subjects ($p = 0.02$). Furthermore, we observed a significant correlation between 1/f slope and verbal and spatial working memory functioning in the brain regions located in the prefrontal and parietal cortex.

Conclusions: In this study, we highlighted the value of the spectral slope, a novel non-invasive biomarker of the E/I ratio, in MS by quantifying the inhibitory effect of benzodiazepines and by putting it forward as a potential biomarker of cognitive deficits in pwMS.

Keywords: Aperiodic; 1/f exponent; Excitation/inhibition balance; Magnetoencephalography; Resting state; Spectral power

1. Introduction

Multiple sclerosis (MS) is an inflammatory and neurodegenerative disease of the central nervous system (1). Synaptic and neuronal loss are two pathological hallmarks in multiple sclerosis that lead to network dysfunction and disability and contribute to cognitive impairment (2–4). Despite the ongoing debate regarding which types of neurons and synapses are more likely to be affected by multiple sclerosis, the overall loss of synapses and neurons results in an imbalance of excitatory and inhibitory synaptic transmission (3,5). As a strict balance between excitatory and inhibitory synapses is a prerequisite for healthy brain function (6,7), the excitation/inhibition (E/I) ratio could be an important parameter to investigate in multiple sclerosis.

Until recently, there was no method to assess the E/I ratio in vivo. However, recent research combining neurocomputational modeling with measurements of local field potential has shown that the 1/f roll-off of power spectrum density of neuronal activity measured by electro- and/or magnetoencephalography (EEG/MEG) is an easily accessible biomarker of the E/I ratio (8). Further, a recent optogenetic study showed that the 1/f exponent captures fluctuations in the E/I balance (9), with lower exponents being associated with a higher E/I ratio (8). This indicates that a steeper roll-off is associated with a stronger inhibition. This concept has been further corroborated by several recent studies on consciousness (10–13). Moreover, the 1/f spectral slope has been found to decrease with age (14,15), and is modulated by task (12), and neuropsychiatric diseases like schizophrenia (16). Finally, the E/I ratio also has been successfully linked to arousal level and cognitive load (10,12,14,17).

Presently, only a few studies have evaluated this biomarker in neurological diseases. The 1/f spectral slope has been found to be flatter in adolescents with Attention-deficit/hyperactivity disorder (ADHD) on a dual performance attention “stopping task” compared to healthy controls (18). Furthermore, the 1/f slope in resting state EEG was shown to be steeper in schizophrenia patients compared to healthy subjects (19). A steeper 1/f slope was also observed following dopaminergic treatment in patients with Parkinson's disease (PD), highlighting the potential utility of 1/f spectral slope to detect the non-invasive treatment response in patients with PD (20).

Despite the evidence that the spectral slope does pick up the E/I ratio in a novel way, we first performed an additional validation study by comparing the slope between pwMS with and without benzodiazepine treatment. As benzodiazepines are known to enhance the inhibitory effect of GABA, we expect a decrease in E/I ratio and, consequently, a steeper slope. Further,

we hypothesized that in pwMS not treated with benzodiazepines, pwMS with increased E/I ratios would demonstrate decreased cognitive functioning.

2. Methods

2.1. Participants

MEG data were acquired in 139 people with MS (pwMS) during resting-state eyes-closed; 23 pwMS treated with benzodiazepines (MS(BZD+)), 72 not treated (MS(BZD-)) and 44 healthy subjects aged between 18 and 65 years. All pwMS were recruited at the National MS Center Melsbroek and had to have been diagnosed with multiple sclerosis according to McDonald's revised criteria (21) and had an Expanded Disability Status Scale (EDSS) score (22) of less than or equal to 6. A relapse or corticosteroid treatment within the preceding six weeks, as well as a pacemaker, dental wires, major psychiatric disorders, or epilepsy were all criteria for exclusion from the study.

2.2. MEG data acquisition

The MEG data was collected at the CUB Hôpital Erasme (Brussels, Belgium) on an Elekta Neuromag Vectorview scanner (Elekta Oy, Helsinki, Finland) for the first 30 pwMS and 14 healthy subjects and the rest of subjects were scanned using an upgraded scanner, Elekta Neuromag Triux scanner (MEGIN, Croton Healthcare, Helsinki, Finland). Both MEG scanners used sensor layout as 102 triple sensors, each consisting of one magnetometer and two orthogonal planar gradiometers and were placed in a lightweight magnetically shielded room (MaxshieldTM, Elekta Oy, Helsinki, Finland).

The MEG data were acquired in the eyes-closed resting-state condition for 8 minutes. All participants were asked to stay still and close their eyes. MEG signals were recorded with a 0.1–330 Hz passband filter at 1 kHz sampling rate. Throughout the whole data acquisition process, the head positions of the subjects were tracked continuously using four head-tracking coils. The location of these coils and at least 400 head-surface points (on the nose, face, and scalp) with respect to anatomical fiducials were determined with an electromagnetic tracker (Fastrak, Polhemus, Colchester, Vermont, USA). An electrooculogram (EOG) and electrocardiogram (ECG) were simultaneously recorded with MEG signal acquisition.

2.3. MEG processing and parcellation

First, we used the temporal extension of the signal space separation algorithm (MaxfilterTM, Elekta Oy, Helsinki, Finland, version 2.2 with default parameters) to remove the external interferences and correct head movements (23). Then, all data were down sampled to 250 Hz.

Using OSL's RHINO (https://github. com/OHBA-analysis) algorithm, data were automatically coregistered with the subject's T1 image: the head shape points were coregistered to the scalp extracted using FSL's BETSURF and FLIRT (24,25), and then transformed into a common MNI152-space (26). Data were filtered between 0.1 and 70 Hz and then a 5th order butterworth filter (between 49.5 and 50.5 Hz) was applied to remove the power line noise.

A semi-automated independent component analysis (ICA) algorithm was employed to visually identify and remove ocular and cardiac artefacts based on the correlation of the components' time series with EOG and ECG signals respectively. Next, we used a linearly constrained minimum variance (LCMV) beamformer to project the MEG data to a source space (27–29). The source-reconstructed data were then parceled using a parcellation atlas including 42 parcels (30). The first principal component (PC) of the different time series within each parcel was used as the time series for that parcel. As previously discussed, the parcellation atlas covered the entire cortex and did not include subcortical areas (30,31).

2.4. Power spectral analysis and estimation of the aperiodic components

For each parcel, the power spectral density (PSD) was calculated using the default Welch function in SciPy with a window length of 500 samples (2 seconds). Further, we used the "fitting oscillations and one over f" (FOOOF) algorithm (32) to estimate the 1/f exponent. This algorithm aims to extract the oscillatory component by applying an iterative Gaussian fit to all periodic components. By modeling the oscillatory part and removing it from the fundamental power spectrum, an ideal 1/f roll-off can be extracted. We defined the fitting frequency range between 20 Hz to 45 Hz to avoid the oscillatory peaks and to be in line with the methodology as presented by Gao et al (8), suggesting the stronger relationship between E/I ratio and spectral slope within the intermediate frequency range.

2.4.1. Static analysis

For the static analysis, we used the first 2-minutes of recorded MEG data in order to reduce the possible effect that pwMS with BZD were more likely to fall asleep during the recording. For each subject, the power spectrum density of 42 brain parcels was calculated separately, and the 1/f slopes were extracted using the FOOOF algorithm.

2.4.2. Dynamic analysis

To perform a dynamic analysis, we used the entire 8 minutes of recorded MEG data, segmenting it into 16 sections of 30 seconds each. We then performed a static analysis within each 30-second data segment to evaluate the 1/f spectral slopes.

2.5. Estimation of the periodic power in the beta frequency range

We defined the beta frequency range between 18 and 30 Hz to avoid the strong peak at 16.6 Hz which was likely caused by mechanical vibrations due to the construction works at the time of data acquisition at the hospital. We averaged the power within the defined frequency band [18-30 Hz] to estimate the beta power.

2.6. Neuropsychological assessment

Neuropsychological tests have been done on the day of the MEG recording for all subjects. The neuropsychological tests included the Symbol Digit Modalities Test (SDMT, (33)) to capture information processing speed, the Dutch version of the California Verbal Learning Test (CVLT-II, (34)), Dutch version: VGLT to assess verbal memory, the Controlled Oral Word Association Test (COWAT, (35)) to assess verbal fluency and the Brief Visuospatial Memory Test (Revised; BVMT-R, (36)) to assess spatial memory. Fatigue is assessed by the Fatigue Scale for Motor and Cognitive Function (FSMC, (37)) and depression by Beck's depression inventory (BDI, (38)).

2.7. Cognitive impairment in multiple sclerosis

The criterion for cognitive impairment was defined as performance below 2 standard deviations (SD) compared to all three normative SDMT, BVMT, and VGLT scores (39). We used a linear regression model similar to the model suggested by Costers et al (40), on the group of healthy subjects (matched in age and gender) to correct the raw cognitive scores.

2.8. Statistics

All statistical analyses used non-parametric measures, which do not rely on assumptions about the distribution of the data. We present the U-statistic and effect size for all pairwise comparisons performed with the Mann-Whitney U test. When calculating tests for multiple cortical parcels, the Benjamini-Yekutieli false discovery rate (FDR) (41) correction for multiple comparisons was applied to the p-values. For all p-values, the cutoff of 0.05 was used. Correlation analyses were controlled for the Expanded Disability Status Scale (EDSS) and disease duration variables using partial Spearman correlation. The effect size of all analyses will be reported.

2.9. Ethics

The research was approved by the University Hospital Brussels's local ethics committees of the University Hospital Brussels (Commissie Medische Ethiek UZ Brussel, B.U.N. 143201423263, 2015/11) and the National MS Center Melsbroek (2015-02-12). All participants also provided written informed consent.

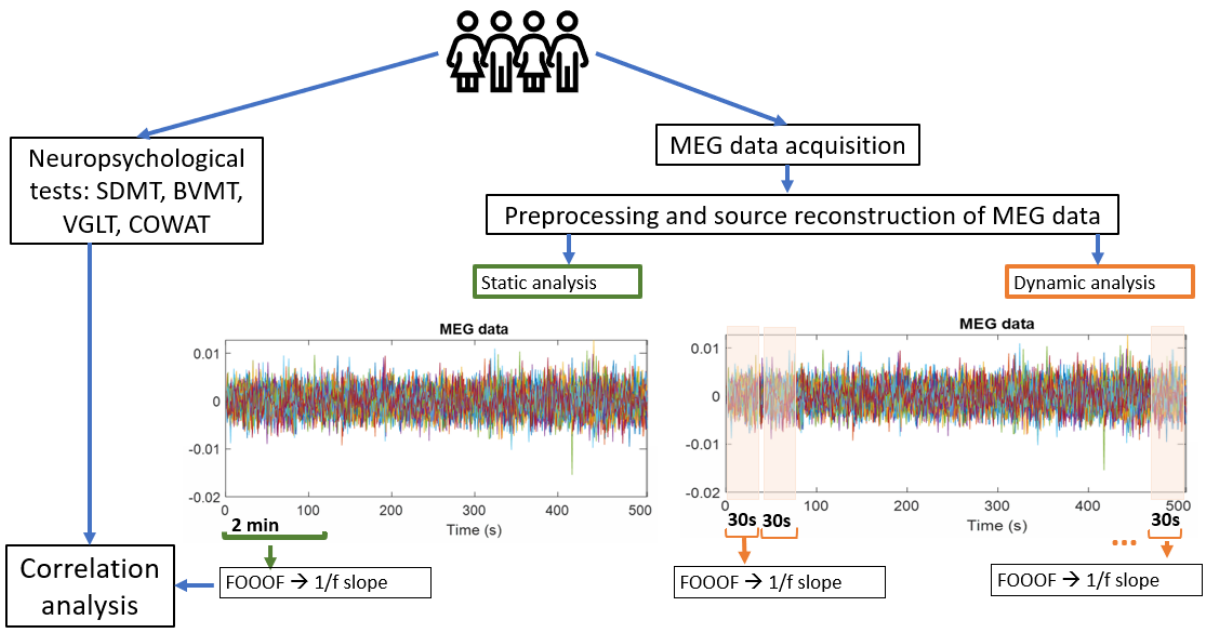

Figure 1. Pipeline of study. We obtained MEG resting-state eyes closed data and neuropsychological test results at the same day. The MEG data was processed to enable the extraction of the 1/f slope. This was then compared between people with and without benzodiazepines and later also correlated with cognitive test results.

3. Results

3.1. Population description

We provide a detailed description of the included cohorts of people with MS (pwMS) and healthy controls in Table 1.

3.2. Static analysis of comparing MS populations: MS(BZD+) vs MS(BZD-)

3.2.1. Whole brain

We start by presenting a whole-brain analysis by averaging the 1/f slope between 20 and 45 Hz across the 42 parcels for each subject. Next, we compared the averaged 1/f slopes between two groups of pwMS (MS(BZD+) vs MS(BZD-)). As expected, we observed a significant difference in 1/f slope in which pwMS treated with benzodiazepines having a steeper roll-off (p-value = 0.014, Mann-Whitney-U = 547, η^2= 0.063, Cohen's d = 0.51).

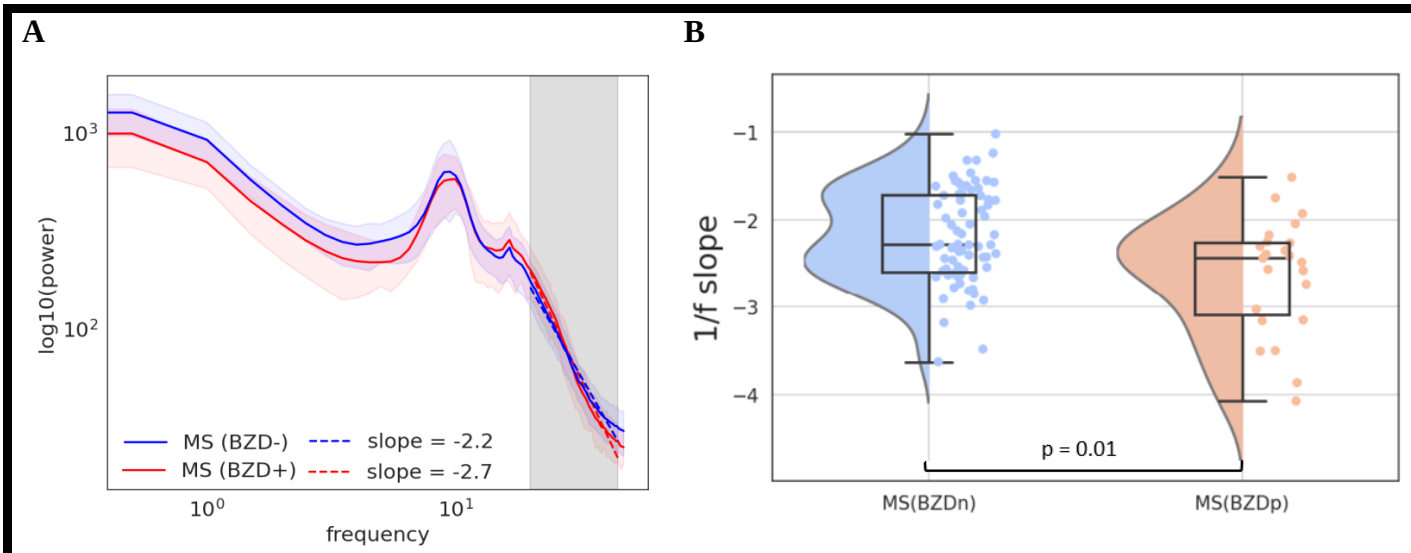

Figure 2. A) The spectra of both cohorts on a log-log scale and the corresponding 1/f slopes. The solid lines present the median and the shaded areas the median absolute deviation. The grey box indicates the area in which the roll-off was calculated [20-45 Hz]. **B)** Averaged 1/f slope across the whole brain for each subject. p-value = 0.014, Mann-Whitney-U = 1109, η2 = 0.063, Cohen's d = 0.51.

	HC	**MS sub-groups**		**MS(BZD-) subgroups**		**Group differences (p-values)**			
	HC	MS(BZD+)	MS(BZD-)	MS(BZD-)_CI	MS(BZD-)_CP	MS(BZD+) **vs** MS(BZD-)	HC **vs** MS(BZD-)_CI	HC **vs** MS(BZD-)_CP	MS(BZD-)_CI **vs** MS(BZD-)_CP
N	44	23	72	16	56				
Gender (M/F)	19/25	2/21	28/44	6/10	22/34	**0.01**	0.92	0.76	0.98
Age (yrs)	47±11	47±8	48±10	52 ±6	47±11	0.75	0.11	0.83	0.06
Education (yrs)	15.2±2	13±2	14±2	13±2	14±3	0.22	**0.007**	**0.02**	0.51
Disease duration(yrs)	-	14±6	15±10	14 ±9	16±9	0.42	-	-	0.57
EDSS median [IQR]	-	3.5[2.75-4.5]	2.5[2-3.625]	3.25 [2-4]	2.5 [2-3.5]	**0.02**	-	-	0.17
Cognitive assessment									
SDMT	54 ±9	46±6	48±12	38±9	51±11	0.53	**P < 0.001**	0.13	**0.0001**
VGLT	66 ±7	63±9	62±11	48±10	66 ±8	0.62	**P < 0.001**	0.82	**P < 0.001**
COWAT	19±4	18±5	15±5	12±4	16±4	0.03	**P < 0.001**	0.052	**0.004**
BVMT	29±5	25 ±6	25±7	17±5	27±5	0.97	**P < 0.001**	0.29	**P < 0.001**

Table 1. Description of subjects. We report the mean values and standard deviations from the different clinical parameters. For EDSS the median and interquartile range (IQR) is shown. The comparisons were performed using permutation testing with N = 5000 for all parameters except gender for which a chi-squared test was used. HC: healthy control, MS(BZD-)_CI: MS(BZD-) cognitively impaired, MS(BZD-)_CP: MS(BZD-) cognitively preserved.

3.2.2. Parcel level

Next, we extracted the 1/f slope for each parcel separately and compared them between MS(BZD+) and MS(BZD-). A significant difference in 1/f slope was observed in 26 brain parcels after correction for multiple comparisons (41). The MS(BZD+) had a steeper roll-off compared to MS(BZD-). As it is shown in Figure 3, pwMS treated with benzodiazepines had steeper slopes in all brain parcels.

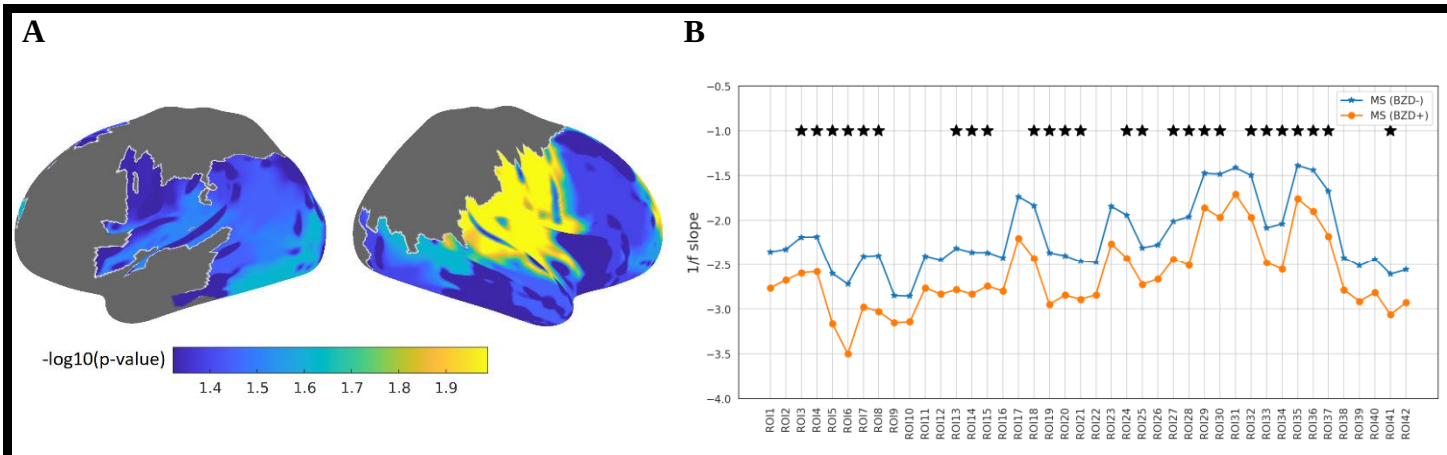

Figure 3. A) Comparisons of slope in the population of pwMS who have received benzodiazepines (MS(BZD+) and pwMS who have not received benzodiazepines (MS(BZD-). MS(BZD+) has steeper slopes compared with MS(BZD-) when comparing across brain parcels. **B)** Slopes were steeper in MS(BZD+) for all brain parcels. *p < 0.05 or -log10(p-value)>1.3, statistical significance after false discovery rate correction for multiple comparison.

3.3. Dynamic analysis of comparing MS populations: MS(BZD+) vs MS(BZD-)

Within each 30-second segment of the data, we compared the 1/f slope of the pwMS that were treated with benzodiazepines and those that were not treated with them. The results of this dynamic analysis demonstrate that the difference between these two groups remains significant over the complete recording session (Figure S1. Supplementary materials).

3.4. Spectral slope and beta activity

One study combining MEG data analysis and conductance-based neuronal network simulation has shown that cortical oscillations in the beta band are modulated by the GABAergic effect of

benzodiazepines through increasing the inhibitory post-synaptic currents (IPSCs) onto inhibitory neurons (42). To rule out the variations in periodic beta activity as a potential explanation for the observed group differences in spectral slope, we estimated the spectral power in the beta band. Figure 4 shows the cortical distributions of average beta power in the two groups of pwMS (MS(BZD+) and MS(BZD-)).

We observed that in both cohorts an increase in the periodic beta power corresponds to an increase in the negative slope of the spectra ($r = -0.61$, $p < 0.001$). However, there was no significant difference in average beta power between the two groups (p-value = 0.289, Mann-Whitney-U = 705.0, $\eta^2 = 0.01$, Cohen's d = 0.22). Effect sizes of differences in 1/f slope between two groups of pwMS for each brain region are quantified by the z-score of the U-statistic.

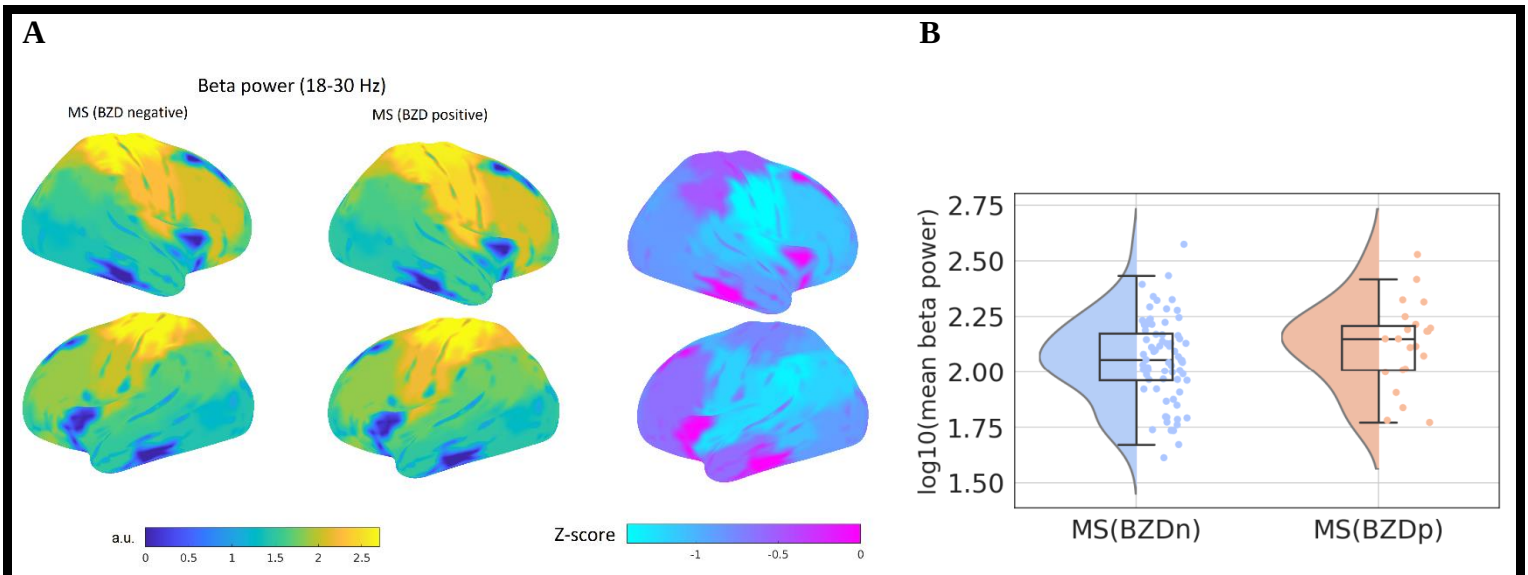

Figure 4. **A**) Cortical distributions of beta power (18-30 Hz). Distributions are shown in the two groups of pwMS. Top: right hemisphere, bottom: left hemisphere. a.u., arbitrary units. **B)** Mean beta power in each group of pwMS (p-value = 0.289, Mann-Whitney-U = 705.0, $\eta^2 = 0.01$, Cohen's d = 0.22).

3.5. Spectral slope and cognitive impairment

In the second part of this study, we investigated the relationship between 1/f slope as an index of excitation/inhibition balance and cognitive functioning in the group of MS(BZD-) patients. Based on the criteria defined above, we divided the MS(BZD-) into two groups (n =72): cognitively impaired (n =16) and cognitively preserved (n = 56). We then evaluated between-

group differences in averaged -across the whole brain- 1/f slope between 3 different groups: cognitively impaired pwMS, cognitively preserved pwMS, and healthy subjects. These 3 groups were matched in gender and age.

We observed a significant difference in slope between cognitively impaired pwMS and cognitively preserved pwMS (p-value = 0.013, Mann-Whitney-U = 262.0, η^2= 0.08, Cohen's d = 0.60). Average slopes were significantly steeper in the cognitively impaired pwMS compared to cognitively preserved pwMS. Similarly, we observed a significant difference in slope between cognitively impaired pwMS and healthy subjects (p-value = 0.029, Mann-Whitney-U = 482.0, η^2= 0.07 , Cohen's d = 0.58). Average slopes were significantly steeper in the cognitively impaired pwMS compared to healthy subjects.

We did not observe a significant difference in slope between cognitively preserved pwMS and healthy subjects (p-value = 0.50, Mann-Whitney-U = 1115.0, η^2 = 0.005, Cohen's d = 0.13).

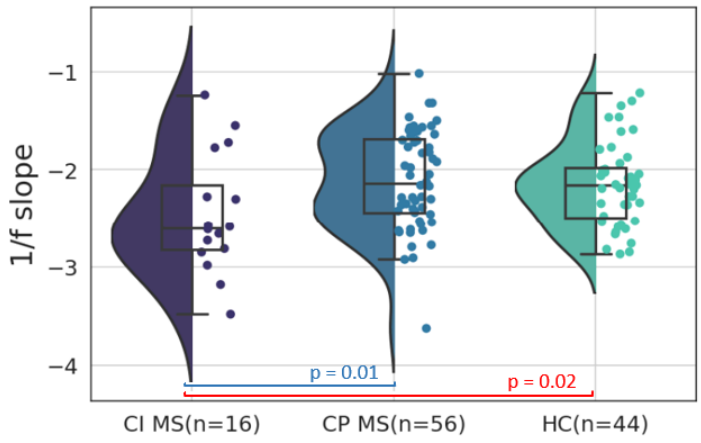

Figure 5. Between-group comparison in averaged 1/f spectral slope across the whole brain.
CI MS: cognitively impaired pwMS.
CP MS: cognitively preserved pwMS.
HC: healthy control subjects.

3.6. Correlation analysis between the 1/f slope and cognitive scores

To further explore the relationship between the 1/f slope and each cognitive score separately, we performed correlation analyses. We observed a significant correlation between the 1/f slope – averaged across the full brain – and BVMT (r=0.26, p=0.02) and VGLT (r=0.24, p=0.03) scores. A steeper slope was associated with lower BVMT and VGLT scores. There was no link between the 1/f slope and the SDMT or COWAT, neither in the whole brain nor in the individual parcels. All correlations were corrected for EDSS and disease duration. In general, the correlation between spectral slope and BVMT and VGLT was more significant in the brain parcels located in the prefrontal and parietal regions.

We repeated the correlation analysis after excluding three pwMS with the lowest VGLT scores (outliers). We did not observe a significant correlation between the 1/f slope – averaged across the full brain – and VGLT (r=0.22, p=0.07) scores. However, the slope was still significantly correlated in the brain parcels located in the prefrontal and parietal regions.

The correlation analysis was repeated for the group of pwMS who had received benzodiazepine treatment. We did not observe a correlation between 1/f slope and cognitive scores.

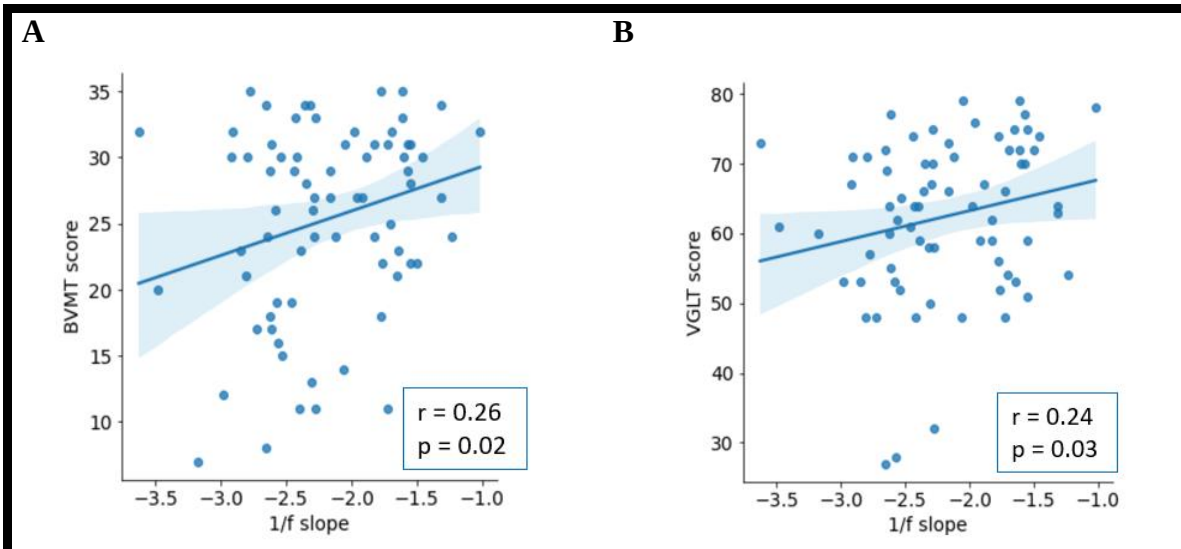

Figure 6. Correlation analysis between 1/f slope and BVMT scores (**A**) and VGLT scores (**B**). All correlations were calculated on the cohort of pwMS who have not treated with benzodiazepine: MS(BZD-).

4. Discussion

In this paper, we demonstrated that the 1/f slope is a non-invasive biomarker to assess changes in the excitation and inhibition ratio in pwMS. We first provided an additional validation by comparing the slope between pwMS with or without treatment of benzodiazepines and next demonstrated that cognitively impaired pwMS (without benzodiazepine treatment) also demonstrate a steeper slope and thus a decreased E/I ratio.

Since benzodiazepines reinforce the inhibitory effects of GABA and therefore increase inhibition, we expected and indeed observed a steeper spectral slope. This provides further evidence of the spectral slope as a non-invasive biomarker of the E/I ratio. The significant difference in 1/f exponent was observed in large parts of the brain, namely the occipital,

temporal, and prefrontal brain regions. In addition, while we focused on the first 2 minutes of data to reduce the potential effects of people falling asleep, we also show that this effect can be consistently observed across the full recording.

As benzodiazepines are well known to increase beta power (e.g. (42)), an important question is whether the differences in slope are a mere epiphenomenon of differences in beta power between the MS(BZD+) and MS(BZD-) group. To investigate this, we estimated the correlation between beta spectral power density (18-30 Hz) and 1/f slope (estimated in the 20-45 Hz band). Despite the presence of a significant correlation between 1/f slope and beta power ($r = -0.61$, $p<0.001$), we found that the latter did not reliably differentiate between pwMS who were treated with benzodiazepines and those who were not whereas the 1/f spectral slope did ($p>0.05$). This suggests that the spectral slope provides information beyond the spectral power of the traditional beta band.

As neuronal and synaptic loss are the well-known features of MS, we further investigated the association between the 1/f slope as an index of excitation/inhibition balance and cognitive functioning in the MS(BZD-) group. We focused on the MS(BZD-) group to avoid potential interference with the use of benzodiazepines in the analyses. We observed a significant correlation between 1/f spectral slope and verbal and spatial working memory, which was strongest in the regions involved in verbal and spatial memory, i.e. parietal and prefrontal regions. Although we had expected to observe flatter spectral slopes (less inhibition) to be associated with worse cognitive functioning, we observed steeper slopes. This suggests that a decrease in E/I ratio is associated with either a higher level of GABA signaling and inhibition or a lower level of excitatory signaling. Importantly, two clinical EEG studies evaluating aperiodic parameters in patients with schizophrenia and children with Attention-deficit/hyperactivity disorder (ADHD) have yielded similar findings (43,44). Although schizophrenia is associated with decreased GABAergic inhibition in the cortex (45), Peterson et al found steeper 1/f slopes in schizophrenia patients compared to healthy control subjects, suggesting a compensatory increase in GABAergic activity. This indicates that a compensatory mechanism may exist to compensate for the decreased number of inhibitory interneurons associated with multiple sclerosis (3). It is also possible that there is a range of 1/f slopes that are considered to be cognitively normal whereas both slopes that are too steep or too flat can indicate cognitive impairment (44).

5. Study limitations

While we did not have benzodiazepine dosage information or treatment duration the obtained results are clearly significant. It could be expected that assessing these effects in a larger group of people treated with benzodiazepines may further elucidate if the observed effects are mainly due to a specific benzodiazepine or rather reflect a common effect.

Another potential limitation of this study was the fact that most of the pwMS who received BZDs were female (21 vs 2). Therefore, we compared the 1/f slope of the 19 male and 25 female healthy control subjects matched in age (p-value = 0.48) to rule out the effect of gender on the 1/f slope. We did not observe a significant difference in 1/f spectral slope between male and female healthy subjects (p-value = 0.76, Mann-Whitney-U = 224, η^2= 0.002, Cohen's d = 0.09). The pwMS who received BZDs also had a slightly higher Expanded Disability Status Scale (EDSS) score. Yet, including the EDSS in an ANCOVA model (46) did not alter the results (p-value = 0.021).

A final limitation is the acquisition of MEG data on two (slightly) different scanners. To investigate the effect of different scanners, we compared the whole brain averaged 1/f slope between 13 healthy subjects and 31 healthy subjects scanned by the first and the second scanner respectively. We did not observe a significant difference in 1/f spectral slope between two healthy groups of the first scanner and second scanner (p-value = 0.27, Mann-Whitney-U = 158, η^2= 0.02, Cohen's d = 0.34).

6. Conclusion

Traditionally, EEG and MEG studies have concentrated on group differences in oscillatory dynamics, but new research underlines the importance of the 1/f slope as a potential biomarker for the excitation/inhibition ratio (8). In this study, we highlighted the utility of 1/f spectral slope to quantify the inhibitory effect of benzodiazepine usage in pwMS. We also demonstrated a correlation between the 1/f spectral slope and cognitive performance in pwMS who are not using benzodiazepines. To the best of our knowledge, this is the first study to investigate the 1/f spectral slope in people with multiple sclerosis.

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Declaration of conflicting interests. The authors report no competing interests.

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Data availability. Data is available upon reasonable request to the corresponding author.

References:

1. Dendrou CA, Fugger L, Friese MA. Immunopathology of multiple sclerosis. Nat Rev Immunol [Internet]. 2015 Sep 7;15(9):545–58. Available from: http://www.nature.com/articles/nri3871

2. di Filippo M, Portaccio E, Mancini A, Calabresi P. Multiple sclerosis and cognition: synaptic failure and network dysfunction. Nat Rev Neurosci [Internet]. 2018 Oct 29;19(10):599–609. Available from: http://www.nature.com/articles/s41583-018-0053-9

3. Mandolesi G, Gentile A, Musella A, Fresegna D, de Vito F, Bullitta S, et al. Synaptopathy connects inflammation and neurodegeneration in multiple sclerosis. Nat Rev Neurol [Internet]. 2015 Dec 20;11(12):711–24. Available from: http://www.nature.com/articles/nrneurol.2015.222

4. Stam CJ. Modern network science of neurological disorders. Nat Rev Neurosci [Internet]. 2014 Oct 4;15(10):683–95. Available from: http://www.nature.com/articles/nrn3801

5. Huiskamp M, Kiljan S, Kulik S, Witte ME, Jonkman LE, GJM Bol J, et al. Inhibitory synaptic loss drives network changes in multiple sclerosis: An ex vivo to in silico translational study. Multiple Sclerosis Journal [Internet]. 2022 Nov 3;28(13):2010–9. Available from: http://journals.sagepub.com/doi/10.1177/13524585221125381

6. Sengupta B, Laughlin SB, Niven JE. Balanced Excitatory and Inhibitory Synaptic Currents Promote Efficient Coding and Metabolic Efficiency. PLoS Comput Biol. 2013;9(10).

7. Bhatia A, Moza S, Bhalla US. Precise excitation-inhibition balance controls gain and timing in the hippocampus. Elife. 2019;8:1–29.

8. Gao R, Peterson EJ, Voytek B. Inferring synaptic excitation/inhibition balance from field potentials. Neuroimage. 2017 Sep 1;158:70–8.

9. Chini M, Pfeffer T, Hanganu-Opatz I. An increase of inhibition drives the developmental decorrelation of neural activity. Elife [Internet]. 2022 Aug 17;11. Available from: https://elifesciences.org/articles/78811

10. Lendner JD, Helfrich RF, Mander BA, Romundstad L, Lin JJ, Walker MP, et al. An electrophysiological marker of arousal level in humans. Elife. 2020 Jul 1;9:1–29.

11. Miskovic V, MacDonald KJ, Rhodes LJ, Cote KA. Changes in EEG multiscale entropy and power-law frequency scaling during the human sleep cycle. Hum Brain Mapp [Internet]. 2019 Feb 1;40(2):538–51. Available from: https://onlinelibrary.wiley.com/doi/10.1002/hbm.24393

12. Waschke L, Donoghue T, Fiedler L, Smith S, Garrett DD, Voytek B, et al. Modality-specific tracking of attention and sensory statistics in the human electrophysiological spectral exponent. Available from: https://doi.org/10.1101/2021.01.13.426522

13. Colombo MA, Napolitani M, Boly M, Gosseries O, Casarotto S, Rosanova M, et al. The spectral exponent of the resting EEG indexes the presence of consciousness during unresponsiveness induced by propofol, xenon, and ketamine. Neuroimage [Internet]. 2019 Apr;189:631–44. Available from: https://linkinghub.elsevier.com/retrieve/pii/S1053811919300242

14. Voytek B, Kramer MA, Case J, Lepage KQ, Tempesta ZR, Knight RT, et al. Age-related changes in 1/f neural electrophysiological noise. Journal of Neuroscience. 2015 Sep 23;35(38):13257–65.

15. Schaworonkow N, Voytek B. Longitudinal changes in aperiodic and periodic activity in electrophysiological recordings in the first seven months of life. Dev Cogn Neurosci [Internet]. 2021;47:100895. Available from: https://doi.org/10.1016/j.dcn.2020.100895

16. Molina JL, Voytek B, Thomas ML, Joshi YB, Bhakta SG, Talledo JA, et al. Memantine Effects on Electroencephalographic Measures of Putative Excitatory/Inhibitory Balance in Schizophrenia. Biol Psychiatry Cogn Neurosci Neuroimaging [Internet]. 2020 Jun;5(6):562–8. Available from: https://linkinghub.elsevier.com/retrieve/pii/S2451902220300471

17. Kosciessa JQ, Lindenberger U, Garrett DD. Thalamocortical excitability modulation guides human perception under uncertainty. Nat Commun [Internet]. 2021 Dec 23;12(1):2430. Available from: http://www.nature.com/articles/s41467-021-22511-7

18. Ostlund BD, Alperin BR, Drew T, Karalunas SL. Behavioral and cognitive correlates of the aperiodic (1/f-like) exponent of the EEG power spectrum in adolescents with and without ADHD. Dev Cogn Neurosci. 2021 Apr;48:100931.

19. Ian Ramsay PLELBSDLSS. Disturbances in aperiodic neural activity during resting state in patients with schizophrenia. Biol Psychiatry. 2021 May 1;89(9):S254–5.

20. Wang Z, Mo Y, Sun Y, Hu K, Peng C, Zhang S, et al. Separating the aperiodic and periodic components of neural activity in Parkinson's disease. European Journal of Neuroscience. 2022 Sep 22;56(6):4889–900.

21. Polman CH, Reingold SC, Banwell B, Clanet M, Cohen JA, Filippi M, et al. Diagnostic criteria for multiple sclerosis: 2010 Revisions to the McDonald criteria. Ann Neurol [Internet]. 2011 Feb 8;69(2):292–302. Available from: https://onlinelibrary.wiley.com/doi/10.1002/ana.22366

22. Kurtzke JF. Rating neurologic impairment in multiple sclerosis: An expanded disability status scale (EDSS). Neurology [Internet]. 1983 Nov 1;33(11):1444–1444. Available from: https://www.neurology.org/lookup/doi/10.1212/WNL.33.11.1444

23. Taulu S, Simola J, Kajola M. Applications of the signal space separation method. IEEE Transactions on Signal Processing. 2005 Sep;53(9):3359–72.

24. JENKINSON M et al. BET2: MR-based estimation of brain, skull and scalp surfaces. In: Eleventh annual meeting of the organization for human brain mapping. 2005;167.

25. Smith SM. Fast robust automated brain extraction. Hum Brain Mapp. 2002 Nov;17(3):143–55.

26. Mazziotta JC, Toga AW, Evans A, Fox P, Lancaster J. A Probabilistic Atlas of the Human Brain: Theory and Rationale for Its Development. Neuroimage. 1995 Jun;2(2):89–101.

27. Oostenveld R, Fries P, Maris E, Schoffelen JM. FieldTrip: Open Source Software for Advanced Analysis of MEG, EEG, and Invasive Electrophysiological Data. Comput Intell Neurosci. 2011;2011:1–9.

28. Quinn AJ, Vidaurre D, Abeysuriya R, Becker R, Nobre AC, Woolrich MW. Task-Evoked Dynamic Network Analysis Through Hidden Markov Modeling. Front Neurosci. 2018 Aug 28;12.

29. Woolrich M, Hunt L, Groves A, Barnes G. MEG beamforming using Bayesian PCA for adaptive data covariance matrix regularization. Neuroimage. 2011 Aug;57(4):1466–79.

30. Vidaurre D, Hunt LT, Quinn AJ, Hunt BAE, Brookes MJ, Nobre AC, et al. Spontaneous cortical activity transiently organises into frequency specific phase-coupling networks. Nat Commun. 2018 Dec 30;9(1):2987.

31. van Schependom J, Vidaurre D, Costers L, Sjøgård M, D'hooghe MB, D'haeseleer M, et al. Altered transient brain dynamics in multiple sclerosis: Treatment or pathology? Hum Brain Mapp. 2019 Nov 1;40(16):4789–800.

32. Donoghue T, Haller M, Peterson EJ, Varma P, Sebastian P, Gao R, et al. Parameterizing neural power spectra into periodic and aperiodic components. Nat Neurosci. 2020 Dec 1;23(12):1655–65.

33. A smith. The symbol-digit modalities test: a neuropsychological test of learning and other cerebral disorders. Learning Disorders. 1968;83–91.

34. Strober L, Englert J, Munschauer F, Weinstock-Guttman B, Rao S, Benedict R. Sensitivity of conventional memory tests in multiple sclerosis: comparing the Rao Brief Repeatable Neuropsychological Battery and the Minimal Assessment of Cognitive Function in MS. Multiple Sclerosis Journal [Internet]. 2009 Sep 25;15(9):1077–84. Available from: http://journals.sagepub.com/doi/10.1177/1352458509106615

35. Ruff RM, Light RH, Parker SB, Levin HS. Benton controlled oral word association test: Reliability and updated norms. Archives of Clinical Neuropsychology [Internet]. 1996 Jan 1;11(4):329–38. Available from: https://academic.oup.com/acn/article-lookup/doi/10.1093/arclin/11.4.329

36. Benedict RHB, Schretlen D, Groninger L, Dobraski M, Shpritz B. Revision of the Brief Visuospatial Memory Test: Studies of normal performance, reliability, and validity. Psychol Assess [Internet]. 1996;8(2):145–53. Available from: http://doi.apa.org/getdoi.cfm?doi=10.1037/1040-3590.8.2.145

37. Penner I, Raselli C, Stöcklin M, Opwis K, Kappos L, Calabrese P. The Fatigue Scale for Motor and Cognitive Functions (FSMC): validation of a new instrument to assess multiple sclerosis-related fatigue. Multiple Sclerosis Journal. 2009 Dec 7;15(12):1509–17.

38. BECK AT. An Inventory for Measuring Depression. Arch Gen Psychiatry. 1961 Jun 1;4(6):561.

39. Fischer M, Kunkel A, Bublak P, Faiss JH, Hoffmann F, Sailer M, et al. How reliable is the classification of cognitive impairment across different criteria in early and late stages of multiple sclerosis? J Neurol Sci. 2014 Aug 15;343(1–2):91–9.

40. Costers L, Gielen J, Eelen PL, Schependom J van, Laton J, Remoortel A van, et al. Does including the full CVLT-II and BVMT-R improve BICAMS? Evidence from a Belgian (Dutch) validation study. Mult Scler Relat Disord. 2017 Nov 1;18:33–40.

41. Benjamini Y, Hochberg Y. Controlling the False Discovery Rate: A Practical and Powerful Approach to Multiple Testing. Journal of the Royal Statistical Society: Series B (Methodological). 1995 Jan;57(1):289–300.

42. JENSEN O, GOEL P, KOPELL N, POHJA M, HARI R, ERMENTROUT B. On the human sensorimotor-cortex beta rhythm: Sources and modeling. Neuroimage. 2005 Jun;26(2):347–55.

43. Peterson EJ, Rosen BQ, Belger A, Voytek B, Campbell D A Neurosciences AM, Program G. Aperiodic neural activity is a better predictor of schizophrenia than neural oscillations Abv. title: Spectral slope predicts schizophrenia. Available from: https://doi.org/10.1101/113449

44. Robertson MM, Furlong S, Voytek B, Donoghue XT, Boettiger CA, Sheridan MA. EEG power spectral slope differs by ADHD status and stimulant medication exposure in early childhood. J Neu-rophysiol [Internet]. 2019;122:2427–37. Available from: www.jn.org

45. Lewis DA, Hashimoto T, Volk DW. Cortical inhibitory neurons and schizophrenia. Nat Rev Neurosci. 2005 Apr;6(4):312–24.

46. Bowman AW AA. R package sm: nonparametric smoothing methods (version 2.2-5.4). University of Glasgow, UK and Universita di padova, Italia.; 2014.

Supplementary materials

Fahimeh Akbarain et al. The spectral slope as a marker of excitation/inhibition ratio and cognitive functioning in multiple sclerosis.

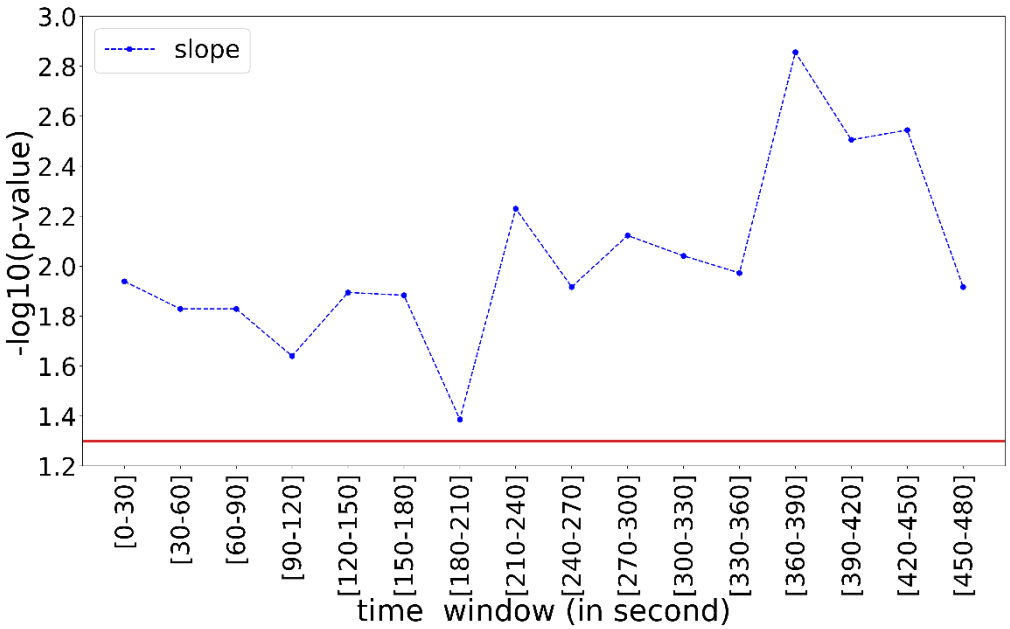

Figure S1. Dynamic analysis. On the y-axis, the p-values of the comparison of 1/f slope between MS(BZD+) and MS(BZD-) for different piece of data. One can clearly appreciate the difference being present in every piece data. Horizontal red line indicates the cut-off value of 1.3. The values of -log10(p-value) above 1.3 correspond to significant results.

Table S1. List of regions used in the parcel level analyses.

	ROI
'ROI 1'	'L Cuneus'
'ROI 2'	'R Cuneus'
'ROI 3'	' L Inf Occ'
'ROI 4'	'R Inf Occ'
'ROI 5'	'L Supramarginal'
'ROI 6'	'R Supramarginal'
'ROI 7'	'L Sup Temp'
'ROI 8'	'R Sup Temp'
'ROI 9'	'L Lat SMC'
'ROI 10'	'R Lat SMC'
'ROI 11'	'L Sup Parietal'
'ROI 12'	'R Sup Parietal'
'ROI 13'	'L Middle Occ'
'ROI 14'	'R Middle Occ'
'ROI 15'	'L Sup Occ'
'ROI 16'	'R Sup Occ'
'ROI 17'	'L Ant Temp'
'ROI 18'	'R Ant Temp'
'ROI 19'	'L Medial SMC'
'ROI 20'	'R Medial SMC'
'ROI 21'	'L Angular'
'ROI 22'	'R Angular'
'ROI 23'	'L VL PFC'
'ROI 24'	'R VL PFC'
'ROI 25'	'L Occ pole'
'ROI 26'	'R Occ pole'
'ROI 27'	'L Sup PFC'
'ROI 28'	'R Sup PFC'
'ROI 29'	'L Sup Dorsal PFC'
'ROI 30'	'R Sup Dorsal PFC'
'ROI 31'	'L Orbitofrontal'
'ROI 32'	'R Orbitofrontal'
'ROI 33'	'L Post Temp'
'ROI 34'	'R Post Temp'
'ROI 35'	'L Inf Dorsal PFC'
'ROI 36'	'R Inf Dorsal PFC'
'ROI 37'	'Medial PFC'
'ROI 38'	'Post Precuneus'
'ROI 39'	' Posterior Cingulate Cortex '
'ROI 40'	'Ant Precuneus'
'ROI 41'	'L Inf Parietal'
'ROI42'	'R Inf Parietal'