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Predicting clinical progression and cognitive decline in patients with relapsing-remitting multiple sclerosis: a 6-year follow-up study

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ABSTRACT

Introduction. Cognitive impairment occurs from the earliest stages of multiple sclerosis (MS) and progresses over time. The introduction of disease modifying therapies (DMTs) has changed the prognosis for MS patients, offering a potential opportunity for improvement in the cognitive arena as well.

Material and methods. 41 patients with relapsing-remitting multiple sclerosis (MS) were recruited to the study. Thirty patients were available for final follow-up and were included in the analysis. Baseline (BL) brain MRI including volumetry and neuropsychological tests were performed. Blood samples were collected at BL and follow-up (FU) and were tested for: vascular endothelial growth factor (VEGF), soluble vascular cell adhesion molecule-1 (sVCAM1), soluble platelet-endothelial CAM-1 (sPECAM1), and soluble intercellular CAM-1 (sICAM-1). Patients were invited for a final neuropsychological follow-up after a median of 6 years. Disease activity (relapses, EDSS increase, new/active brain lesions on MRI) was analysed between BL and FU.

Results. The study group deteriorated in the Rey–Osterrieth Complex Figure (ROCF) test ($p = 0.001$), but improved significantly in three other tests, i.e. semantic fluency test ($p = 0.013$), California Verbal Learning Test (CVLT, $p = 0.016$), and Word Comprehension Test (WCT, $p < 0.001$). EDSS increase correlated negatively with semantic fluency and WCT scores ($r = –0.579$, $p = 0.001$ and $r = –0.391$, $p = 0.033$, respectively). Improvements in semantic fluency test and WCT correlated positively with baseline deep grey matter, grey matter, and cortical volumes ($p < 0.05$, $r > 0$).
Higher EDSS on FU correlated significantly negatively with baseline left and right pallidum, right caudate, right putamen, right accumbens, and cortical volume ($p < 0.05$, $r < 0$). No significant relationship was found between the number of relapses and EDSS on FU or neuropsychological deteriorations. Improvements in WCT and CVLT correlated positively with baseline sPECAM1 and sVCAM1 results, respectively ($r > 0$, $p < 0.05$). Deterioration in ROCF test correlated significantly with higher levels of baseline VEGF and sVCAM1 ($p < 0.05$).

Conclusions. Brain volume is an important predictor of future EDSS and cognitive functions outcome. MS patients have a potential for improving in neuropsychological tests over time. It remains to be established whether this is related to successful disease modification with immunotherapy. Baseline volumetric measures are stronger predictors of cognitive performance than relapse activity, which yet again highlights the importance of atrophy in MS prognosis.

Keywords: multiple sclerosis, cognitive functions, brain atrophy, CVLT, predictors, ROCF, semantic fluency, choroid plexus, EDSS

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Introduction

Multiple sclerosis (MS) is a chronic demyelinating disorder of both inflammatory and neurodegenerative aetiologies [1]. Cognitive impairment (CI) occurs in MS patients from the earliest stages of the disease, progresses over time, and affects up to 65% of the MS population [2].

Patients with MS perform worse in cognitive tests compared to the healthy population [3]. Cognitive domains that are typically affected in MS include information processing speed, learning and memory, visuospatial skills, and executive function [2]. Importantly, cognitive complaints observed in patients during the prodromal stage of MS have been shown to be predictors of a worse clinical outcome [4].

The introduction of effective disease-modifying therapies (DMTs) has markedly reduced the annual relapse rate and radiological progression. Moreover, DMTs also seem to have an impact on reducing the rate of cognitive impairment (CI) and the potential for improving cognitive parameters [5].

The risk factors associated with the development of CI in MS include older age, greater disability in EDSS scale [6], brain atrophy [7], fewer years of education, and male sex [8], although the data differs despite the studies. Factors that have a protective effect on the development of CI are DMTs [5] and the cognitive reserve [9]. There is little data on biochemical factors as predictors of CI in MS, studies having focused mainly on light neurofilaments, with contradictory results [10, 11].

Clinical rationale for study

Conclusions regarding prognostic factors in MS differ between studies. Our objective was to analyse them in our patient population, with a longer follow-up period than that adopted in most studies.

In addition to the well-known prognostic factors, we also investigated those rarely considered, e.g. neurotrophins and endothelial molecules.

Our study aimed to analyse how performance in specific cognitive domains evolves as the disease worsens in a typical relapsing-remitting MS (RRMS) population. Additionally, we analysed which baseline biochemical and imaging parameters were the best predictors of Expanded Disability Status Scale (EDSS) worsening, and which were predictive of cognitive deterioration, or protective of cognitive function, in specific neuropsychological domains.

Material and methods

Participants were recruited from a single MS centre at the Department of Neurology, Poznan University of Medical Sciences, Poland. Forty-one adult patients with RRMS were recruited for the study consecutively between 2014 and 2015. Baseline (BL) brain MRI, including volumetry, was performed on a 3.0 Tesla scanner at the Centre for Modern Interdisciplinary Technologies, Nicolaus Copernicus University in Torun, Poland. Extensive cognitive assessment was done by a trained neuropsychologist, and blood samples collected and stored until further analysis. After baseline assessment, out of 41 originally recruited patients, nine were lost to follow-up at some point and two declined to participate, leaving 30 patients who were recruited for the final follow-up including neuropsychological re-assessment, conducted between 2020 and 2022.

On the day of of study inclusion, all patients had been treated with platform disease-modifying therapies for a median of 3 years (range 0.5–6, IQR 3) and were regularly being seen at the outpatient MS clinic of the Department of Neurology, Poznan University of Medical Sciences. EDSS scores were recorded by treating neurologists at the time of enrollment into the study, and every 3–6 months thereafter during regular follow-up visits to the MS centre. A control brain MRI was performed yearly on a 3.0 T scanner available at the local hospital, recording the presence of new or enlarging T2 or gadolinium enhancing lesions.

Clinical data concerning the onset of first MS symptoms and of MS diagnosis was obtained from patient records and verified, with the data being entered by treating neurologists into the central nationwide register under the public healthcare system (the National Health Fund electronic database).

The median follow-up (FU) of the study cohort who showed up for the final cognitive follow-up (n = 30) was 6 years. Patients who did not consent to participate in the study, and those with suspected depressive disorders (as verified by the Beck's Depression Inventory) were excluded from the study.

This study was approved by the Internal Review Board of Poznan University of Medical Sciences, Poland and all participants signed written informed consent.

Neuroimaging data acquisition and analysis

Brain structural imaging was performed using high resolution T1-weighted sequence obtained using 3 Tesla MRI scanner (MR750, GE Healthcare; Waukesha, WI, USA) and 32-channel dedicated head coil. Protocol included high-resolution T1-weighted BRAVO acquisition, CUBE T2-FLAIR, and multidirection T2-weighted diffusion acquisition. Extracted dicom images were converted to nifti format and processed using comprehensive segmentation pipeline recon-all using Freesurfer 5.3 to extract brain volume and evaluate cross-sectional regional brain volume difference across the entire group. Freesurfer (Martinos Centre for Biomedical Imaging, Harvard-MIT, Boston; available at http://surfer.nmr.mgh.harvard.edu/) consists of surface based analysis and volumetric segmentation. It involves intensity non-uniformity correction affine transformation to a MNI305 template,

intensity normalisation, removal of non-brain tissue, linear and non-linear transformations to a probabilistic brain atlas, and labelling of cortical and subcortical structures. It uses a Markov Random Field model for each structure for each point in space. Spatial localisation priors are used in determining the right label per voxel. Since Freesurfer version 5.2, surface-based calculations are used to calculate various brain volumes to get better accuracy. In our study, segmentation was performed using default settings (i.e. using the command: 'recon-all'). For our study, we used the compartment measurements reported by Freesurfer. All volumes were extracted from the stat files that Freesurfer produces using the 'asegstats2table' command. Since Freesurfer estimates ICV and does not perform segmentation of extracerebral CSF, we obtained the CSF volume by subtracting TBV from the estimated ICV.

Detailed and technically acceptable baseline MRI data was available for 27/30 patients who showed up for the final cognitive follow-up. Yearly follow-up MRI was performed on a 3.0 T scanner available at the local hospital (Magnetom Skyra 3.0 T, Siemens Healthineers, 20-channel dedicated head coil).

Neuropsychological assessment

A broad battery of neuropsychological tests was applied by a trained neuropsychologist in standardised clinical conditions. The following neuropsychological tests were used: Stroop Colour-Word Interference Test, Rey–Osterrieth Complex Figure test, Benton Visual Retention Test, Colour Trails Test, semantic and phonemic fluency test, Word Comprehension Test, Raven's Progressive Matrices, and California Verbal Learning Test. (see Tab. 1 for summary of tests with assessed domains).

Blood biomarkers

The following molecules were measured with the use of the ELISA method in serum samples collected at baseline, according to the manufacturer's instructions: vascular endothelial growth factor (VEGF), soluble vascular cell adhesion molecule-1 (sVCAM1), soluble platelet-endothelial cell adhesion molecule-1 (sPECAM1), and soluble intercellular adhesion molecule-1 (sICAM-1) (BioVendor Laboratory Medicine Inc., Czech Republic). The levels of sVCAM1, sPECAM1, and sICAM1 were calculated from standard curves in nanograms per millilitre (ng/mL). Nerve growth factor (NGF), brain derived neurotrophic factor (BDNF), and neutrophin 4/5 (NT4/5) were measured with the use of ELISA kits, according to the manufacturer's instructions (Multi-Neurotrophin Rapid Screening ELISA Kit: Human, Biosensis Pty Ltd, Thebarton, Australia) in lysates of peripheral blood mononuclear cells fraction, which was isolated at baseline and then frozen until further analysis, according to the procedure described previously [12]. Their concentrations were expressed as relevant weight units per 1 mg of the protein.

Statistical analyses

The results were reported as counts (percentage) for the qualitative variables, mean ± standard deviation if normally distributed, and median (interquartile range) if not normally distributed quantitative variables. Quantitative variables were summarised using either the mean and standard deviation, or median and IQR. Qualitative variables were summarised using percentages.

A comparison of the two repeated measurements of quantitative variables was performed using the Wilcoxon signed-rank test. Correlations among continuous variables were assessed by the Spearman rank correlation coefficient. Statistical significance was defined as p-value < 0.05. Therefore, those test results with $p < 0.05$ were interpreted as indicating significant relationships. The analysis was performed in R, version 4.3.1. (R Core Team (2023). (R: A language and environment for statistical computing, R Foundation for Statistical Computing, Vienna, Austria, URL https://www.R-project.org).

Results

In the final analysis, we included 30 subjects with RRMS who completed the long-term cognitive follow-up, of whom 27 were women (90%) and three men (10%). The mean age at MS diagnosis (years) was 31 ± 7; median was 30 (range 18–45, IQR 10). The mean age at baseline (years) was 36 ± 7; median 35 (range 23–50, IQR 11), and at the final follow-up it was 42 ± 6.8; median 41 (range 29–57, IQR 11.25).

The mean disease duration in years from experiencing first MS symptoms to baseline was 6 ± 4, median 6 (range 1–26, IQR 3). At baseline, median EDSS was 1.0 (range 0–3.5), while at the final follow-up, median EDSS was 2.0 (range 0–6, IQR 2). Only four (13.3%) patients achieved EDSS > 3 at FU. 23/30 (76.7%) patients had higher education.

At the start of the study, 29 patients were on moderate efficacy treatment (injectables such as beta-interferons and glatiramer acetate), and one patient was receiving high efficacy therapy (fingolimod). Between the baseline and follow-up visits, nine (30%) patients switched their treatments because of a lack of efficacy and 12 (40%) due to side effects and inconvenience related to long-term injections. Fourteen (46.7%) patients experienced at least one relapse between BL and FU. Annual brain MRIs revealed new active lesions in eight (26.7%) patients and new T2 lesions in 15 (30%) patients.

By the end of the study period, 26 patients remained on moderate efficacy treatment (dimethyl fumarate, beta-interferons, teriflunomide, glatiramer acetate), three patients were on high efficacy drugs (ofatumumab, cladribine), and one patient had resigned from treatment.

Table 1. Comparison of neuropsychological test results measured at baseline and after mean 6-year follow-up

Neuropsychological test	Domain	Timepoint	Mean	SD	P-value
Semantic fluency test (SF)	Verbal functions	Baseline	22.53	5.37	0.013*
		Follow-up	24.93	6.21	
Phonemic fluency test (PF)	Verbal functions	Baseline	17.13	6.11	0.124
		Follow-up	18.93	5.67	
Raven's Progressive Matrices	Abstract reasoning	Baseline	51.67	15.76	0.179
		Follow-up	48.53	9.06	
Word Comprehension Test (WCT)	Verbal functions	Baseline	21.27	5.15	< 0.001*
		Follow-up	26.27	5.41	
Stroop Interference Test (time 1: colour)	Working memory Executive functions	Baseline	81.53	31.22	0.516
		Follow-up	75.73	14.67	
Stroop Interference Test (time 2: interference)	Working memory Executive functions	Baseline	119.80	23.21	0.509
		Follow-up	116.70	26.94	
Benton Visual Retention Test (correct score)	Spatial and visuospatial memory	Baseline	7.87	1.50	0.234
		Follow-up	7.57	1.63	
Benton Visual Retention Test (error score)	Spatial and visuospatial memory	Baseline	3.13	2.39	0.073
		Follow-up	3.90	3.14	
Colour Trails Test (time 1)	Attention and executive functions	Baseline	38.30	14.95	0.574
		Follow-up	39.40	18.86	
Colour Trails Test (time 2)	Attention and executive functions	Baseline	73.17	25.68	0.65
		Follow-up	74.20	28.99	
Rey–Osterrieth Complex Figure (ROCF) test (copy)	Spatial and visuospatial memory	Baseline	36.00	0.00	0.001*
		Follow-up	34.77	1.65	
Rey–Osterrieth Complex Figure (ROCF) test (recall)	Spatial and visuospatial memory	Baseline	23.10	5.73	1
		Follow-up	23.13	6.71	
California Verbal Learning Test (CVLT)	Verbal memory	Baseline	58.90	8.34	0.016*
		Follow-up	62.47	7.31	

Grey background — tests with significant change between two; $p < 0.05$ — clinically significant; SD — standard deviation; *$p < 0.05$ clinically significant

Neuropsychological assessment

At the group level, we found significant changes in neuropsychological scores from baseline to the final follow-up in four tests (Tab. 1).

Patients deteriorated significantly in the Rey–Osterrieth Complex Figure test (from mean 36.00 points to 34.77, $p = 0.001$).

On the other hand, patients improved significantly in the semantic fluency test (from mean 22.53 to 24.93 points, $p = 0.013$), California Verbal Learning Test (from mean 58.90 to 62.47 points, $p = 0.016$), and Word Comprehension Test (from mean 21.27 to 26.27 points, $p < 0.001$).

There were no significant relationships between the number of relapses, new active or T2 lesions between BL and FU, and changes in specific neuropsychological tests. Patients with a higher increase in EDSS had a lower likelihood of reaching improvement in the semantic fluency test ($r = –0.579$, $p = 0.001$) and WCT ($r = –0.391$, $p = 0.033$). On follow-up, patients who achieved worse results on baseline CVLT and CTT tests deteriorated in EDSS significantly more often, and subsequent relapses were significantly more common in patients with worse baseline Benton and WCT test results ($p < 0.05$).

Imaging predictors of cognitive decline, disease activity, and disability progression

Improvement in the semantic fluency test correlated positively ($p < 0.05$, $r > 0$) with baseline volumes of right caudate and left hemisphere cortex (Tab. 2). Improvement in the Word Comprehension Test correlated positively ($p < 0.05$, $r > 0$) with baseline volumetric parameters of the right pallidum, right nucleus accumbens, left and right hemisphere cortex, and total grey matter (Tab. 2).

None of the volumetric parameters was a significant predictor for achieving improvement in CVLT or for deterioration in ROCF test.

The total number of relapses between BL and FU correlated significantly negatively with the left and right choroid plexus volumes ($r = –0.452$, $p = 0.02$ and $r = –0.402$, $p = 0.042$). Accumulation of a new T2 lesion between BL and FU correlated negatively with the volume of the left and right cerebellum white matter, left choroid plexus, and right thalamus ($p < 0.05$, $r < 0$) (Tab. 2).

Deterioration in EDSS on FU correlated significantly negatively with the following baseline volumes: left and right globus pallidus, right caudate, right putamen, right nucleus accumbens, left and right hemisphere cortex, and total grey

Table 2. Imaging predictors of cognitive decline, disease activity, and disability progression

	EDSS increases between BL and FU	Semantic fluency test FU	Word Comprehension Test FU	Relapses between BL and FU	New T2 lesions between BL and FU
R thalamus	r = –0.185, p = 0.356	r = 0.281, p = 0.155	r = 0.276, p = 0.163	r = –0.323, p = 0.107	r = –0.395, p = 0.046*
L thalamus	r = –0.249, p = 0.211	r = 0.121, p = 0.549	r = 0.229, p = 0.251	r = –0.176, p = 0.39	r = –0.231, p = 0.257
R choroid plexus	r = –0.124, p = 0.536	r = –0.068, p = 0.736	r = –0.091, p = 0.651	r = –0.402, p = 0.042*	r = –0.24, p = 0.238
L choroid plexus	r = –0.146, p = 0.467	r = 0.103, p = 0.61	r = –0.159, p = 0.428	r = –0.452, p = 0.02*	r = –0.517, p = 0.007*
R accumbens	r = –0.444, p = 0.02*	r = 0.366, p = 0.061	r = 0.488, p = 0.01*	r = –0.12, p = 0.559	r = –0.13, p = 0.527
L accumbens	r = –0.262, p = 0.186	r = 0.184, p = 0.359	r = 0.244, p = 0.219	r = –0.099, p = 0.629	r = 0.022, p = 0.915
R pallidum	r = –0.519, p = 0.006*	r = 0.353, p = 0.071	r = 0.453, p = 0.018*	r = –0.371, p = 0.062	r = –0.249, p = 0.219
L pallidum	r = –0.467, p = 0.014*	r = 0.339, p = 0.084	r = 0.337, p = 0.086	r = –0.295, p = 0.143	r = –0.091, p = 0.659
R caudate	r = –0.424, p = 0.027*	r = 0.429, p = 0.025*	r = 0.302, p = 0.126	r = –0.247, p = 0.224	r = –0.133, p = 0.518
L caudate	r = –0.265, p = 0.181	r = 0.207, p = 0.301	r = 0.107, p = 0.594	r = –0.308, p = 0.126	r = –0.084, p = 0.684
R putamen	r = –0.397, p = 0.04*	r = 0.124, p = 0.537	r = 0.319, p = 0.105	r = –0.361, p = 0.07	r = –0.055, p = 0.79
L putamen	r = –0.33, p = 0.093	r = 0.154, p = 0.442	r = 0.179, p = 0.373	r = –0.377, p = 0.058	r = –0.053, p = 0.796
Cortex	r = –0.583, p = 0.001*	r = 0.381, p = 0.05*	r = 0.416, p = 0.031*	r = –0.239, p = 0.239	r = –0.086, p = 0.678
Total grey matter	r = –0.517, p = 0.006*	r = 0.34, p = 0.083	r = 0.4, p = 0.039*	r = –0.305, p = 0.129	r = –0.139, p = 0.499
R hemisphere cortex	r = –0.579, p = 0.002*	r = 0.353, p = 0.071	r = 0.391, p = 0.044*	r = –0.246, p = 0.227	r = –0.036, p = 0.86
L hemisphere cortex	r = –0.61, p = 0.001*	r = 0.402, p = 0.038*	r = 0.425, p = 0.027*	r = –0.243, p = 0.232	r = –0.109, p = 0.597
R cerebellum WM	r = –0.16, p = 0.425	r = 0.231, p = 0.247	r = 0.027, p = 0.893	r = –0.208, p = 0.307	r = –0.421, p = 0.032*
L cerebellum WM	r = –0.272, p = 0.169	r = 0.259, p = 0.192	r = 0.08, p = 0.691	r = –0.153, p = 0.456	r = –0.491, p = 0.011*

BL — baseline; EDSS — Expanded Disability Scale score; FU — follow up; L — left; R — right; WM — white matter; *p < 0.05 statistically significant, r > 0 positive association, r < 0 negative association; yellow background — association with significant correlation

matter ($p < 0.05$, $r < 0$) (Tab. 2). We did not identify a correlation between the number of relapses and EDSS deterioration from baseline.

Blood markers as predictors of cognitive decline, disease activity, and disability progression

Improvement in the Word Comprehension Test correlated positively with baseline sPECAM1 results ($r = 0.402$, $p = 0.028$), while improvement in the CVLT correlated positively with baseline sVCAM1 results ($r = 0.401$, $p = 0.028$). On the other hand, deterioration in the ROCF test correlated significantly with higher levels of VEGF ($r = –0.395$, $p = 0.031$) and sVCAM1 ($r = –0.392$, $p = 0.032$). There was no correlation between baseline blood biomarkers results and subsequent relapses, EDSS worsening, or new active or T2 lesions on MRI.

Discussion

Our study provides long-term detailed cognitive follow-up data for a subgroup of classic RRMS patients from a Polish population treated with the use of moderate efficacy DMTs from disease onset. We aimed to determine baseline MRI and biochemical predictors of future disease activity, disability progression, and cognitive decline.

Firstly, our patient group was representative of the general RRMS population treated at our centre, as we recruited patients consecutively. Notably, our study group had a moderately active MS course. Namely, within the six years of follow-up, only 30% had to switch their primary DMT due to a lack of efficacy, less than 50% had relapses, and 30% had new T2 lesions in MRI.

As for the cognitive aspect, on the group level the study subjects worsened from baseline to follow-up only in the ROCF copy test, while they improved significantly in three other tests, two of them assessing verbal functions (semantic fluency test and Word Comprehension Test), which we find informative, as it is believed that language functions are generally spared in MS [2]. Importantly, the standard test batteries used in the MS population do not include the assessment of this domain. However, verbal fluency is perceived to be as sensitive as the widely used SDMT [13].

The third test where patients improved over time was the California Verbal Learning Test, which is used for evaluating verbal learning and memory [3]. These results were consistent with other studies where patients on DMTs have shown improvement in CVLT-II [14–17].

The only test where patients deteriorated on FU was the ROCF test, which assesses visuospatial abilities, memory, attention, and executive functions [18]. Patients with MS generally perform worse in the ROCF test than healthy controls, and the results are correlated with grey matter and deep

grey matter volume [18]. To date, no other studies concerning longitudinal ROCF scores in the MS population have been published. Our data indicates that a nonverbal test such as the Ray-Osterrieth Complex Figure might be useful in longitudinal assessment of complex planning and memorisation independent of language. In standard conditions, ROCF performance is characterised by a ceiling effect. However, in clinical settings of chronic disease, minor deficiencies in copying reveal subtle cognitive impairment which might be useful for the international community to replicate.

In our study group, improvement in language tests (SF and WCT) correlated positively with baseline volumes of deep grey matter structures (right caudate nucleus in FS, right globus pallidus, and right accumbens in WCT) and cortex. Additionally, for WCT there was also a significant positive correlation between baseline total grey matter volume and better results on FU. This is in line with previous studies, where cortical thickness, grey matter and deep grey matter atrophy have been associated with cognitive impairment [7, 19, 20]. On the other hand, grey matter volume has increased in MS patients who have learnt a second language, which is suggestive of the efficacy of cognitive rehabilitation [21]. Matias-Guiu et al. [22] reported that semantic fluency results correlated with thalamus and caudate volumes in both hemispheres, while Crosson et al. [23] revealed that both left and right basal ganglia play a role in language generation. Total cortical volume might be perceived as a surrogate biomarker of cognitive reserve, and could in this way explain the fluctuations of the change over time.

Choroid plexus (CP) volume is yet another important imaging marker that has recently surfaced in MS research. CP volume has been found to be larger in patients with RRMS than in neuromyelitis optica spectrum disorder (NMOsd) or in healthy controls [24–26]. In our study, baseline CP volume correlated significantly negatively with relapses and new T2 lesions during the follow-up period, which is a novel finding.

Most of the previous studies focused on the positive correlation between choroid plexus and brain atrophy [24, 27]. Also, a larger CP was found in patients with cognitive impairment [24] and was associated with an increased risk of chronic lesions expansion [27]. CP might be a better indicator of disability than relapses, because of its close association with brain atrophy; to date, data concerning CP and relapses is divergent. Jankowska et al. [28] reported that a larger plexus correlated with relapses during 12-month FU, but not with the change in brain volume or lesions volume, albeit in a small sample of 14 subjects. Similarily, Ricigliano et al. [25] connected CP volume with relapses. However, Muller et al. [26] found no association between CP and relapses, and Klistorner et al. [27] found no correlation between choroid plexus and new T2 lesions or EDSS deterioration during a 4-year FU.

As for predicting EDSS score on follow-up, we found several correlations with the baseline brain volume parameters. Deterioration in EDSS correlated significantly with cortical, total grey matter and deep grey matter (left and right pallidum, right caudate, right putamen, right accumbens) volumes, which confirms the role of these parameters as good predictors of future disability and is consistent with the literature [19, 29–34].

Despite the fact that almost half of the patients experienced at least one relapse between BL and FU, they were not relevant predictors of EDSS or cognitive outcomes. This is in line with the published data showing that only early relapses have an impact on future disability, while relapses throughout the subsequent disease course are not associated with long-term disability measures [35, 36].

On the contrary, baseline volumetric parameters had a significant impact on both EDSS and cognitive performance, which confirms that brain atrophy might be more relevant in predicting future disability. Therefore, atrophy assessment should become a standard part of MRI protocol in MS [37].

Interestingly, higher baseline sPECAM-1 and sVCAM-1 serum concentrations correlated significantly with improvements in the WCT and CVLT tests, respectively. The opposite results were obtained for the ROCF test, where baseline higher sVCAM1 and VEGF values correlated significantly positively with deterioration in the ROCF test. So far, data on the role of sPECAM -1, sVCAM-1 and VEGF as potential markers in MS has yielded contradictory results.

Both sPECAM -1 and sVCAM-1 are considered markers of MS activity and blood-brain barrier disruption [38]. sPECAM-1 plasma concentrations have been shown to be significantly increased in MS patients and to be elevated during relapses [39]. However, they may also have a neuroprotective effect: an sVCAM-1 increase was associated with a better treatment outcome [40], while lower sPECAM-1 and sVCAM-1 increased the risk of progression from CIS to MS [38]. Higher baseline endothelial molecules concentration may reflect greater inflammatory disease activity, and thus greater therapeutic potential, due to the fact that DMTs main mode of action is anti-inflammatory. However, in our study, this was confirmed only with regards to the language domain tests.

Our group has previously shown that patients with MS have a higher level of serum VEGF compared to non-MS individuals, and that similar concentrations were found in MS and NMOsd patients [41]. A different group showed that a higher VEGF level shortened the remission phase in MS [42], but its role remains ambiguous.

In the natural history of MS, we expect a constant deterioration of cognitive performance over time. Also in previous studies, patients' cognitive function was observed to deteriorate over time [43].

We did not consider the influence of the initial neuropsychological test results on the FU cognitive scores; it is known that the initial CI is a negative factor for further development of CI. However, we found that patients who achieved worse results on baseline CVLT and CTT tests significantly more often deteriorated in EDSS on FU, which is coherent with other studies [44].

However, one should consider that most of the previous data was derived from a different (historical) MS population, as current patients typically have a lower median EDSS and start DMT earlier, which is related to the new diagnostic criteria [45]. Recently, Katsari et al. found that during a 10-year follow up there was a group of patients that did improve cognitive functions [46].

The improvement that was observed in our study cohort could partly be explained by the relatively mild disease course (mean EDSS on FU = 2.0) and by a relatively high educational level in our group, as cognitive reserve has an important protective function [9]. Finally, all our patients were treated with DMTs.

We do recognise some limitations of our study

Firstly, the final sample size was small, and we did not use a control group. Fortunately, all of the neuropsychological tests we used are standardised and have established normative data. Additionally, there were disproportionately few men in our population, which may also influence the results, as male gender has been reported as a negative prognostic factor for the development of CI in MS.

Another limitation is that we did not have volumetric parameters from FU MRIs to determine brain atrophy rate. We also did not consider the potential impact of DMTs switching on the final result.

On the other hand, the strengths of the study include a relatively long observation period and the fact that patients were assessed with a wide battery of neuropsychological tests applied by a trained neuropsychologist. Also, a large number of radiological parameters and blood markers were included as potential predictors.

Clinical implications/future directions

We have shown that MS patients may improve in cognitive performance while on DMTs. We found that patients with higher baseline volumes of grey matter, deep grey matter and cortex improved in tests evaluating language functions. Baseline brain atrophy was a more significant factor influencing the EDSS and cognitive outcomes than relapse rate. Detecting cognitive deterioration in the course of a long chronic disease poses a difficult challenge. On the one hand, the repetitive assessment using psychometric tests designed for cross-sectional assessment might detect a spurious improvement in test performance that might be caused by learning the test heurystic.

Yet on the other hand, the clinician's drive to define actionable deterioration in cognitive performance is desired to modify the clinical management and adjust to the progressive nature of the disease process. There is still an unmet need to implement volumetric assessment into routine MRI protocol. The role of endothelial molecules in stratification of patients with regards to future disease progression requires further studies.

Article information

Data availability statement: *Anonymised data and documentation may be made available to qualified investigators upon reasonable request.*

Ethics statement: *This study was approved by the Internal Review Board of Poznan University of Medical Sciences, Poland and all participants signed an informed consent form.*

Authors' contributions: *Karolina Kania — drafting/revision of manuscript for content, including medical writing for content, major role in acquisition of data, analysis or interpretation of data; Mikołaj A. Pawlak — drafting/revision of manuscript for content, including medical writing for content, major role in acquisition of radiographic data, analysis or interpretation of data, study concept; Maria Forycka — drafting/revision of manuscript for content, major role in acquisition of neuropsychological data, analysis/interpretation of data; Monika Wiłkość-Dębczyńska — major role in acquisition of neuropsychological data, analysis/interpretation of data; Sławomir Michalak — analysis/interpretation of data, study concept; Agnieszka Łukaszewska — acquisition of neuropsychological data; Aleksandra Wyciszkiewicz — major role in laboratory procedure design and performance, analysis/interpretation of data; Aleksandra Wypych, Zbigniew Serafin — major role in acquisition of radiographic data, analysis/interpretation of data; Justyna Marcinkowska — analysis/interpretation of data, statistics; Wojciech Kozubski — analysis/interpretation of data; Alicja Kalinowska-Łyszczarz — drafting/revision of manuscript for content, including medical writing for content, major role in acquisition of data, study concept or design, analysis or interpretation of data, supervision of project.*

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References

1. Reich DS, Lucchinetti CF, Calabresi PA. Multiple sclerosis. N Engl J Med. 2018; 378(2): 169–180, doi: 10.1056/NEJMra1401483, indexed in Pubmed: 29320652.
2. Portaccio E, Amato M. Cognitive impairment in multiple sclerosis: an update on assessment and management. NeuroSci. 2022; 3(4): 667–676, doi: 10.3390/neurosci3040048.
3. Betscher E, Guenter W, Langdon DW, et al. Polish validation of the Brief International Cognitive Assessment for Multiple Sclerosis (BICAMS battery): correlation of cognitive impairment with mood

disorders and fatigue. Neurol Neurochir Pol. 2021; 55(1): 59–66, doi: 10.5603/PJNNS.a2020.0080, indexed in Pubmed: 33047783.
4. Kania K, Ambrosius W, Dyczkowska K, et al. Prevalence and prognostic value of prodromal symptoms in relapsing-remitting multiple sclerosis. Neurol Neurochir Pol. 2023; 57(3): 289–296, doi: 10.5603/PJNNS.a2023.0035, indexed in Pubmed: 37144905.
5. Kania K, Ambrosius W, Kozubski W, et al. The impact of disease modifying therapies on cognitive functions typically impaired in multiple sclerosis patients: a clinician's review. Front Neurol. 2023; 14: 1222574, doi: 10.3389/fneur.2023.1222574, indexed in Pubmed: 37503514.
6. Ruano L, Portaccio E, Goretti B, et al. Age and disability drive cognitive impairment in multiple sclerosis across disease subtypes. Mult Scler. 2017; 23(9): 1258–1267, doi: 10.1177/1352458516674367, indexed in Pubmed: 27738090.
7. Eijlers AJC, van Geest Q, Dekker I, et al. Predicting cognitive decline in multiple sclerosis: a 5-year follow-up study. Brain. 2018; 141(9): 2605–2618, doi: 10.1093/brain/awy202, indexed in Pubmed: 30169585.
8. Johnen A, Bürkner PC, Landmeyer NC, et al. German Competence Network Multiple Sclerosis (KKNMS). Can we predict cognitive decline after initial diagnosis of multiple sclerosis? Results from the German National early MS cohort (KKNMS). J Neurol. 2019; 266(2): 386–397, doi: 10.1007/s00415-018-9142-y, indexed in Pubmed: 30515631.
9. Artemiadis A, Bakirtzis C, Ifantopoulou P, et al. The role of cognitive reserve in multiple sclerosis: A cross-sectional study in 526 patients. Mult Scler Relat Disord. 2020; 41: 102047, doi: 10.1016/j.msard.2020.102047, indexed in Pubmed: 32182469.
10. Williams T, Tur C, Eshaghi A, et al. Serum neurofilament light and MRI predictors of cognitive decline in patients with secondary progressive multiple sclerosis: Analysis from the MS-STAT randomised controlled trial. Mult Scler. 2022; 28(12): 1913–1926, doi: 10.1177/13524585221114441, indexed in Pubmed: 35946107.
11. Friedova L, Motyl J, Srpova B, et al. The weak association between neurofilament levels at multiple sclerosis onset and cognitive performance after 9 years. Mult Scler Relat Disord. 2020; 46: 102534, doi: 10.1016/j.msard.2020.102534, indexed in Pubmed: 33032055.
12. Kalinowska-Łyszczarz A, Pawlak MA, Michalak S, et al. Cognitive deficit is related to immune-cell beta-NGF in multiple sclerosis patients. J Neurol Sci. 2012; 321(1-2): 43–48, doi: 10.1016/j.jns.2012.07.044, indexed in Pubmed: 22877509.
13. Henry JD, Beatty WW. Verbal fluency deficits in multiple sclerosis. Neuropsychologia. 2006; 44(7): 1166–1174, doi: 10.1016/j.neuropsychologia.2005.10.006, indexed in Pubmed: 16293271.
14. Nygaard GO, Celius EG, de Rodez Benavent SA, et al. A longitudinal study of disability, cognition and gray matter atrophy in early multiple sclerosis patients according to evidence of disease activity. PLoS One. 2015; 10(8): e0135974, doi: 10.1371/journal.pone.0135974, indexed in Pubmed: 26280173.
15. Ozakbas S, Piri Cinar B, Yigit P, et al. Multiple Sclerosis Research Group. Five-year real-world data on fingolimod treatment's effects on cognitive function. Mult Scler Relat Disord. 2021; 54: 103089, doi: 10.1016/j.msard.2021.103089, indexed in Pubmed: 34198030.
16. Skorve E, Lundervold AJ, Torkildsen Ø, et al. A two-year longitudinal follow-up of cognitive performance assessed by BICAMS in newly diagnosed patients with MS. Mult Scler Relat Disord. 2020; 46: 102577, doi: 10.1016/j.msard.2020.102577, indexed in Pubmed: 33296975.
17. London F, De Haan A, Benyahia Z, et al. Cognitive trajectories in relapsing-remitting multiple sclerosis: Evidence of multiple evolutionary trends. Mult Scler Relat Disord. 2023; 77: 104848, doi: 10.1016/j.msard.2023.104848, indexed in Pubmed: 37390678.
18. Dimitrov I, Kirkova V, Kaprelyan A, et al. Application of the Rey-Osterrieth complex figure test for assessment of cognitive impairment in multiple sclerosis. Scripta Scientifica Medica. 2015; 47(3): 59, doi: 10.14748/ssm.v47i3.1303.
19. Moridi T, Stawiarz L, McKay KA, et al. Association between brain volume and disability over time in multiple sclerosis. Mult Scler J Exp Transl Clin. 2022; 8(4): 20552173221144230, doi: 10.1177/20552173221144230, indexed in Pubmed: 36570871.
20. Cruz-Gomez ÁJ, Forero L, Lozano-Soto E, et al. Cortical thickness and serum nfl explain cognitive dysfunction in newly diagnosed patients with multiple sclerosis. Neurol Neuroimmunol Neuroinflamm. 2021; 8(6), doi: 10.1212/NXI.0000000000001074, indexed in Pubmed: 34465616.
21. Ehling R, Amprosi M, Kremmel B, et al. Second language learning induces grey matter volume increase in people with multiple sclerosis. PLoS One. 2019; 14(12): e0226525, doi: 10.1371/journal.pone.0226525, indexed in Pubmed: 31869402.
22. Matías-Guiu JA, Cortés-Martínez A, Montero P, et al. Identification of Cortical and Subcortical Correlates of Cognitive Performance in Multiple Sclerosis Using Voxel-Based Morphometry. Front Neurol. 2018; 9: 920, doi: 10.3389/fneur.2018.00920, indexed in Pubmed: 30420834.
23. Crosson B, Benefield H, Cato MA, et al. Left and right basal ganglia and frontal activity during language generation: contributions to lexical, semantic, and phonological processes. J Int Neuropsychol Soc. 2003; 9(7): 1061–1077, doi: 10.1017/S135561770397010X, indexed in Pubmed: 14738287.
24. Wang X, Zhu Q, Yan Z, et al. Enlarged choroid plexus related to iron rim lesions and deep gray matter atrophy in relapsing-remitting multiple sclerosis. Mult Scler Relat Disord. 2023; 75: 104740, doi: 10.1016/j.msard.2023.104740, indexed in Pubmed: 37146422.
25. Ricigliano VAG, Morena E, Colombi A, et al. choroid plexus enlargement in inflammatory multiple sclerosis: 3.0-t mri and translocator protein pet evaluation. Radiology. 2021; 301(1): 166–177, doi: 10.1148/radiol.2021204426, indexed in Pubmed: 34254858.
26. Müller J, Sinnecker T, Wendebourg MJ, et al. Choroid plexus volume in multiple sclerosis vs neuromyelitis optica spectrum disorder: a retrospective, cross-sectional analysis. Neurol Neuroimmunol Neuroinflamm. 2022; 9(3), doi: 10.1212/NXI.0000000000001147, indexed in Pubmed: 35217580.
27. Klistorner S, Barnett MH, Parratt J, et al. Choroid plexus volume in multiple sclerosis predicts expansion of chronic lesions and brain atrophy. Ann Clin Transl Neurol. 2022; 9(10): 1528–1537, doi: 10.1002/acn3.51644, indexed in Pubmed: 36056634.
28. Jankowska A, Chwojnicki K, Grzywińska M, et al. choroid plexus volume change-a candidate for a new radiological marker of ms progression. Diagnostics (Basel). 2023; 13(16), doi: 10.3390/diagnostics13162668, indexed in Pubmed: 37627928.
29. Steenwijk MD, Geurts JJG, Daams M, et al. Cortical atrophy patterns in multiple sclerosis are non-random and clinically relevant. Brain. 2016; 139(Pt 1): 115–126, doi: 10.1093/brain/awv337, indexed in Pubmed: 26637488.
30. Jacobsen C, Hagemeier J, Myhr KM, et al. Brain atrophy and disability progression in multiple sclerosis patients: a 10-year follow-up study. J Neurol Neurosurg Psychiatry. 2014; 85(10): 1109–1115, doi: 10.1136/jnnp-2013-306906, indexed in Pubmed: 24554101.
31. Roosendaal SD, Bendfeldt K, Vrenken H, et al. Grey matter volume in a large cohort of MS patients: relation to MRI

parameters and disability. Mult Scler. 2011; 17(9): 1098–1106, doi: 10.1177/1352458511404916, indexed in Pubmed: 21586487.
32. Eshaghi A, Prados F, Brownlee WJ, et al. MAGNIMS study group. Deep gray matter volume loss drives disability worsening in multiple sclerosis. Ann Neurol. 2018; 83(2): 210–222, doi: 10.1002/ana.25145, indexed in Pubmed: 29331092.
33. Tao G, Datta S, He R, et al. Deep gray matter atrophy in multiple sclerosis: a tensor based morphometry. J Neurol Sci. 2009; 282(1-2): 39–46, doi: 10.1016/j.jns.2008.12.035, indexed in Pubmed: 19168189.
34. Slezáková D, Kadlic P, Jezberová M, et al. Brain volume loss in multiple sclerosis is independent of disease activity and might be prevented by early disease-modifying therapy. Neurol Neurochir Pol. 2023; 57(3): 282–288, doi: 10.5603/PJNNS.a2023.0031, indexed in Pubmed: 37144903.
35. Novotna M, Paz Soldán MM, Abou Zeid N, et al. Poor early relapse recovery affects onset of progressive disease course in multiple sclerosis. Neurology. 2015; 85(8): 722–729, doi: 10.1212/WNL.0000000000001856, indexed in Pubmed: 26208962.
36. Lublin FD, Häring DA, Ganjgahi H, et al. How patients with multiple sclerosis acquire disability. Brain. 2022; 145(9): 3147–3161, doi: 10.1093/brain/awac016, indexed in Pubmed: 35104840.
37. Hartel M, Kluczewska E, Pierzchała K, et al. What you cannot get from routine MRI of MS patient and why — the growing need for atrophy assessment and seeing beyond the plaque. Neurol Neurochir Pol. 2016; 50(2): 123–130, doi: 10.1016/j.pjnns.2016.01.007, indexed in Pubmed: 26969569.
38. Kalinowska-Łyszczarz A, Michalak S, Pawlak MA, et al. Serum sPECAM-1 and sVCAM-1 levels are associated with conversion to multiple sclerosis in patients with optic neuritis. J Neuroimmunol. 2016; 300: 11–14, doi: 10.1016/j.jneuroim.2016.10.003, indexed in Pubmed: 27806869.
39. Kuenz B, Lutterotti A, Khalil M, et al. Plasma levels of soluble adhesion molecules sPECAM-1, sP-selectin and sE-selectin are associated with relapsing-remitting disease course of multiple sclerosis. J Neuroimmunol. 2005; 167(1-2): 143–149, doi: 10.1016/j.jneuroim.2005.06.019, indexed in Pubmed: 16040131.
40. Rieckmann P, Altenhofen B, Riegel A, et al. Correlation of soluble adhesion molecules in blood and cerebrospinal fluid with magnetic resonance imaging activity in patients with multiple sclerosis. Mult Scler. 1998; 4(3): 178–182, doi: 10.1177/13524585980040031, indexed in Pubmed: 9762670.
41. Jasiak-Zatońska M, Michalak S, Osztynowicz K, et al. Relationship between blood-brain permeability and antibodies against aquaporins in neuromyelitis optica spectrum disorders and multiple sclerosis patients. Neurol Neurochir Pol. 2022; 56(4): 308–317, doi: 10.5603/PJNNS.a2022.0007, indexed in Pubmed: 35029294.
42. Amini Harandi A, Siavoshi F, Shirzadeh Barough S, et al. Vascular endothelial growth factor as a predictive and prognostic biomarker for multiple sclerosis. Neuroimmunomodulation. 2022; 29(4): 476–485, doi: 10.1159/000525600, indexed in Pubmed: 35981507.
43. Amato MP, Ponziani G, Siracusa G, et al. Cognitive dysfunction in early-onset multiple sclerosis: a reappraisal after 10 years. Arch Neurol. 2001; 58(10): 1602–1606, doi: 10.1001/archneur.58.10.1602, indexed in Pubmed: 11594918.
44. Moccia M, Lanzillo R, Palladino R, et al. Cognitive impairment at diagnosis predicts 10-year multiple sclerosis progression. Mult Scler. 2016; 22(5): 659–667, doi: 10.1177/1352458515599075, indexed in Pubmed: 26362896.
45. Przybek-Skrzypecka J, Małecka I, Członkowska A, et al. Demographic and clinical profile of patients with multiple sclerosis diagnosed over the last 30 years according to different diagnostic criteria. Neurol Neurochir Pol. 2020; 54(2): 169–175, doi: 10.5603/PJNNS.a2020.0027, indexed in Pubmed: 32242912.
46. Katsari M, Kasselimis DS, Giogkaraki E, et al. A longitudinal study of cognitive function in multiple sclerosis: is decline inevitable? J Neurol. 2020; 267(5): 1464–1475, doi: 10.1007/s00415-020-09720-8, indexed in Pubmed: 32008073.