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Multiple brain networks support processing speed abilities of patients with multiple sclerosis

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Abstract

Objectives: Many people affected by multiple sclerosis (MS) experience cognitive impairment, especially decreases in information processing speed (PS). Neural disconnection is thought to represent the neural marker of this symptom, although the role played by alterations of specific functional brain networks still remains unclear. The aim is to investigate and compare patterns of association between PS-demanding cognitive performance and functional connectivity across two MS phenotypes.

Methods: Forty patients with relapsing-remitting MS (RRMS) and twenty-five with secondary progressive MS (SPMS) had neuropsychological and MRI assessments. Multiple regression models were used to investigate the relationship between performance on tests of visuo-motor and verbal PS, and on the verbal fluency tests, and functional connectivity of four cognitive networks, i.e. left and right fronto-parietal, salience and default-mode, and two control networks, i.e. visual and sensorimotor.

Results: Patients with SPMS were older and had longer disease history than patients with RRMS and presented with worse overall clinical conditions: higher disease severity, total lesion volume and cognitive impairment rates. However, in both patient samples, cognitive performance across tests was negatively correlated with functional connectivity of the salience and default-mode networks, and positively with connectivity of the left fronto-parietal network. Only the visuo-motor PS scores of the RRMS group were also associated with connectivity of the sensorimotor network.

Conclusions: PS-demanding cognitive performance in patients with MS appears mainly associated with strength of functional connectivity of frontal networks involved in evaluation and manipulation of information, as well as the default mode network. These results appear in line with the hypothesis that multiple neural networks may be needed to support normal

cognitive performance across MS phenotypes. However, different PS measures showed partially different patterns of association with functional connectivity. Therefore, further investigations are needed to clarify the contribution of inter-network communication to specific cognitive deficits due to MS.

Keywords

MS phenotypes; salience; default mode; cognition; functional connectivity; disconnection

1. Introduction

Cognitive impairment due to multiple sclerosis (MS) represents a pervasive symptom with negative consequences on quality of life of patients [1]. Processing speed (PS), attention and memory are the most commonly affected domains [2]. PS deficits have long been considered as the most characteristic cognitive sign of MS, possibly driving decline in other cognitive functions [3]. However, the cognitive profile of patients with MS has been recently shown to be highly heterogeneous, since subgroups of patients may present with deficits either in isolated or multiple cognitive domains [4].

A possible explanation of such variability in the observed symptomatology may relate to the fact that MS neuropathological hallmarks, i.e. demyelinating lesions, spread randomly and affect all brain tissues [5], thus causing a wide range of deficits according to their location [6]. In fact, global volumetric indices of lesioned tissue and atrophy are only mildly associated with declining performance in specific cognitive domains, such as PS function [7]. Currently, neural disconnection is considered the general mechanism underlying cognitive dysfunction in MS [8,9], but little is known about the neural correlates of PS and other specific cognitive deficits.

Functional magnetic resonance imaging (MRI) has been used to investigate how networks of functionally related brain areas at rest rearrange as a consequence of MS and how these alterations may be linked to declining cognitive performance. Many studies focused on the so-called Default Mode Network (DMN), a set of areas that usually deactivates when engagement in goal-directed behaviors is requested [10]. In people with relapsing-remitting MS (RRMS) and cognitive impairment this network appears to become more central in the brain organization, probably suggesting an inability to shift brain activation towards other task-related networks [11]. However, alterations in functional connectivity in cognitively

impaired patients have been observed both in the anterior [12] and posterior hubs of the DMN [13].

More specific analyses of the resting-state functional neural correlates of MS-related PS deficits have been carried out by a handful of studies with quite variable and inconsistent outcomes [14]. The Paced Auditory Serial Addition Test (PASAT) has been extensively used as a measure of PS function of people with RRMS and performance on this task appears to be associated with functional connectivity of several distinct brain networks: the DMN [15], the executive control and the medial visual networks [16], and the thalamic network as reported by Tona et al. [17] but not by Zhou et al. [18].

Consistent findings across studies point at a preferential involvement of the left hemisphere in supporting PS functions in MS [15,19,20]. This may be explained by the fact that most PS tests used in these studies required processing of verbal information. However, the role of the left fronto-parietal network in cognitive decline caused by MS is still unclear since it appears to be altered also in cognitively preserved patients and deficits may emerge as a consequence of a more widespread disruption [21].

More limited is the evidence in secondary progressive MS (SPMS), although PS deficits have been consistently reported to a greater extent in people with SPMS compared to those with RRMS [22-26]. Resting-state functional MRI analyses revealed both decreases [27] and increases [28] in resting-state activity of the DMN in people with SPMS compared to healthy controls. Nonetheless, both studies revealed a similar involvement of functional connectivity of the anterior cingulate cortex (as part of the DMN) in performance of patients on the PASAT, but not of the sensorimotor network [28]. Instead, the role of other networks, e.g. the salience network [29] and the fronto-parietal networks [30], in MS-related PS function decline has been scarcely or not investigated, although the correct functioning of these

networks is to some degree connected to the correct functioning of the DMN and successful task execution depends on the coordination of their activation and deactivation patterns [31]. Therefore, this study aimed at investigating: 1) whether alternative measures of PS function correlate differentially with functional connectivity of various brain networks; 2) which networks are preferentially associated with PS abilities in people with MS; and 3) whether such patterns of associations differ across MS phenotypes.

2. Methods

2.1. Participants

People with MS recruited for two prospective studies were included in the present work: forty people affected by RRMS and 25 with SPMS [32] were recruited from the MS Clinic at Sheffield Teaching Hospital NHS Foundation Trust (Sheffield, UK) and the MS Clinic of the IRCCS Fondazione Ospedale San Camillo (Venice, Italy), respectively. Inclusion criteria for both studies are reported in **Table 1**. These studies were carried out according to the Declaration of Helsinki and ethical approval was obtained from the Yorkshire and Humber Regional Ethics Committee Ref No: 12/YH/0474 (protocol version 4.0) and the Institutional Review Board of the IRCCS Fondazione Ospedale San Camillo (Venice, Italy) (Ref No. 11/09 version 2). All participants were provided with written information material prior to recruitment and gave written consent to take part in these studies.

- Insert Table 1 about here -

2.2. Neuropsychological assessment

Global cognitive status was assessed by means of the Mini Mental State Examination [33] for people with RRMS and by means of the Raven's Colored Progressive Matrices [34] for

people with SPMS. Additionally, a neuropsychological battery was administered to both patient cohorts (in English for people with RRMS and in Italian for people with SPMS) and included four tests with high PS demands from which we derived different indices of PS function, either in terms of the amount of information processed per time unit or of the time needed to processes a given amount of information:

- the Trail Making Test - part A (TMT-A) [35]: the total time (sec) to connect with straight lines a series of 25 numbered dots on a sheet of paper;
- the short version of the Stroop test [36]: the average of the completion time (sec) on the first two trials (word reading and color naming);
- the Semantic and Phonemic Fluency tests [37]: the total count of words recalled for three categories (cities, animals and fruits) and three letters (F, L and P) over a period of time of 60 seconds for each trial.

Cognitive deficits were defined as performance at least 2 standard deviations below (for the Fluency tasks) or above (for the TMT-A and the Stroop Test) the mean of normative data from age-matched healthy controls. For the RRMS cohort, normative data from a local database of healthy controls were used, while for the SPMS cohort Italian norms were used for the Fluency tasks [38], the TMT-A [39] and the Stroop test [36].

2.3. MRI acquisition and pre-processing

All participants underwent an MRI scanning session. The scanner characteristics and the protocols used for the two studies are described in **Table 2**.

- Insert Table 2 about here -

All MRI analyses were carried out in parallel, but independently, for the two studies by means of the Statistical Parametric Mapping software (SPM 12, Wellcome Centre for Human Neuroimaging, London, UK) running on MATLAB R2008a, version 7.6.0 (The Mathworks, Natick, Massachusetts, USA). Reoriented T1-weighted and Fluid-Attenuated Inversion Recovery (FLAIR) scans were fed to the Lesion Segmentation Toolbox v1.2.3 [40] and white matter lesions were automatically segmented by means of the lesion growth algorithm (threshold k = 0.3). These two types of images were automatically combined to improve segmentation of white matter lesions and the output was visually inspected to ensure no lesions were missed and that normal appearing white matter was not included in segmented lesions. The lesion-filled T1-weighted images were segmented into grey matter, white matter and cerebrospinal fluid. The volume in millilitres of all segmented images was extracted by means of the MATLAB function "get_totals" and their sum provided the total intracranial volume.

All resting-state scans were first slice-time corrected and realigned to correct for possible artifacts. Head movements did not exceed ±3 mm and ±3° for any of the participants. Realigned images were normalized to the default echo-planar imaging template and voxel size was isotropied at 2.0 × 2.0 × 2.0 mm to correct for possible inter-individual differences in head size and shape. Subsequently, the REST toolbox [41] was used to apply a band-pass filter (0.008-0.1 Hz) to remove non-neural frequencies [42]. Finally, images were spatially smoothed (6 mm) to improve signal-to-noise ratio. After pre-processing, group-level independent component analysis was performed on resting-state MRI scans using the GIFT toolbox for SPM12 (GIFT v1.3i; mialab.mrn.org/software/gift) [43] to separate independent sources of signal and identify networks of functionally related areas. The Infomax algorithm was used and the number of components to be extracted was set to twenty [44]. Finally, spatial maps of each component were reconstructed for each participant.

Four functional networks were visually identified and selected for statistical analyses: the default mode network, the salience network, the right and left fronto-parietal networks. The visual and the sensorimotor networks were selected as control networks due to their prevalent involvement in non-cognitive functions.

2.4. Statistical analyses

Independent sample t-test and Mann-Whitney U test analyses were carried out to compare demographic, clinical and global neural characteristics of the two samples using SPSS Statistics Version 21 (IBM, Chicago, IL, USA). Additionally, English and Italian normative values were used to quantify the total count of people with cognitive impairment in each neuropsychological test and χ^2 was used to assess differences in rates of cognitive impairment between MS phenotypes. Indeed, this procedure was favored to an independent sample t-test due to the fact that verbally based tests are sensitive to language and culture, thus undermining a direct between-group comparison of raw scores.

Multiple regression models were created in SPM12 to investigate the association between PS measures and the maps of each one of the six functional networks for both RRMS and SPMS cohorts. Age was used as covariate of no interest to rule out the effect of ageing processes on cognition that may be independent of any MS pathological changes [45]. Similarly, education and total intracranial volume were included in the analyses to account for possible effects of cognitive [46] and brain reserve [475].

Firstly, analyses of the relationship between the DMN and PS measures were carried in consideration of the prominent role of this network in the literature. Secondly, the salience network and the fronto-parietal networks were analyzed to ascertain their possible association with PS abilities of people with MS beyond the DMN. Finally, the visual and the

sensorimotor networks were investigated to test the specificity of the association between PS performance and functional connectivity of cognitive networks.

Only clusters that survived statistical correction for multiple comparisons at a Family Wise Error (FWE) threshold of $p < 0.05$ were considered. Peaks within significant clusters of gray matter were identified with the Talairach Daemon (http://www.talairach.org/daemon.html), after converting their coordinates from the Montreal Neurological Institute (MNI) to the Talairach reference system.

3. Results

3.1. Clinical and cognitive results

The Shapiro-Wilk test was used to perform a preliminary check of the clinical data and only age, total intracranial and gray matter volumes resulted to be normally distributed. Therefore, independent sample t-tests were used to compare these variables across MS phenotypes, while Mann-Whitney U tests were used for the remaining variables.

All results on cognitive and clinical characteristics of the samples of patients are reported in **Table 3**. People with SPMS were older and less educated and presented with a globally worse profile than the RRMS group. Although having higher total intracranial volume, as expected, the SPMS group presented with lower gray matter volume and higher cerebrospinal fluid volume. In general, people with SPMS showed higher rates of cognitive deficits in almost all PS-demanding tests except for the Stroop speed index.

- Insert Table 3 about here -

3.2. Resting-state fMRI results

First, correlations between scores of all the cognitive tests and functional connectivity maps of the four cognitive networks were investigated. The left fronto-parietal network was observed to be the one most consistently associated with PS-demanding cognitive performance across a range of different tests in both RRMS (**Table 4** and **Figure 1**) and SPMS groups (**Table 5** and **Figure 2**). In particular, performance on the Stroop test was negatively correlated with functional connectivity of the left fronto-parietal network, differentially across MS phenotypes: in the right posterior cingulate cortex in the RRMS group and in the left inferior frontal gyrus in the SPMS groups.

- Insert Table 4 and Figure 1 about here –

Strength of connectivity of the DMN in the cerebellum was negatively associated with performance of people with RRMS on the Semantic Fluency test. Instead, functional coupling of the salience network in different fronto-parietal areas was shown to be positively correlated with TMT-A scores in the RRMS group and Stroop test scores in the SPMS group.

- Insert Table 5 and Figure 2 about here -

Additionally, analyses carried out on the two control networks revealed that TMT-A scores obtained by people with RRMS correlated with functional connectivity of the sensorimotor network in motor areas, positively, and in the right inferior frontal gyrus, negatively (**Table 6** and **Figure 3**). No significant associations between cognitive performance and functional connectivity of either the visual or the sensorimotor networks were observed for people with SPMS.

- Insert Table 6 and Figure 3 about here -

4. Discussion

The results of this study suggest that the left fronto-parietal network, a set of brain areas involved in attentional and executive processes necessary to coordinate other cognitive functions [30], may play a central role in supporting performance of people with MS on tests requiring fast information processing. In fact, this network emerged consistently in the analyses carried out on two cohorts of patients affected by RRMS and SPMS and across tests characterized by PS demands. Significant findings were highlighted for the Stroop speed index in both patient groups and, in the RRMS group only, for more complex PS measures, i.e. the Fluency tasks. However, it was systematically noticed that better PS performance was correlated with stronger functional connectivity of the left fronto-parietal network in frontal and parieto-limbic areas that differed across tests. These findings support the hypothesis that efficient integration of information across several areas involved in cognitive control contributes to faster processing, yet with possible differences in distinct phases of the disease. Indeed, the dorsal posterior cingulate, found to be linked to performance on the Stroop speed index in the RRMS group, is connected at rest with attentional networks and may influence attention allocation [48]. Instead, faster performance on the same test in the SPMS group was associated with lower functional connectivity of this network in the left inferior frontal gyrus: an area associated with oral verbal production and semantic processing [49,50] as well as cognitive control [51]. These results appear consistent with the identification of different frontal WM tracts as the main structural neural correlates of PS performance of people with SPMS [52].

The right fronto-parietal network was not associated with any PS-demanding tasks in any of the two patient groups. Negative results were found even for the TMT-A, which mainly taps into visuo-spatial attentional processes supported by right frontal and parietal areas [53]. Therefore, the left-lateralized findings observed in this study may be explained either by the prominent role of the left hemisphere in PS performance or by the fact that all of the tests found to be associated with the left fronto-parietal network require processing of verbal information, mainly elaborated by areas in the left hemisphere.

Cruz-Gomez et al. [21] have found alterations not only in the left but also in the right fronto-parietal network, as well as in the salience network, in patients with MS and cognitive impairment. In our study the salience network and, to a lesser extent, the DMN have emerged to be associated with performance on different PS-dependent tests. In particular, TMT-A scores, in the RRMS group, and Stroop speed index scores, in the SPMS group, positively correlated with the strength of functional coupling of the salience network in fronto-limbic and cerebellar areas involved in different executive processes [54,55]. Hence, a speculative interpretation of these findings may be that lower functional connectivity of the salience network in areas known as part of the DMN as well as of the right fronto-parietal network might be associated with faster PS performance. Indeed, these networks are negatively correlated with one another [56] and the integrity of the salience network has been observed to be a predictor of DMN functionality after traumatic brain injury [57]. Therefore, the salience network may play a crucial role in enabling efficient deployment of attentional resources that allow effective information processing and faster responses to stimuli.

The investigation of the DMN showed that only performance of patients with RRMS on the Semantic Fluency test was negatively correlated with the strength of functional connectivity of this network in the right cerebellum. This result might suggest that higher PS abilities are

associated with weaker representation of cerebellar areas consistently observed to contribute to execution of this test [58,59]. However, none of the other PS measures correlated with resting-state activity of this network, in line with findings by Janssen et al. [60] but contrarily to what reported by other studies on MS [15,20].

It follows that dysfunction due to MS pathology in different brain networks may contribute to the emergence of cognitive impairments. In fact, it cannot be ignored that the DMN, the salience and the fronto-parietal networks are functionally associated with one another [31] and fast cognitive processing may be affected by between- rather than within-network disconnections, e.g. decoupling between the DMN and the salience network. Indeed, Gamboa et al. [61] found that lack of communication between functionally specialized brain modules in MS may affect fast cognitive operations that depend on integration of different types of information across brain areas and hemispheres. Therefore, understanding the relevance of balance between functional segregation and integration across brain networks seems to be a relevant issue for further investigating PS function decline in MS.

Additionally, bidirectional correlations emerged between completion time on the TMT-A and connectivity of the sensorimotor network, possibly due to the importance of eye-hand coordination for the execution of this test. More specifically, higher completion times were predicted by higher connectivity in areas of the network and by lower connectivity in the right inferior frontal gyrus. These findings seem to suggest that functional segregation of the sensorimotor network and lack of integration with executive networks may impact motor execution as well as psycho-motor PS function. In the SPMS group no correlations between PS measures and the two control networks emerged, possibly suggesting that tests based on processing of verbal material might be more sensitive to neural disruption in later stages of MS.

The variability observed in the patterns of associations between PS-dependent measures and functional networks appears to rely on the diversity of cognitive functions underlying the PS measures collected. This means that different cognitive tests may not be interchangeable in the assessment of the same function and may depend on different neural correlates. Another caveat may be in the fact that networks' resting-state activity probably enables only partial characterization of the status of a cognitive function that is essentially dynamic, such as quickly performing cognitive processes [62]. Instead, incongruences in the results across MS phenotypes are only partially interpretable due to linguistic and cultural difference that may have an impact on test performance (e.g. on the Fluency tasks), as well as any difference in MRI parameters. The discrepancy in the sample size of the two groups might also play a role, although the more limited results for the SPMS group may be a consequence of the general worse health status of this patient group compared to patients with RRMS. In fact, it may be argued that accumulation of structural damage is the main cause driving cognitive symptoms [63] and functional alterations in MS [64]. Therefore, reorganization of functional brain architecture may occur in the early stages of the disease and contribute to support cognitive performance [65]. The increasing severity of brain insults may lead to the depletion of these adaptations and produce a drop in variance both in cognitive performance and in functional connectivity that may, in turn, prevent the identification of associations between cognitive and neural variables. However, future longitudinal studies including the assessment of both cognitive and neuroimaging changes over time are needed to clarify this point.

Some limitations of our study must be mentioned, since they may influence the interpretation of the differences observed in the results across MS phenotypes. First, the two cohorts of patients were recruited in two different countries, thus introducing cultural differences across the two groups. Second, the design of this study is cross-sectional, making any interpretation

on the possible evolution of cognitive and neural changes due to MS only speculative. Third, it cannot be ignored the fact that the limited sample size, especially of the SPMS group, might have negatively affected the statistical power of the study and the detection of significant associations between cognitive and neural features. Fourth, fMRI data of MS patients were not compared to healthy controls, therefore no characterization of functional connectivity alterations was possible. Finally, differences in MRI acquisition parameters allowed only qualitative comparisons between the two cohorts of patients, thus limiting the interpretations of the results.

5. Conclusions

The functional neural correlates of PS performance of patients affected by MS in different disease stages appear to be mainly consisting of different frontal networks involved in a range of executive functions. In particular, weaker strength of functional connectivity in the salience and the left fronto-parietal networks and other frontal and parieto-limbic areas is likely to affect fast information processing. Therefore, dysfunction caused by MS lesions across multiple networks may be particularly relevant for the emergence of PS deficits. Indeed, studies have shown cognitive impairments due to MS are likely to be dependent on scattered disruptions in communications between brain areas beyond the most commonly studied DMN [66-68]. Further investigations combining neuroimaging with tests that require fast integration of information across brain areas may clarify the neural correlates of cognitive symptoms experienced by people affected by MS, but also help identifying possible MRI markers for clinical trials to test the mechanisms of action and effectiveness of treatments.

Disclosures

The authors have no disclosures regarding financial benefits related to this study.

Data availability

The authors have no permission from participants to share their research data with others not members of the research team and their collaborators.

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Figure captions

Figure 1 Negative (blue) and positive (red) correlations between functional connectivity of: the left fronto-parietal network and A) the Stroop speed index, B) the Phonemic Fluency test, C) the Semantic fluency test; the default mode network and Semantic Fluency test; the salience network and the Trail Making Test - part A in the RRMS group ($p < .05$ FWE)

Figure 2 Correlations (negative in blue and positive in red) between the Stroop speed index and functional connectivity of the left fronto-parietal and salience networks in the SPMS group ($p < .05$ FWE)

Figure 3 Negative (blue) and positive (red) correlations between TMT-A scores and functional connectivity of the sensorimotor network in the RRMS group ($p < .05$ FWE)

Table 1 Inclusion criteria for the two studies.

RRMS	SPMS
Stable disease status for at least three months prior to recruitment	No relapses for at least three months prior to recruitment
Stable treatment for at least three months prior to recruitment	Stable treatment for at least three months prior to recruitment
Self-reported cognitive symptoms	Self-reported cognitive symptoms
MMSE ≥ 24	CPM ≥ 17
Objective cognitive impairment defined as a score of 2 standard deviations below normative values in at least one of the tests included in the neuropsychological battery	Objective cognitive impairment defined as a score of 2 standard deviations below normative values in at least one of the tests included in the neuropsychological battery
Absence of other neurological or psychiatric comorbidities	Absence of other neurological or psychiatric comorbidities
Age between 25 and 65	
EDSS ≤ 6	

CPM: Colored Progressive Matrices, EDSS: Expanded Disability Status Scale, MMSE: Mini Mental State Examination, RRMS: Relapsing-Remitting Multiple Sclerosis, SPMS: Secondary Progressive Multiple Sclerosis

Table 2 Scanning protocol for the two cohorts of patients.

MRI characteristic	RRMS	SPMS
Scanner	Ingenia, Philips Healthcare	Achieva, Philips Healthcare
Magnetic field	3T	1.5T
Structural imaging	• Sagittal T1-weighted magnetization prepared rapid acquisition gradient-echo (repetition time = 8.1 ms; echo time = 3.7 ms; inversion time = 1000 ms; slices = 170; voxel dimension = 1 mm × 1 mm × 0.94 mm); • Sagittal Fluid-Attenuated Inversion Recovery (repetition time = 4800 ms; echo time = 289 ms; slices = 326; voxel dimension = 1.1 mm × 1.1 mm × 0.56 mm)	• Sagittal T1-weighted (repetition time = 7.4 ms; echo time = 3.4 ms; no inversion time; slices = 280; voxel dimension = 1.1 mm × 1.1 mm × 0.6 mm); • Coronal Fluid-Attenuated Inversion Recovery (repetition time= 8000 ms; echo time = 125 ms; slices = 20; voxel dimension = 0.75 mm x 0.75 mm x 4.5 mm
Resting-state imaging	• Axial T2*-weighted Echo Planar Imaging (repetition time = 2600ms; echo time = 35ms; slices = 35; thickness = 4 mm; no gap; volumes = 200; matrix size = 96 x 94; field of view = 230 x 230 mm2)	• Axial T2*-weighted Echo Planar Imaging (repetition time = 2000 ms; echo time = 50 ms, slices = 20; thickness = 6 mm; no gap; volumes = 240; matrix size = 72 x 71; field of view = 230 x 230 mm2)

RRMS: Relapsing-Remitting Multiple Sclerosis, SPMS: Secondary Progressive Multiple Sclerosis

Table 3 Clinical characteristics and count of patients with deficits in each cognitive test.

Variable	RRMS (n=40)	SPMS (n = 25)	Test statistic	p
Clinical[a]				
Age (years)[b]	44.6 (8.8)	53.0 (12.0)	-3.25	0.002
Education (years)	14.0 (2.7)	10.0 (2.6)	183.00	< 0.001
Duration (years)	9.7 (7.2)	15.5 (7.6)	746.00	0.001
EDSS	3.4 (1.6)	6.5 (1.2)	932.50	< 0.001
Total intracranial volume (ml)[b]	1503.2 (184.9)	1669.6 (169.8)	-3.64	0.001
Gray matter volume (ml)[b]	637.63 (82.69)	569.95 (57.73)	3.81	0.001
White matter volume (ml)	423.25 (132.46)	418.33 (65.41)	363.00	0.065
Cerebrospinal fluid volume (ml)	303.17 (129.80)	681.27 (131.22)	967.00	< 0.001
Total lesion volume (ml)	10.6 (13.4)	26.2 (18.4)	789.00	< 0.001
Cognitive[c]				
TMT-A	6 (15%)	15 (60%)	14.24	< 0.001
Stroop speed index	5 (12.5%)	7 (28%)	2.45	0.117
Phonemic Fluency	1 (2.5%)	5 (20%)	5.62	0.018
Semantic Fluency	1 (2.5%)	5 (20%)	5.62	0.018

[a] Mean (standard deviation); Mann-Whitney U test

[b] Independent sample t-test

[c] Total count (percentage); χ^2 test

EDSS: Expanded Disability Status Scale, RRMS: Relapsing-Remitting Multiple Sclerosis, SPMS: Secondary Progressive Multiple Sclerosis, TMT: Trail Making Test

Table 4 Correlations between performance on PS-dependent tests and functional connectivity of the left fronto-parietal network, the default mode network and the salience network in the RRMS group ($p < .05$ FWE)

Cognitive variable	Cluster extent	r	Side	Brain region	*t* value	MNI coordinates		
						x	*y*	*z*
Left fronto-parietal network								
Stroop speed index[a]	73	-.628	R	PCC (BA 31)	5.26	28	-44	30
			R	PCC (BA 31)	4.05	28	-52	28
Phonemic Fluency[b]	115	.605	R	SFG (BA 8)	4.86	4	32	56
			R	SFG (BA 8)	4.10	2	36	46
Semantic Fluency[b]	100	.481	R	PCG (BA 4)	5.06	60	-6	22
			R	PCG (BA 6)	4.14	54	-8	34
			R	PCG (BA 6)	3.73	46	-8	32
Default mode network								
Semantic Fluency[a]	69	-.541	R	Nodule	5.82	8	-64	-32
			R	Uvula	4.21	0	-64	-36
			R	Declive	4.08	20	-60	-28
Salience network								
TMT-A[b]	91	.745	L	PCC (BA 30)	5.07	-16	-68	4
			L	Cuneus (BA 18)	4.48	-12	-72	16
			L	LG (BA 18)	4.32	-8	-72	4

102	.678	R	Declive	4.97	-14	-78	-26
		R	Declive	4.69	-22	-76	-28
		R	Declive	4.07	-30	-78	-30
140	.714	R	IFG (BA 44)	4.84	46	10	20
		R	IFG (BA 44)	4.26	52	4	12
		R	IFG (BA 45)	4.20	46	18	18

[a] Negative correlation

[b] Positive correlation

BA: Brodmann area, IFG: inferior frontal gyrus, LG: lingual gyrus, PCC: posterior cingulate cortex, PCG: precentral gyrus, SFG: superior frontal gyrus

Table 5 Correlations between performance on the Stroop speed index and functional connectivity of the left fronto-parietal and salience networks in the SPMS group ($p < .05$ FWE)

Cognitive variable	Cluster extent	r	Side	Brain region	*t* value	MNI coordinates		
						x	*y*	*z*
				Left fronto-parietal network[a]				
Stroop speed index	90	-.810	L	IFG (BA 47)	6.31	-36	22	-12
			L	IFG (BA 47)	5.09	-36	22	-2
				Salience network[b]				
	108	.784	L	ACC (BA 33)	4.95	-8	8	30
				ACC (BA 24)	4.82	-4	12	30

[a] Negative correlation

[b] Positive correlation

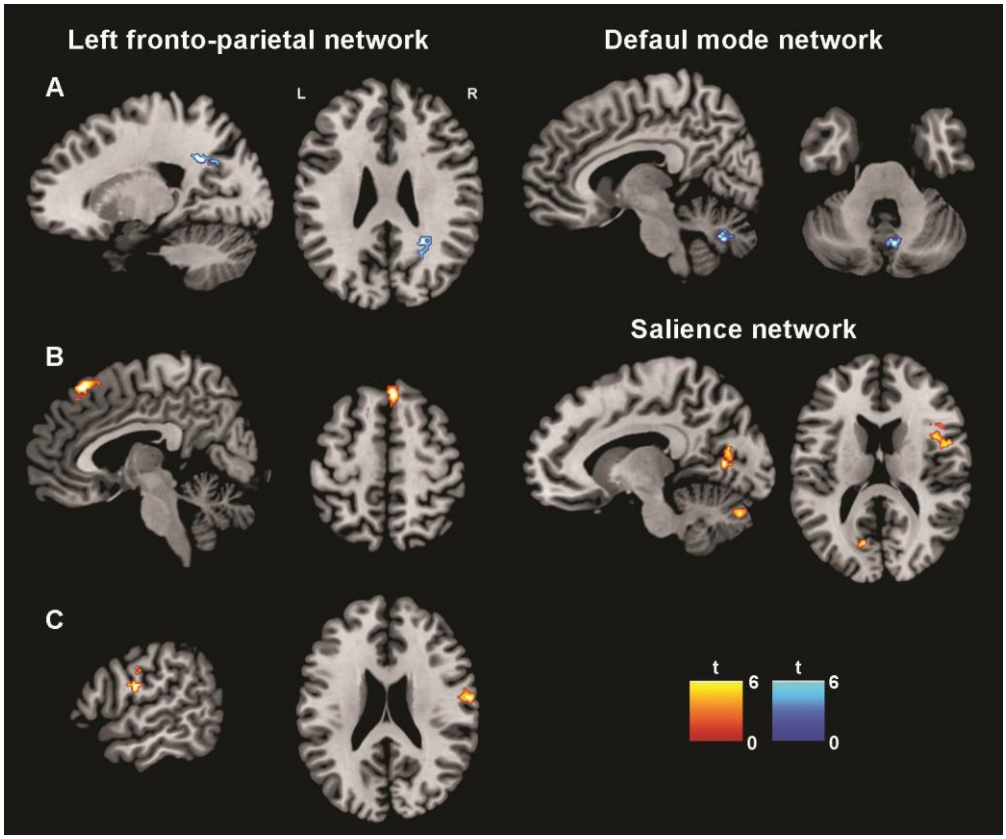
ACC: posterior cingulate cortex, BA: Brodmann area, IFG: inferior frontal gyrus

Table 6 Correlation between TMT-A scores and functional connectivity of the sensorimotor network in the RRMS group ($p < .05$ FWE)

Cognitive variable	Cluster extent	r	Side	Brain region	*t* value	MNI coordinates		
						x	*y*	*z*
				Positive correlation				
TMT-A	398	.676	R	PCG (BA 6)	5.80	48	-16	26
			R	PCG (BA 43)	4.97	60	-10	10
			R	PCG (BA 4)	4.94	64	-10	28
	315	.604	L	PCG (BA 43)	5.19	-54	-6	14
			L	Insula (BA 13)	5.05	-44	-12	26
			L	PCG (BA 4)	4.22	-60	-4	28
				Negative correlation				
	73	-.687	R	IFG (BA 44)	5.25	48	6	16
			R	IFG (BA 44)	4.28	54	12	18
			R	IFG (BA 9)	4.13	46	14	24

BA: Brodmann area, IFG: inferior frontal gyrus, PCG: Precentral gyrus, TMT: Trail Making Test

Figure 1


Left fronto-parietal network
Defaul mode network
A
L
R
B
Salience network
C
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0
t
6
0

Figure 2

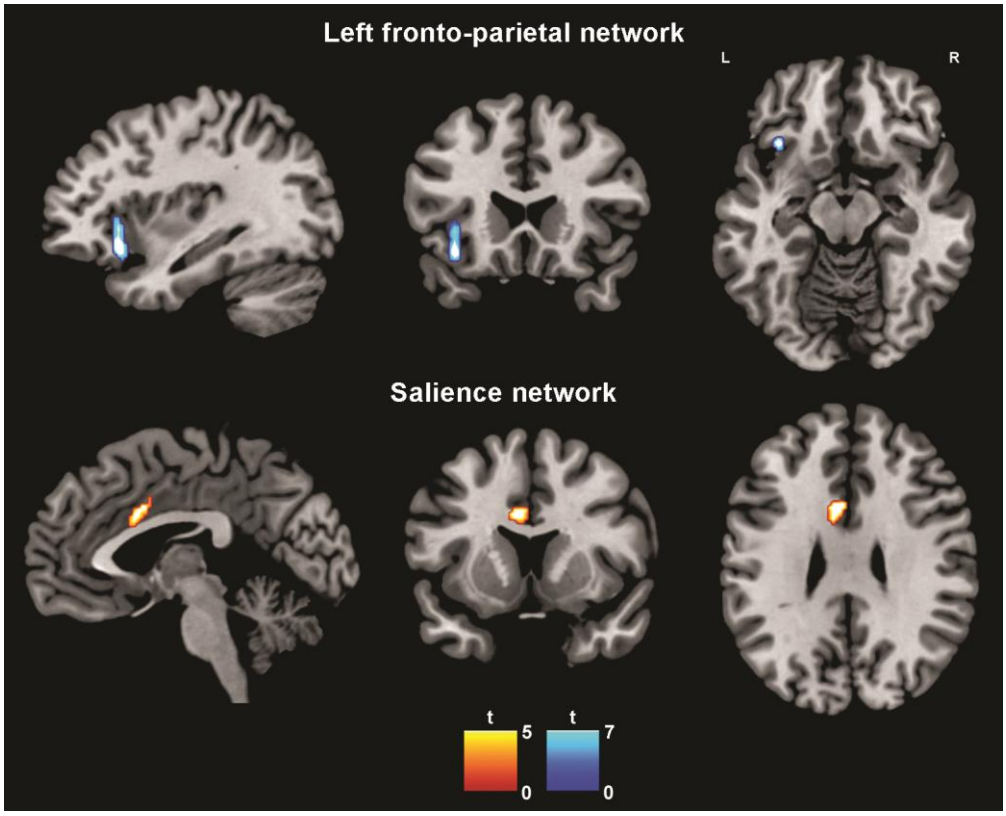

Figure 3