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Posted Date: 25 July 2025

doi: 10.20944/preprints202507.2165.v1

Keywords: SDMT; Digit Span Forward; Digit Span Backward; GABA-edited 1H – MRS; Multiple Sclerosis



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Article

Digit Span Tests Are More Sensitive than SDMT for Detecting Working Memory Impairment and Correlate with Metabolic Alterations in White Matter and Deep Gray Matter Nuclei in Multiple Sclerosis: A GABA-Edited Magnetic Resonance Spectroscopy Study

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Highlights:

Digit span forward (DSF), and Digit span backward (DSB) tests show high efficiency to test working memory deficit in MS
DSF and DSB are comparable to Single Digit Modality Test (SDMT) to measure cognitive problems in MS
DSB is the most sensitive in identifying MS patients
SDMT correlates with inflammatory, degenerative metabolites and neurotransmitters (Glx, GABA) in the thalamus, hippocampus, and corpus callosum
DSF and DSB correlate with inflammatory, degenerative metabolites and neurotransmitter GABA in caudate nucleus and hypothalamus

Abstract

Background: Single Digit Modality Test (SDMT) has been claimed to be one of the most sensitive tests to measure attention and information processing speed in multiple sclerosis (MS). **Objectives:** We aimed to evaluate the efficacy and usefulness of three brief, easy-to-administer, and repeatable tests: SDMT, Digit Span Forward (DSF), and Digit Span Backward (DSB) in MS patients (MSp), and compare the results with those of healthy volunteers (CON). We were hoping to identify the most sensitive test that would be used in clinical practice on a regular basis. In addition, we tried to identify the metabolic background of the cognitive setting using the advanced radiological method, Mescher-Garwood (MEGA)-edited 1H - Magnetic Resonance Spectroscopy (1H - MRS). **Methods:** 22 relapsing MSp and 23 age-, education-, and gender-matched CON were enrolled. In MSp, clinical disability was evaluated using Expanded Disability Status Scale (EDSS). The SDMT, DSF, and DSB tests were used in all participants. They also underwent a 1H - MRS brain examination. In addition to N-Acetyl-Aspartate (tNAA), Myoinositol (mIns), and Choline (tCho), and Creatine (tCr) containing compounds were also evaluated GABA and Glutamate-Glutamine (Glx) ratios. **Results:** CON were superior to MSp in results of all neurocognitive tests. The DSB was found to be the most sensitive test to identify MSp. The SDMT in MSp correlated with inflammatory and degenerative metabolites in the thalamus, hippocampus, and corpus callosum. A correlation between increased Glx- and GABA-

ratios and SDMT was found. Unlike the SDMT, the DSF and DSB showed correlations with inflammatory metabolites in the caudate nucleus and hypothalamus. DSF correlated with GABA-ratios in hippocampus. **Conclusion:** Our study confirms efficacy of DSF and DSB tests in evaluating working memory cognitive impairment in MS, showing an association of the tests with specific brain metabolites.

Keywords: SDMT; digit span forward; digit span backward; GABA-edited 1H – MRS; multiple sclerosis

1. Introduction

Multiple sclerosis (MS), a chronic, immune-mediated neurodegenerative disorder of the central nervous system (CNS), which collectively contribute to progressive neurological disability [1,2]. While motor and sensory deficits are hallmark clinical features, cognitive dysfunction (CD) represents a profound burden, significantly impairing quality of life, vocational capacity, and psychosocial well-being [3]. CD may manifest early in the disease course, even without significant physical disability, and correlate poorly with conventional magnetic resonance imaging (MRI) markers such as T2 hyperintense lesion load or gadolinium-enhancing activity [4]. This dissociation underscores the limitations of structural imaging in capturing the complex neurobiological substrates of cognitive decline, which are hypothesized to involve microstructural damage, metabolic dysregulation, and network-level disconnection. Emerging evidence suggests that cortical and deep gray matter (GM) atrophy and diffuse white matter (WM) pathology in normal-appearing tissue may drive cognitive impairment. Yet, the molecular mechanisms linking neuroinflammation, neurodegeneration, and neuropsychological deficits remain not well understood [5–7].

One of the advanced MRI methods is 1-proton Magnetic Resonance Spectroscopy (1H - MRS) [8], which detects biochemical alterations in lesioned and normal-appearing brain tissue [4]. In this study was used Mescher-Garwood (MEGA)-edited 1H - MRS [9,10], which allows evaluation of neurotransmitters such as Glutamate with its precursor Glutamine (Glx), and γ -Aminobutyric Acid (GABA), in addition to traditional metabolites reflecting tissue alteration such as N-Acetyl-Aspartate (tNAA), Myoinositol (mIns), and Choline (tCho), and Creatine (tCr)-containing compounds [9,10]. In a short summary, tNAA is a marker of neuronal density and mitochondrial function synthesized in neuronal mitochondria; tCr is a compound central to adenosine triphosphate (ATP) buffering and cellular energy homeostasis; tCho is indicative of membrane phospholipid turnover and upregulated in neuroinflammatory states; mIns is a glial-specific osmolyte elevated in astrogliosis and microglial activation; Glx is an excitatory neurotransmitter involved in synaptic signaling and implicated in excitotoxic injury; GABA has a role as the major inhibitory neurotransmitter referred to as a marker of neuro-plasticity [11,12].

Cognitive function and affective symptoms have been evaluated using a standardized battery of neuropsychological tests and validated self-report measures selected for their established sensitivity to MS-related deficits. The Symbol Digit Modalities Test (SDMT) [13–15], is a validated measure of information processing speed (IPS) and working memory, the primary endpoint for assessing cognitive efficiency in MS. The SDMT emphasizes rapid visual scanning, sustained attention, and cognitive flexibility, domains disproportionately impaired in MS. It has been identified as the most sensitive cognitive marker of disease progression [14,17].

Verbal working memory and attentional capacity could be assessed using the Digit Span subtest from the Wechsler Adult Intelligence Scale (WAIS-IV), administered in both forward (DSF) and backward (DSB) conditions [18]. DSB requires mental manipulation by reversing digit sequences, taxing executive control, and working memory updating. While DSF primarily reflects phonological loop integrity, DSB engages the central executive component of Baddeley's working memory model, making it sensitive to frontal-subcortical circuit dysfunction common in MS [19].

In this study, we aimed to evaluate the efficacy and usefulness of three brief, easy-to-administer, and repeatable tests: SDMT, DSF, and DSB in MS patients, and compare the results with those of healthy volunteers (CON). We were hoping to identify the most sensitive test that would be used in clinical practice on a regular basis. In addition, we tried to find the metabolic background of the cognitive setting using the advanced radiological method, MEGA-edited 1H - MRS. Selected areas of white and gray matter in the brains of MS patients and CON were evaluated; data were compared and correlated with the results of SDMT, DSF, and DSB tests.

2. Methods

2.1. Patients

Patients fulfilling the criteria for definite MS according to McDonald 2017 criteria [2] were randomly selected from the Multiple Sclerosis Centre, University Hospital in Martin, Slovakia, and they were entered the study after signing the written consent. The inclusion criteria were: relapsing course of the disease, the absence of clinical relapse and corticosteroid treatment within at least three months before study entry, EDSS ≤ 5.0 , aged between 18 and 55 years, and the ability and willingness to cooperate in examinations. The exclusion criterion was serious comorbidities and acute depressive symptomatology confirmed by psychological examination.

We recruited 24 MS patients and 23 (7 males, 16 females) age-, education-, and gender-matched healthy participants (CON). Out of all participants, data from two MS patients (1 male 1 female) were not complete, and 1H - MRS data from one CON subject (female) did not meet the technical quality criteria for evaluation, so these subjects were excluded from the analyses. Finally, 22 MS patients (9 males 40.9% and 13 females 59.1%) participated in the study.

In MS patients, clinical disability was evaluated by a neurologist certified in Expanded Disability Status Scale (EDSS) assessment. Cognitive status was tested using the SDMT, DSF and DSB from a neuropsychological battery WAIS-IV by an experienced neuropsychologist.

2.2. Mescher-Garwood GABA-Edited 1H - MRS

Measurements were performed on a 3 T whole-body MR scanner (TIM Trio®, Siemens Healthcare, Erlangen, Germany) using a 32-channel head coil (Siemens Healthcare, Erlangen, Germany) for signal reception. To ensure accurate and reproducible slice positioning, auto-align and three-dimensional T1-weighted anatomical reference images were used (magnetization-prepared two rapid acquisition gradient echoes – MP2RAGE, TR/TE = 4600/3.2 ms, resolution = $0.8 \times 0.8 \times 0.8$ mm³). MRS- measurements were performed using a three-dimensional MEGA-edited sequence (TR/TE = 1600/68 ms, 32 averages, with a measurement time of ~20 min) with localized adiabatic spin echo refocusing selection and spiral encoding [9,10]. For real-time correction, the volumetric, dual-contrast, echo planar imaging-based navigators that update B0 shim, frequency, and motions artifacts were used. All 1H - MRS slices were placed parallel to the anterior commissure–posterior commissure line and covered the centrum semiovale and basal ganglia, with volume of interest-VOI = $90 \times 80 \times 50$ mm³, field of view-FOV = $160 \times 160 \times 80$ mm³, and ~2 cc nominal resolution. The acquired matrix size of $10 \times 10 \times 10$ voxels was interpolated to the $16 \times 16 \times 16$ matrix of multivoxel (CSI, chemical shift imaging) representation.

Required regions were derived based on T1-weighted images resulting in the selection of areas without detectable manifestation of focal WM or GM lesions. All MR-spectra were evaluated in LCModel software (Version 6.3–1, S. Provencher, LC Model, Oakville, ON, Canada) for obtaining following metabolite ratios tNAA/tCr, tCho/tNAA, tCho/tCr, mIns/tNAA, mIns/tCr, Glx/tNAA, Glx/tCr, GABA/tNAA, and GABA/tCr.

For further analytical processes, all selected voxels had to fulfill quality criteria including more than 10 signal-to-noise ratio and less than 0.10 ppm full width at half maximum.

2.3. Statistical Analyses

All statistical analyses were performed in NCSS (version 9.0, LLC. Kaysville, Utah, USA). One-way ANOVA was used for statistical analyses that found no differences between metabolic values in left and right voxels. Differences in demographic and clinical parameters (age, SDMT, DSF, DSB, EDSS) as well as GABA-edited 1H - MRS metabolite ratios between patients and controls were evaluated using Welch two-sample t-test or one-way ANOVA with Bonferroni post-hoc test. Cochran - Armitage test was applied to compare trends of results of DSF and DSB series. Random Forest test was used to stratify variables (cognitive tests and metabolites) according to their predictive power to discriminate between MSp and CON.

3. Results

3.1. Differences Between MSp and CON

CON were superior over MSp in results of all the cognitive tests (SDMT $p < 0.00008$, DSF $p = 0.0008$, DSB $p = <0.0014$). Demographic data are in Table 1.

Table 1. Demographic and basic information about the study groups.

Characteristic	control, N = 22/14F	RR, N = 23	p-value	q-value	Median diff (95% CI)
age	30.00 (25.00, 34.00)	34.00 (27.00, 41.00)	0.2	0.4	-3.0 (-9.0, 2.0)
EDSS	NA	3.28 (1-5)	NA	NA	NA
RR (year)	NA	0.25	NA	NA	NA
Disease Duration (months)	NA	76.5 (10-162)	NA	NA	NA
MRI activity (newT2/Gd + lesions)	NA	3/22 (13.6%)	NA	NA	NA
DMT	NA	NAT 9/22(40.9%) DMF 3/22(13.6%) GA 3/22 (13.6%) FIN 2/22/(9.9%) ALEM 2/22 (9.9%) TERI 1/22(4.5%) None (2(9.9%)			
SDMT	53.50 (47.00, 60.50)	43.00 (35.50, 49.00)	<0.001	0.003	11 (6.0, 17)

Characteristic	control, N = 22/14F	RR, N = 23	p-value	q-value	Median diff (95% CI)
DSF	7,86	6,08	0.001	0.010	
3	0 (0%)	1 (4.3%)			
4	0 (0%)	4 (17%)			
5	1 (4.5%)	6 (26%)			
6	1 (4.5%)	3 (13%)			
7	5 (23%)	4 (17%)			
8	8 (36%)	3 (13%)			
9	7 (32%)	0 (0%)			
10	0 (0%)	1 (4.3%)			
11	0 (0%)	1 (4.3%)			
DSB	7,14	5,09	<0.001	0.003	
3	0 (0%)	2 (8.7%)			
4	1 (4.5%)	10 (43%)			
5	0 (0%)	3 (13%)			
6	4 (18%)	4 (17%)			
7	9 (41%)	2 (8.7%)			
8	6 (27%)	1 (4.3%)			
9	2 (9.1%)	0 (0%)			
10	0 (0%)	1 (4.3%)			

EDSS = Expanded Disability Status Scale, RR = Annualized Relapse Rate, MRI = Magnetic Resonance Imaging, T2 = T2 weighted lesions, Gd+ = gadolinium enhancing lesions, DMT = Disease Modifying Treatment, DSF = Digit Span Forward, DSB = Digit Span Backward.

3.2. Correlations of Brain Metabolites with Cognitive Tests Are in Tables 2–4.

Table 2. Correlation of SDMT with brain metabolites in MSp.

Thalamus R	tNAA/tCr	pval = 0.04	cor = 0.43
Hypothalamus L	Glx/tCr	pval = 0.032	cor = 0.447
Hippocampus L	mIns/tNAA	pval = 0.014	cor = -0.507
CC_splenium	tCho/tNAA	pval = 0.048	cor = -0.417
CC_rostral	GABA/tCr	pval = 0.038	cor = 0.436
CC_genu	tCho/tNAA	pval = 0.021	cor = -0.479

tNAA = N-acetyl Aspartate, Glx = Glutamate and Glutamine, tCho = Choline, GABA = γ -amino butyric acid, mIns = Myoinositol, tCr = Creatine; CC = corpus callosum; R = right, L = Left, Welsh two-sample t-test, $\alpha < 0.05$.

Table 3. Correlation of Digit Span Forward series with brain metabolites in MSp.

Caudate R	mIns/tNAA	pval = 0.022	cor = 0.474
Caudate R	mIns/tCr	pval = 0.021	cor = 0.477
Hypothalamus R	Glx/tCr	pval = 0.01	cor = -0.527
Hypothalamus R	tNAA/tCr	pval = 0.04	cor = -0.432
Hypothalamus L	tCho/tNAA	pval = 0.012	cor = 0.517
Hippocampus L	GABA/tNAA	pval = 0.007	cor = 0.545
Hippocampus L	GABA/tCr	pval = 0.026	cor = 0.464
CC_splenium	mIns/tCr	pval = 0.041	cor = -0.429
CC_genu	NAA/tCr	pval = 0.021	cor = -0.479

tNAA = N-Acetyl Aspartate, Glx = Glutamate and Glutamine, tCho = Choline, GABA = γ-Aminobutyric Acid, mIns = Myoinositol, tCr = cCreatine; Caudate = Caudate Nucleus, CC = Corpus Callosum; R = right, L = left, Welsh two-sample t-test, $\alpha < 0.05$.

3.3. Evaluation of the Most Significant Predictors of Multiple Sclerosis

RF as the significant predictors identified See Figure 1.

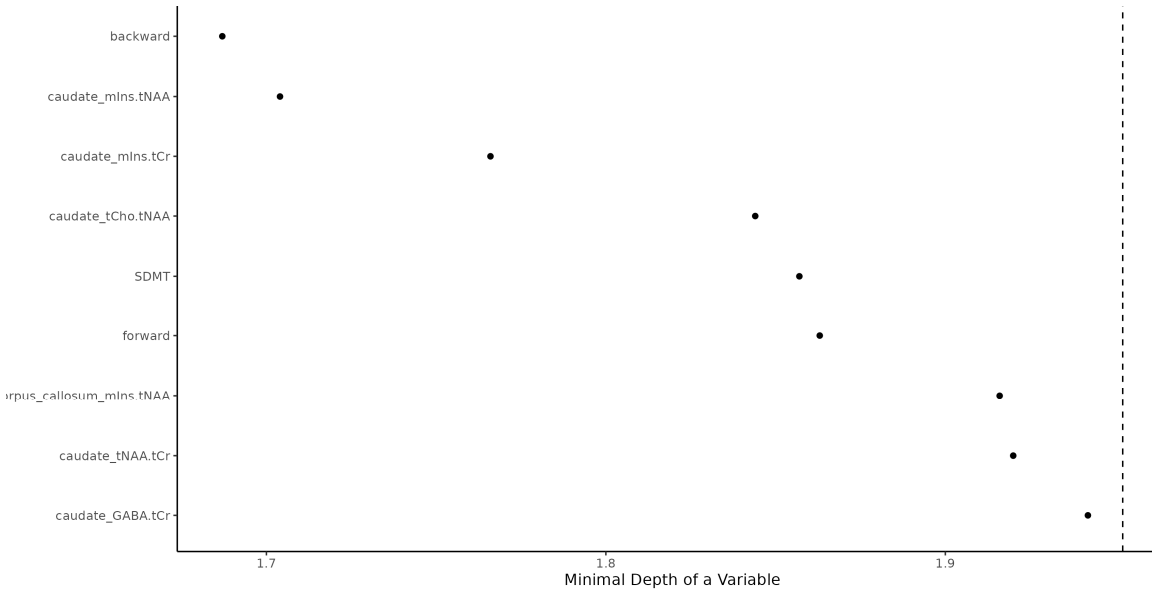


Figure 1. Random Forest identification of multiple sclerosis patients. Backward = Digit Span Backward; caudate_mIns.tNAA = Caudate Nucleus Myoinositol/tN-Acetyl Aspartate; caudate_mIns.tCr = Caudate Nucleus Myoinositol/t-Creatinin; caudate_tCho.tNAA = Caudate Nucleus tCholin/tN-Acetyl Aspartate; SDMT = Single Digit Modality Test; forward = Digit Span Forward; corpus callosum_mIns.tNAA = Corpus Callosum Myoinositol/tN-Acetyl Aspartate; caudate_tNAA.tCr = Caudate Nucleus tN-Acetyl Aspartate/tCreatinin; caudate_GABA.tCr = Caudate Nucleus γ-Aminobutyric acid/tCreatinin.

1. DSB series (Fisher exact test pval < 0.001),
- 2.caudate nucleus mIns/tNAA (Wilcoxon pval < 0.001; increased in MS),
- 3.caudate nucleus mIns/tCr (Wilcoxon pval < 0.001; increase in MS),
- 4.caudate nucleus tCho/tNAA (Wilcoxon pval < 0.001; increased in MS),
- 5.SDMT (Wilcoxon pval < 0.001; decreased in MS),
- 6.DSF forward series (Fisher exact test pval = 0.001),
- 7.corpus callosum mIns/tNAA (Wilcoxon pval = 0.001; increased in MS),
- 8.caudate nucleus tNAA/tCr (Wilcoxon pval = 0.016; decreased in MS),
- 9.caudate nucleus GABA/tCr (Wilcoxon pval = 0.001; decreased in RR).

Some 20% false positive and 20% false negative rate should be expected when using the RF machine learning algorithm (trained with the above predictors) when used for predicting whether a person is MS or CON. RF results prediction power was high, area under a curve was 86%.

As the DSB and DSF series increases, the relative proportion of MSp decreases. It was hard to find MSp among people with high DSB and DSF series value (Figure 2).

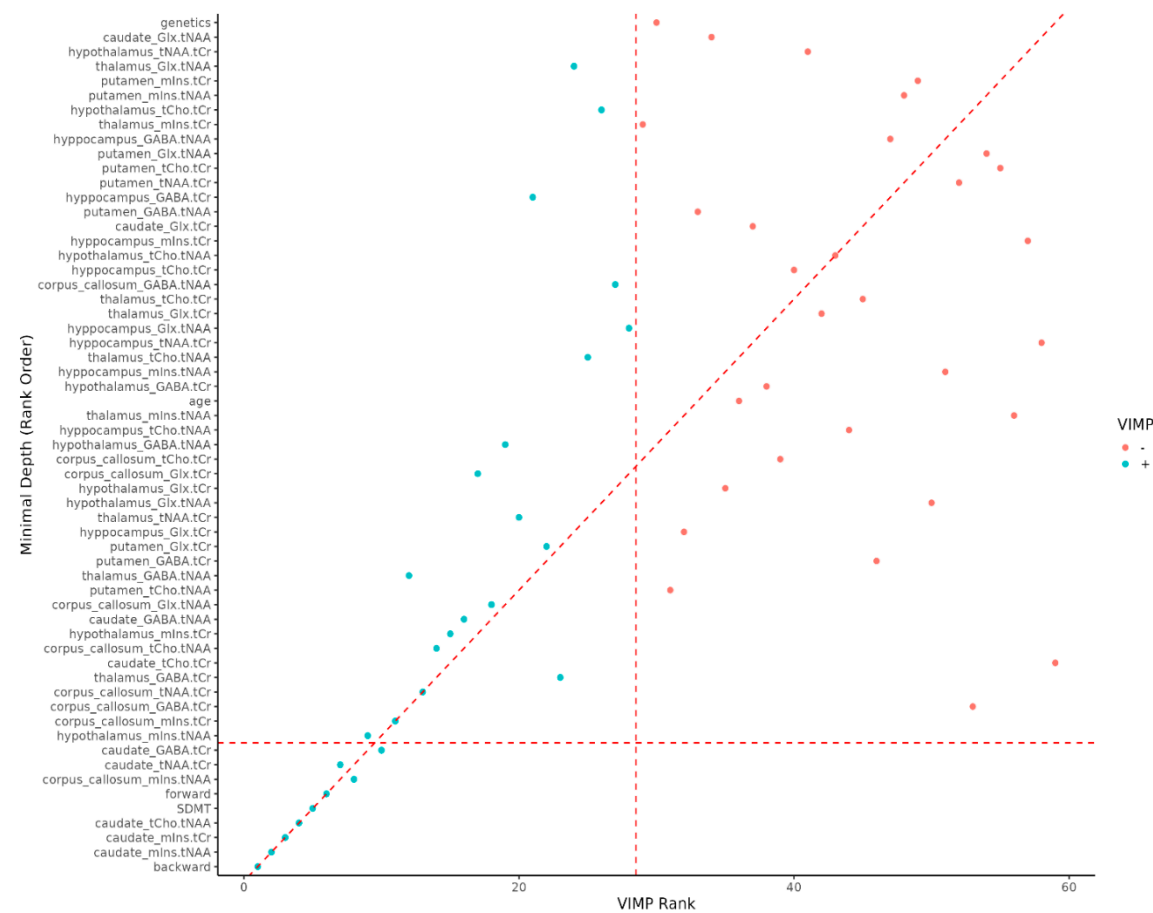


Figure 2. Variable importance rank. VIMP = variable importance, Backward = Digit Span Backward; caudate_mIns.tNAA = Caudate Nucleus Myoinositol/tN-Acetyl Aspartate; caudate_mIns.tCr = in Caudate Nucleus Myoinositol/t-Creatinin; caudate_tCho.tNAA = Caudate Nucleus tCholin/tN-Acetyl Aspartate; SDMT = Single Digit Modality Test; forward = Digit Span Forward; corpus callosum_mIns.tNAA = Corpus Callosum Cyoinositol/tN-acetyl aspartate; caudate_tNAA.tCr = Caudate Nucleus tN-Acetyl Aspartate/tCreatinin; caudate_GABA.tCr = Caudate Nucleus γ -Aminobutyric acid/tCreatinin.

4. Discusssion

Our results confirm that MS patients performed significantly worse than healthy controls on neurocognitive tests. The SDMT is currently considered the most sensitive instrument for evaluating processing speed in MS. Impairment in processing speed in MS has been shown to underlie deficits in working memory, executive function, and learning [13–15]. However, we also found substantial deficits in working memory and attention in MS_p, as assessed by Digit Span tests. The Digit Span task assesses attention, encoding, auditory processing, and working memory capacity. Moreover, DSB was found as the most sensitive test to identify MS_p. These results support Beatty’s [20] hypothesis that MS patients experience both a generalized difficulty in sustaining concentration and a more specific deficit in attentional resource allocation under multi-task demands.

To explore the neural correlates of cognitive dysfunction, we analyzed brain metabolites linked to performance on different cognitive tests.

4.1. SDMT

In our study, lower SDMT scores in GM of MS patients correlated with thalamic neuronal loss (decreased tNAA/tCr ratio reflects diminished neuronal integrity), hippocampal microglial activation (elevated mIns/tNAA was associated with poorer SDMT performance), and hypothalamic glutamate reduction (decreased Glx/tCr correlated with lower SDMT scores).

In the corpus callosum (WM), SDMT impairment was associated with increased choline (tCho/tCr), indicating intensified demyelination, decreased GABA (GABA/tCr), suggesting reduced inhibitory neurotransmission.

SDMT performance has been previously linked to volumes of the thalamus, cerebellum, putamen, and occipital cortex [21,22]. Although we did not measure thalamic volume directly, metabolite-based findings (i.e., reduced tNAA/tCr) support the association between thalamic integrity and SDMT scores. In early-stage MS, we previously reported correlations between elevated tCho and mIns ratios and lower SDMT scores [23], suggesting that metabolic alterations may anticipate neuronal loss. Additionally, 7 T MRS studies in older versus younger individuals report higher glia-related metabolites (mIns, tCr, tCho) particularly in hippocampus and thalamus [24]. Our hippocampal mIns/tNAA findings align with those results, supporting a potential role of sustained microglial activity in metabolic dysfunction and neurotoxicity in MS.

Heightened hypothalamic glutamate levels correlated with higher SDMT scores, potentially reflecting increased excitatory neurotransmission to support attention and processing speed via cortico-subcortical pathways.

Higher tCho/tNAA ratios in the corpus callosum corresponded with lower SDMT scores—consistent with increased demyelination and axonal loss [8,25]. Morphometric and diffusion studies have reinforced the role of corpus callosum integrity in cognitive processing speed, including SDMT performance [26]. Moreover, we observed positive correlation between SDMT and GABA levels in the rostral corpus callosum. GABA is a key inhibitory neurotransmitter crucial for maintaining excitatory-inhibitory balance in cortical circuits. Previous GABA-focused MRS studies in MS did not specifically assess the corpus callosum [27–29], highlighting the novelty of our findings.

4.2. Digit Span Forward and Digit Span Backward Series

In contrast to SDMT scores, higher Digit Span Forward and Backward scores—indicating better working memory—were positively correlated with increased mIns ratios in the caudate nucleus, suggesting microglial activation. Although this association has not previously been reported in MS, parallel findings in Human Immunodeficiency Virus (HIV) patients show that working memory deficits are linked to elevated glial metabolites, including mIns in the caudate and associated white matter networks [30].

The caudate nucleus plays a vital role in strategic planning, goal-directed behavior, and both visual and verbal working memory via cortico-subcortical circuits [31–34]. Functional MRI studies show either caudate nucleus or hippocampus activation specifically during spatial working memory tasks [31,32]. Conversely, SDMT performance relies primarily on information processing speed and engages frontoparietal, visual-attention, and cerebellar networks, with minimal basal ganglia involvement [26].

In our cohort, higher DSF scores correlated with increased hippocampal GABA levels. Preclinical and clinical studies indicate that reduced GABAergic inhibition impairs memory and attention [35], and that lower GABA levels in the prefrontal cortex are linked to greater working memory decline under cognitive load [36]. Moreover, reduced hippocampal GABA has been observed in MS patients [27,28], and is associated with cognitive and motor deficits [23,37].

The GABAergic system is critical for synchronizing large neuronal populations. Inhibitory projections, such as those connecting the medial septum, hippocampus, and entorhinal cortex, are crucial for modulating oscillatory activity underlying episodic memory formation [38].

Additionally, both DSF and DSB scores were negatively correlated with hypothalamic tNAA/tCr and positively with tCho/tNAA, indicating neuronal loss and demyelination. Although comparable

data are scarce, these findings align with the known role of the hypothalamus in cortico-subcortical pathways that support learning and motivated behavior [39].

Finally, our data extend findings by Llufríu et al [40], who identified mIns/tNAA in normal-appearing white matter as predictive of disability in MS. We similarly observed correlations of mIns/tCr and tNAA/tCr in the corpus callosum with reduced DSF and DSB scores, reinforcing the use of these metabolite ratios as biomarkers of cognitive decline.

4.3. Limitation of the Study and Suggestion for Further Research

Our study has also several limitations. First, we tested relatively small number of participants. Second, from a technical perspective, employing absolute metabolite quantification rather than ratio-based analysis could provide more precise biochemical measurements. Additionally, advanced 1H-MRS protocols with higher spatial resolution (7T MRI systems) may improve the detection of localized pathological changes in the brain subregions and also brain cortex. fMRI studies could also bring better insights into functional connections between activated brain regions. These methodological refinements could significantly enhance future investigations of structure-function relationships in MS-related cognitive pathology.

5. Conclusion

We can conclude, SDMT but also DSB and DSF are the most reliable and sensitive neuropsychological test, leading to the idea that it may serve as a screen for neuropsychological impairment in MS in daily bases. Moreover, DSB test was found to be the most sensitive in determining MS-related working memory impairment. Additionally, our data show unique metabolic changes in deep GM and WM of the brain in correlation with cognitive tests. The results confirm a hypothesis about SM as a whole brain disease.

Supplementary Materials: The following supporting information can be downloaded at the website of this paper posted on Preprints.org.

Author Contributions: EK, PH, WB contributed to the conception and design of the study; JG, PH, MG, LK contributed to the acquisition and analysis of data; JG, NK, PH, EK contributed to drafting the text or preparing the figures; EK, PH, MG revising it critically for important intellectual content;

Authorship contribution statement: **Ján Grossmann:** Writing – original draft, Visualization, Methodology, Investigation, Data curation, Conceptualization; **Marián Grendár:** Writing – review & editing, Supervision, Methodology, Funding acquisition, Conceptualization; **Petra Hnilicová:** Writing – review & editing, Supervision, Methodology, Funding acquisition, Conceptualization; **Wolfgang Bogner:** Supervision, Methodology, Funding acquisition, Conceptualization; **Nina Kováčiková:** Writing – original draft, Visualization, Methodology, Investigation, Data curation; **Lucia Kotuľová:** Methodology, Investigation, Data curation, Conceptualization; **Ema Kantorová:** Writing – review & editing, Supervision, Methodology, Funding acquisition, Conceptualization.

Funding sources: This work was supported by the Grant VEGA of The Ministry of Education, Research, Development and Youth of the Slovak Republic, No: 1/0092/22. The funding sources had no involvement in the study design, the collection, analysis, and interpretation of the data, or in the decision to submit this article for publication. The corresponding author had the final responsibility for the decision to submit for publication.

Institutional Review Board Statement: The study was conducted in accordance with the Declaration of Helsinki, and approved by the Institutional Ethics Committee of Jessenius Faculty of Medicine in Martin, Comenius University Bratislava No. EK1849/2016 and No. EK 27/2021, for studies involving humans.

Informed Consent Statement: Informed consent was obtained from all subjects involved in the study. Written informed consent has been obtained from the patients to publish this paper.

Data Availability Statement:

Acknowledgments: Generative AI and AI-assisted technologies was used in the writing process to improve the readability and language of the manuscript.

Conflicts of Interest: Ján Grossmann has nothing to declare; Marián Grendár has nothing to declare; Petra Hnilicová has nothing to declare; Nina Kováčiková has nothing to declare; Lucia Kotuľová has nothing to declare; Wolfgang Bogner has nothing to declare; Ema Kantorová in the last five years, has had travel expenses or registration fees prepaid or reimbursed to present at international conferences.

Abbreviations

MS: Multiple Sclerosis
 MSp: Multiple Sclerosis Patients
 DSF: Digit span forward
 DSB: Digit span backward
 SDMT: Single Digit Modality Test
 1H-MRS: 1-Proton Magnetic Resonance Spectroscopy
 CD: cognitive dysfunction
 WM: white matter
 GM: gray matter
 Glx: Glutamine and Glutamate
 GABA: γ -Amino Butyric Acid
 CON: healthy volunteers
 MEGA-edited: Mescher-Garwood -edited
 tNAA: N-acetyl-aspartate
 mIns: myoinositol
 tCho: choline
 tCr: creatine
 ATP: adenosine triphosphate
 EDSS: Expanded Disability Status State
 IPS: information processing speed
 WAIS-IV: Wechsler Adult Intelligence Scale
 CSI: chemical shift imaging
 fMRI: functional Magnetic Resonance Imaging
 HIV: Human Immunodeficiency Virus
 7 T: 7 Tesla

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