Resting-State EEG Microstates as Dynamic Biomarkers of Network Dysfunction and Cognitive Impairment in Multiple Sclerosis

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Introduction

Multiple Sclerosis (MS) progression is characterised by **large-scale brain network dysfunctions**, yet **not fully captured by conventional clinical and MRI measures.**

EEG **microstates** analysis offers a a cost-effective, non-invasive, high-temporal-resolution approach to track these dynamics

Aim

- **To assess whether EEG microstate metrics distinguish MS patients from healthy volunteers (HVs).**
- **To determine whether they differentiate cognitively impaired (CI) from cognitively preserved (CP) patients.**
- **To explore associations between microstate features and clinical disability.**

Methods

We included **88 patients with MS** (2017 McDonald criteria, EDSS 0–3.0, age 18–55, relapse-free ≥30 days) and 46 HVs.
Cognitive status was classified using the Brief Repeatable Battery of Neuropsychological Tests (CI if ≤5th percentile in ≥1 test). Clinical assessment included disease duration, treatment, EDSS, 9HPT, T25FW, and SDMT.

Resting-state eyes-open EEG (32 channels, 5 min) was recorded and preprocessed with EEGLAB and ICA-based artifact removal.

Microstate analysis was performed on 2–20 Hz filtered data using k-means clustering at GFP peaks, with a 6-class solution **(A, B, C, D, F, G)** applied across all participants. For each class, explained variance, mean duration, occurrence, and coverage were extracted.

For the **primary analysis**, a subgroup of patients with **MS** was age- and sex-matched to **HVs**. A **secondary analysis** compared patients with **CI to CP patients.**

Group differences in **temporal** parameters were tested with Mann–Whitney U and topographical differences with **TANOVA**. Stepwise linear discriminant analysis (LDA) was used for classification. Associations with clinical measures were explored with partial correlations controlling for age and education.

Results

Table 1. Demographic and Clinical Characteristics of HVs and MS

Variable	HVs	MS	Statistic (p-value)
N	45	46	
Age, mean (SD)	33.7 (10.6)	34.6 (9.6)	t = -0.44 (0.658)
Sex (M/F)	22/23	21/25	Fisher p = 0.835
Education, median [IQR]	18.0 [3.0]	17.0 [5.0]	Z = 1.42 (0.157)
Disease duration, median [IQR]		5.5 [9.0]	
EDSS, median [IQR]		1.2 [1.0]	
9HPT, median [IQR]		21.4 [4.8]	
SDMT, median [IQR]		48.5 [11.0]	
T25FW, median [IQR]		4.8 [1.1]	
Cognitively Impaired (n)		19	
Cognitively Preserved (n)		27	
DMT (Naive/platform/S1P/Cla/mAb)		3/11/2/3/27	

Table 2. Demographic and Clinical Characteristics of CP and CI patients

Variable	CP	CI	Statistic (p-value)
N	31	57	
Age, mean (SD)	35.5 (9.6)	46.7 (11.6)	t = -4.86 (< 0.001)
Sex (M/F)	11/20	17/40	Fisher p = 0.636
Education, median [IQR]	13.0 [5.0]	16.0 [5.0]	Z = -0.59 (0.557)
Disease duration, median [IQR]	5.0 [8.8]	12.0 [16.0]	Z = -3.56 (< 0.001)
EDSS, median [IQR]	1.5 [1.8]	2.0 [2.6]	Z = -2.65 (0.008)
9HPT, median [IQR]	21.1 [2.7]	23.0 [4.6]	Z = -4.27 (< 0.001)
SDMT, median [IQR]	50.0 [8.5]	41.0 [14.2]	Z = 4.14 (< 0.001)
T25FW, median [IQR]	4.6 [0.9]	6.1 [4.5]	Z = -4.70 (< 0.001)
DMT (Naïve-No/platform/S1P/Cla/mAb)	1/4/0/2/24	5/17/10/7/18	

Results

Fifty-seven (68%) MS patients showed CI in at least one domain, most frequently in processing speed and working memory **(Table 1 and 2).**

- **MS (46) vs HVs (45):** Patients with MS displayed increased presence of microstate B (visual network) and reduced duration of microstates C (posterior DMN), F (salience) and G (sensorimotor) **(Fig.1).** LDA using microstates B and D (DAN) classified MS vs HVs with ~77% accuracy. No topographic differences **(Fig.3A).**

- **CI (57) vs CP (31) patients**: CI patients showed altered topographies for microstates C, F, and G **(Fig.3B)**, with significantly reduced expression of microstate F **(Fig.2).** This alone discriminated CI from CP with ~65% accuracy.

- **Clinical correlations:** Class A presence correlated with longer disease duration, higher EDSS, and worse ambulation. Class D inversely correlated with EDSS, while Class B presence was associated with poorer information processing speed **(Fig.4).**

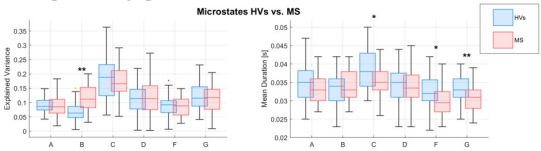

Fig.1

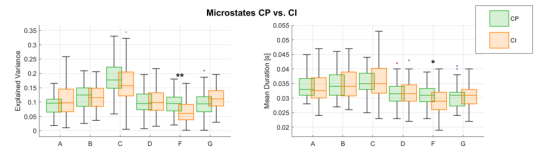

Fig.2

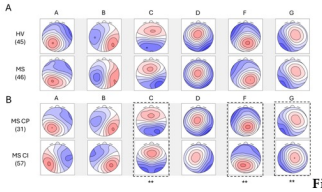

Fig.3

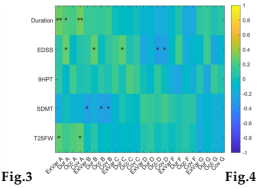

Fig.4

Conclusions

EEG microstates reveal early and clinically relevant alterations of large-scale brain dynamics in MS. Increased expression of sensory networks and reduced stability of cognitive/salience networks differentiate patients from healthy volunteers and are linked to disability and cognitive impairment. Class F (salience) alterations strongly characterize cognitive decline, while class B (visual) and D (DAN) features discriminate MS from HVs with good accuracy.

These findings support microstates as promising tools for diagnosis, monitoring, and stratification in MS, warranting longitudinal validation.

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