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Baseline cerebral metabolism predicts fatigue and cognition in Multiple Sclerosis patients

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ABSTRACT

Background: Cerebral metabolic rate of oxygen ($CMRO_2$), a measure of global oxygen metabolism, reflects resting cellular activity. The mechanisms underlying fatigue and cognitive dysfunction in multiple sclerosis (MS) remain unknown. If fatigue indeed reflects ongoing autoimmune activity and cortical reorganization, and cognitive decline is the result of gray matter atrophy and white matter degeneration, we postulate that changes in $CMRO_2$ should reflect disease activity and predict these symptoms.

Objective: We sought to utilize T_2-Relaxation-Under-Spin-Tagging (TRUST) and phase-contrast (PC) MRI to measure global $CMRO_2$ to understand its relationships to white matter microstructure, fatigue and cognitive dysfunction.

Methods: We measured venous oxygenation (TRUST) and cerebral blood flow (PC-MRI) in superior sagittal sinus to calculate global $CMRO_2$ and diffusion tensor imaging (DTI) to evaluate white matter microstructure in healthy controls (HC) and MS patients. Participants underwent neuropsychological examinations including Modified Fatigue Impact Scale (MFIS) and Symbol-Digit-Modalities Test (SDMT).

Results: We observed lower $CMRO_2$ in MS patients compared to HC. After controlling for demographic and disease characteristics (i.e., age, education, disability, lesion volume), $CMRO_2$ predicted increased fatigue (MFIS) and reduced cognitive performance (SDMT) in MS patients. Finally, MS patients with higher $CMRO_2$ have reduced FA in normal-appearing white-matter.

Conclusion: Altogether, these results suggest that increased $CMRO_2$ reflects ongoing demyelination and autoimmune activity which plays an important role in both fatigue and cognitive dysfunction.

1. Introduction

Multiple Sclerosis (MS) is an immune-mediated, demyelinating disease of the central nervous system. Approximately 85% of MS patients present with relapsing-remitting disease course (RRMS) resulting in recurrent and reversible neurological deficits. Fatigue and cognitive dysfunction are two of the most prominent symptoms in MS and there are currently no available therapies. Furthermore, mechanisms underlying these symptoms remain poorly understood.

Cerebral metabolic rate of oxygen ($CMRO_2$) is a measure of oxygen metabolism throughout the brain, reflecting resting cellular activity. Previous work in MS has elucidated general cerebral hypometabolism in gray matter and stable lesions, but hypermetabolism in Gadolinium-enhancing lesions with axonal damage (D'haeseleer et al., 2015; Schiepers et al., 1997; Sivakolundu et al., 2019a; Sun et al., 1998). Similarly, pathological studies have shown that autoimmune activity and demyelination lead to higher density of axon channels and thus, probably increased neuronal energy demand (e.g., oxygen metabolism; Ames, 2000; Blokhin et al., 2008; Craner et al., 2004; Witte et al., 2014). Together, these findings suggest that measures of oxygen metabolism, such as $CMRO_2$, could (1) provide valuable insight into MS pathophysiology, (2) distinguish active from stable disease, and (3) provide outcome measures for therapies aimed at ameliorating MS symptoms (see also Paling et al., 2011).

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One of the most common symptoms in Multiple Sclerosis is fatigue that occurs in ~75% of patients (Krupp et al., 1988; Lerdal et al., 2007). Fatigue is also considered to be one of the most debilitating, with associations with reduced quality of life (Krupp et al., 1988). The pathophysiology of fatigue remains poorly understood but the primary mechanism proposed to underlie fatigue involves autoimmune inflammatory activity of the central nervous system, leading to demyelination and axonal loss (Kos et al., 2008). However, fatigue may be multifactorial including associations with affective disorders and sleep disturbance; thus, fatigue remains difficult to treat (Braley and Chervin, 2010; Kos et al., 2008). Additionally, current metrics of fatigue rely on subjective self-report surveys (Flachenecker et al., 2002; Kos et al., 2008) and there are no objective techniques capable of monitoring or predicting fatigue.

Cognitive dysfunction occurs in ~43 to 70% of MS patients (Chiaravalloti and DeLuca, 2008). The cognitive domain most commonly impaired is processing speed, which can be measured with the Symbol Digit Modalities Test (SDMT; Benedict et al., 2006). Previous studies suggest that gray matter atrophy and white matter degeneration contribute to reduced processing speed (Benedict et al., 2004; Christodoulou et al., 2003; Sivakolundu et al., 2019b) because performance relies on rapid communication between distant brain regions via white matter tracts. Performance on SDMT also declines during acute relapses (Benedict et al., 2014) suggesting a role for inflammatory pathology in cognitive dysfunction. However, the mechanism leading to acute and sustained cognitive decline remains largely unknown.

To the extent that fatigue reflects ongoing autoimmune activity and cortical reorganization, and cognitive decline is the result of inflammation and neurodegeneration, we postulate that change in cerebral metabolic rate of oxygen ($CMRO_2$) is a common component of the underlying mechanism for both of these symptoms (Krupp and Elkins, 2000). One previous study utilized MRI to demonstrate relationships between whole-brain $CMRO_2$, lesion volume, and physical disability (Ge et al., 2012), but the relationships to fatigue and cognitive dysfunction remain unknown. Thus, we sought to test the hypothesis that changes in $CMRO_2$ reflect disease activity and can predict fatigue and cognitive dysfunction. Understanding these relationships between $CMRO_2$ and symptomology could provide new insights into the pathophysiology of MS-related fatigue and cognitive decline and indicate a fast, clinically feasible prognostic measure.

2. Methods

2.1. Participants

Thirty-three MS patients and 16 age- and sex-matched HC (see Table 1) were recruited from the Clinical Center for MS at the University of Texas Southwestern (UTSW) Medical Center, local MS support groups, and from advertisements and flyers distributed throughout the Dallas-Fort-Worth Metroplex. All study procedures were approved by the University of Texas at Dallas and University of Texas Southwestern Medical Center Institutional Review Boards in accordance with the guidelines of the Declaration of Helsinki and Belmont Report.

Table 1
Sample characteristics.

	HC (n = 16)	MS patients (n = 33)	p-value
Demographics			
Age (years)	42.56 (10.67)	46.52 (10.31)	0.220
Gender (M/F)	6/10	14/19	0.742[a]
Education (years)	17.00 (2.07)	16.25 (2.58)	0.317
Patient characteristics			
Disease Duration (years)	–	9.31 (8.08)	–
Lesion volume (mL)	–	5.27 (6.17)	–
EDSS	–	2.58 (1.76)	–
MS Disease-Modifying Treatments (%)	–	67.74%[b]	–
Anti-Depressants (%)	–	31.25%[c]	–

[a] Pearson chi-square test.
[b] Information on DMT was unavailable for 2 participants.
[c] Information on anti-depressants was unavailable for 1 participant.

All participants underwent screening procedures. Individuals enrolled in the study were free from MR-contraindicators, substance abuse, and significant medical, neurological (other than MS), or psychiatric conditions unrelated to their MS disease course. All participants were right-handed, at least high-school educated (i.e., 12 years of education), and native English speakers. Due to the inclusion of hypercapnia, all individuals were required to be non-smokers with no history of cardiorespiratory or cerebrovascular conditions. All prospective participants underwent the Telephone Interview for Cognitive Status Modified (TICS-m; Brandt et al., 1988) to ensure cognitive capability to sign informed consent (TICS-m score > 22).

All MS patients had confirmed Relapsing-Remitting MS (RRMS) diagnoses by the 2010 McDonald criteria (Polman et al., 2011), were > 6 months post last exacerbation, were > 30 days post-corticosteroid treatment, and were either treatment-naïve or treatment-stable (> 3 months). 67.74% of MS patients were on disease-modifying therapies. Concurrent use of CNS-modifying drugs (e.g., anti-depressants) were allowed if individuals were treatment-stable.

3. Clinical assessment

After scanning, all participants underwent neuropsychological evaluation including expanded disability status scale (EDSS; Verdier-Taillefer et al., 1994), Beck Depression Index (BDI-II; Beck and Steer, 1993), Modified Fatigue Index scales (MFIS; Flachenecker et al., 2002) and Symbol-Digit-Modalities Test (SDMT; Benedict et al., 2006).

3.1. Imaging data and acquisition

All imaging was conducted at the UTSW Advanced Imaging Research Center (AIRC) on a Philips 3T MRI system (Philips Medical Systems, Best, The Netherlands) with a 32-channel head coil. T_2-Relaxation-Under-Spin-Tagging (TRUST) was acquired to assess venous oxygenation (Y_v; Lu and Ge, 2008). Phase contrast (PC) MRI was acquired to estimate whole-brain cerebral blood flow (CBF; Liu et al., 2013). Using Y_v and CBF from the superior sagittal sinus, baseline global $CMRO_2$ was calculated (*See* 2.3 Data Analysis). High resolution magnetization prepared rapid gradient echo (MPRAGE) was acquired for co-registration and atrophy estimates. T_2-weighted fluid-attenuated inversion recovery (T_2-FLAIR) was used to assess RRMS lesion volume and Diffusion Tensor Imaging (DTI) was acquired to assess white matter microstructure (WMMS) integrity. One HC did not have diffusion data and one HC was not included due to image artifacts resulting in 14 HC and 33 MS patients for diffusion analyses.

The TRUST sequence was acquired in a transverse slice located parallel to the anterior commissure-posterior commissure (AC-PC) line and going through the superior sagittal sinus (20.1–20.7 mm above the sinus congruence for each participant). The sequence utilized alternating label/control RF pulses to separate pure venous blood from surrounding tissue. The labeling slab was 100 mm thick and the labeling slab gap was 22.5 mm above the imaging slice. A non-selective T_2-preparation train of 180° pulses was also applied for T_2-weighting (*See* Supplemental Fig. 1). The specific sequence parameters were as follows: single-shot echo-planar imaging, excitation flip angle = 90°, T_R = 3000 ms, T_I = 1200 ms, T_E = 3.8 ms, voxel size = 3.44 × 3.44 × 5 mm^3, four different T_2 weightings with eT_Es of 0, 40, 80, and 160 ms, t_{CPMG} = 10 ms, scan duration = 1.2 min (Liu et al., 2013; Lu et al., 2012; Lu and Ge, 2008; Xu et al., 2012).

Phase contrast (PC) MRI was performed using the same imaging

slice location as used in TRUST. PC MRI utilizes phase information to encode the velocity of the venous blood. PC was acquired with the following scan parameters: single slice, flip angle = 15°, T_R = 20 ms, T_E = 6.9 ms, voxel size = 0.45 × 0.45 × 5 mm^3, maximum velocity encoding (VENC) = 80 cm/s, 4 signal averages, scan duration = 0.5 min (Ge et al., 2012; Liu et al., 2013). VENC = 80 cm/s was used here for consistency with the protocol for PC during hypercapnia in the same scan session (though these data were not used in the current study); hypercapnia induces higher velocities, requiring a higher VENC parameter.

High-resolution anatomical data were acquired using a 3D T_1-weighted MPRAGE pulse sequence to provide regions of interest and brain volume with the following parameters: flip angle = 12°, 1 mm^3 isotropic voxel-size, matrix size = 256 × 204 × 160, scan time ~4 min. Diffusion Tensor Imaging (DTI) was acquired with a pulsed gradient spin echo sequence with an echo planar imaging (EPI) readout, SENSE-factor = 2.2, 2 zero and 1 non-zero b-value (1000 s/mm^2) across 30 directions (Jones et al., 1999), T_R/T_E = 6500/69 ms, voxel size = 2.0 × 2.0 mm^2 (reconstructed to 0.88 × 0.88 mm^2), in-plane matrix size = 112 × 112, 65 axial slices, slice thickness = 2.2 mm, no gap, scan time ~ 5 min. 3D sagittal T_2-FLAIR imaging was used to assess RRMS lesion burden with the following scan parameters: effective $T_R/T_I/T_E(T_{E,eq})$ = 4800/1600/344(117) ms, TSE readout = 178 echoes, echo-spacing = 3.5 ms, refocusing flip angle = 120°, matrix size = 228 × 227 × 163, and 1.1 mm^3 isotropic resolution (reconstructed to 1 mm^3), SENSE-factor = 2.6 × 2.

3.2. Data analysis

TRUST and PC images were analyzed using in-house MATLAB code to obtain $CMRO_2$. Multi-echo TRUST provided estimates of T_2 in the superior sagittal sinus (SSS) venous blood, directly related to Yv via $1/T_2 = A + B\cdot(1-Y_v) + C\cdot(1-Y_v)^2$, where A = 6.80/s, B = 0.38/s, and C = 60.3/s for macrovascular hematocrit = 0.42 (Lu et al., 2012; Lu and Ge, 2008). PC magnitude images were manually segmented to obtain the area of the SSS and the mask was applied to phase images to obtain CBF. To obtain whole brain CBF (mL/100 g/min), SSS CBF was multiplied by 2 based on SSS reflecting ~50% of whole brain venous drainage (De Vis et al., 2018) and then normalized to the brain parenchyma volume (gray matter (GM) + white matter (WM) volume; from MPRAGE). A pulse oximeter was also clipped to the index finger of each participant to measure arterial oxygen saturation (SpO_2) during the experiment. Using Y_v and CBF, baseline $CMRO_2 = CBF(Y_a - Y_v)\,C_a$, where Y_a = average arterial O_2 saturation from pulse-oximeter and C_a = 833.7 μmol O2/100 mL blood (*See* Supplemental Fig. 1; Ge et al., 2012; Guyton and Hall, 2005).

Total lesion volume (TLV) was calculated from T_2-FLAIR images with the lesion prediction algorithm in the Lesion Segmentation Toolbox (LST v2.0.15; Schmidt, 2017). The GM, WM, and intracranial volumes (ICV) were estimated from MPRAGE using Freesurfer to estimate brain atrophy as brain parenchymal fraction (BPF = [GM + WM]/ICV[7]).

DTI images were corrected for eddy-current distortions and motion (FSL EDDY tool) and co-registered to MPRAGE. Diffusion Kurtosis Estimator (DKE) software (Tabesh et al., 2011) was used to obtain estimates of tensor parameters, such as fractional anisotropy (FA). All DTI parameter maps were aligned to whole-brain skeletons using Tract-Based Spatial Statistics (TBSS; Smith et al., 2007) yielding metrics of average FA across the normal-appearing white matter (NAWM) skeleton.

3.3. Statistical analysis

Statistical analyses were conducted using SPSS (2017. Version 25.0. Armonk, NY: IBM Corp.). Group demographic comparisons (age, education) were conducted via independent samples t-test ($p < 0.05$) and gender distributions were tested via chi-square test as described in Table 1. Analysis of covariance (ANCOVA) was used to test main effects of group in all other variables of interest with age and education as covariates. Associations (between $CMRO_2$ and MFIS or SDMT) were assessed using Pearson correlations ($p < 0.05$). Hierarchical regression was performed to further evaluate the predictive power of $CMRO_2$ to fatigue and cognitive performance in MS patients beyond disease characteristics (e.g. age, EDSS, TLV, education). Further, MS patients were separated into three groups based on the $CMRO_2$ interquartile range (45.28 μmol O_2/100 g/min) and ANOVA was performed to test main effects of group in all variables of interest between HC and these 3 MS groups. Following significant main effect of group, Tukey's honestly significant difference (HSD) post-hoc tests were performed ($p < 0.05$).

Table 2
Mean (standard deviation) of physiologic parameters from healthy controls (HC) and MS patients (MS patients).

	HC (n = 16)	MS patients (n = 33)	F	p-value
Imaging				
CBF (mL/100 g/min)	64.93 (15.88)	57.03 (13.85)	2.92	0.095
Y_v (%)	62.98 (4.67)	61.87 (4.44)	0.205	0.653
$CMRO_2$ (μmol O2/100 g/min)	**184.59 (34.93)**	**164.32 (31.05)**	**4.425**	**0.041**
BPF (%)	73.60 (6.04)	71.10 (5.71)	1.165	0.286
FA	**0.537 (0.02)**	**0.506 (0.35)**	**7.501**	**0.009**
Neuropsychological				
MFIS	**15.73 (11.75)**	**35.00 (15.82)**	**12.843**	**0.001**
SDMT	58.80 (8.59)	53.34 (8.90)	1.950	0.170
BDI	**3.19 (3.51)**	**8.16 (6.71)**	**6.133**	**0.017**
Physiologic				
HR (beats/min)	71.97 (12.90)	70.88 (8.98)	0.089	0.766
SpO_2 (%)	97.80 (1.74)	97.39 (1.66)	0.393	0.534
Systolic BP (mm Hg)	123.31 (19.13)	123.27 (13.22)	0.151	0.699
Diastolic BP (mm Hg)	74.88 (14.10)	77.18 (9.87)	0.022	0.883

4. Results

Table 1 describes the HC and MS-patient sample characteristics. There were no significant differences between the two groups in age, sex, or education. We evaluated differences between HC and MS patients in the structural and metabolic imaging, neuropsychological, and physiologic measures (see Table 2). After covarying for age and education, MS patients had significantly lower $CMRO_2$ (p = 0.041) and FA from the NAWM skeleton (p = 0.009) compared to HC. MS patients had significantly higher MFIS scores (p = 0.001) and BDI indices (p = 0.017). Gender was matched between groups to minimize gender effects, but all significant differences remained when gender was included as an additional covariate. There were no differences between groups in CBF, Y_v, BPF, SDMT, heart rate, SPO_2 or blood pressure.

To evaluate if the addition of $CMRO_2$ improved the prediction of fatigue and cognitive performance in MS patients, we performed hierarchical multiple regression. For MFIS, a two-block hierarchical regression was performed with age, EDSS, and TLV (Kos et al., 2008) entered in block one and $CMRO_2$ entered in block two. Age, EDSS, and TLV accounted for 18.5% of the variance and the overall model was not significant ($F(3,28)$ = 2.78, p = 0.06). Adding $CMRO_2$ explained an additional 10% of the variance resulting in a significant regression model (p = 0.02) and a significant change in R^2 ($F(1,27)$ = 4.05, p = 0.05, ΔR^2 = 0.10). For SDMT, a two-block hierarchical regression was performed with age, EDSS, TLV, and education (Benedict et al., 2010) entered in block one and $CMRO_2$ entered in block two. Age, EDSS, TLV, and education accounted for 52.2% of the variance with an overall significant regression model ($F(4,26)$ = 7.09, p = 0.001). Adding $CMRO_2$ explained an additional 10.2% of the variance resulting

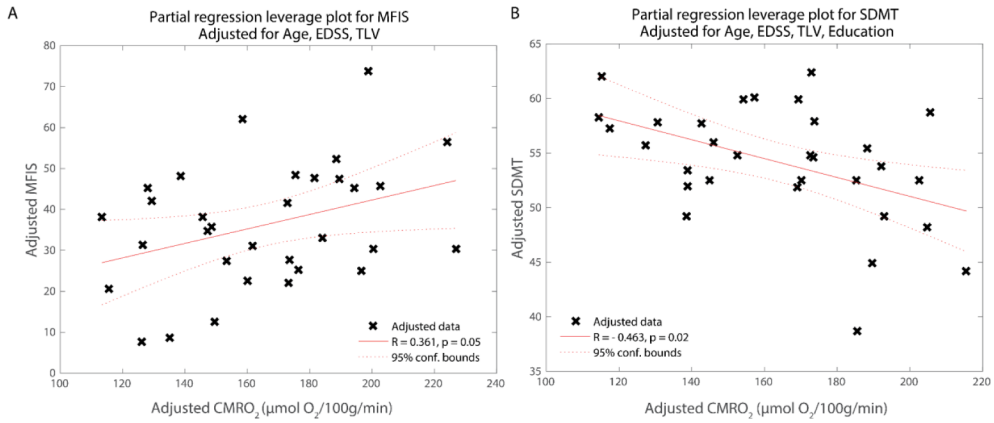

Fig. 1. (Left) Partial linear regression of $CMRO_2$ with MFIS in MS patients after adjusting for age, EDSS, and TLV ($R = 0.361$, $p = 0.05$). (Right) Partial linear regression of $CMRO_2$ with SDMT scores in MS patients after adjusting for age, EDSS, TLV, and education ($R = -0.463$, $p = 0.015$.).

in a significant change in R^2 ($F(1,25) = 6.81$, $p = 0.015$, $\Delta R^2 = 0.102$). Fig. 1A displays the partial regression plot for $CMRO_2$ and MFIS with higher fatigue associated with higher $CMRO_2$ ($r = 0.361$, $p = 0.05$). Fig. 1B shows the negative relationship between $CMRO_2$ and SDMT ($r = -0.463$, $p = 0.015$), with higher $CMRO_2$ resulting in worse performance on SDMT. There were no significant correlations between $CMRO_2$ and EDSS, TLV, Disease Duration, BPF, or FA.

Finally, to evaluate the potential mechanism of white matter damage leading to changes in $CMRO_2$, we split MS patients into three groups based on the $CMRO_2$ interquartile range (45.28 μmol O_2/100 g/min; see Table 3). The first group was MS patients with the lowest 25% $CMRO_2$ ($n = 8$, mean ± std = 124.93 ± 12.05 μmol O_2/100 g/min), the second group was MS patients with the middle 50% $CMRO_2$ ($n = 17$, mean ± std = 165.52 ± 12.97 μmol O_2/100 g/min), and the third group was MS patients with the highest 25% $CMRO_2$ ($n = 8$, mean ± std = 207.44 ± 14.07 μmol O_2/100 g/min). Table 3 displays average values for each MS patient group, p-values reflect a significant main effect of group by ANOVA between HC and all MS groups, and bolded values reflect significant differences from HC via Tukey HSD test. The low $CMRO_2$ MS patient groups had significantly lower CBF ($p = 0.001$) and $CMRO_2$ $p < 0.001$) compared with HC. Fig. 2 displays the average FA for each group with significantly lower FA only in the high $CMRO_2$ MS patients compared with HC ($p = 0.007$). Additionally, the middle and high $CMRO_2$ MS patients had significantly increased fatigue ($p = 0.005$, $p = 0.001$, respectively) and the high $CMRO_2$ MS

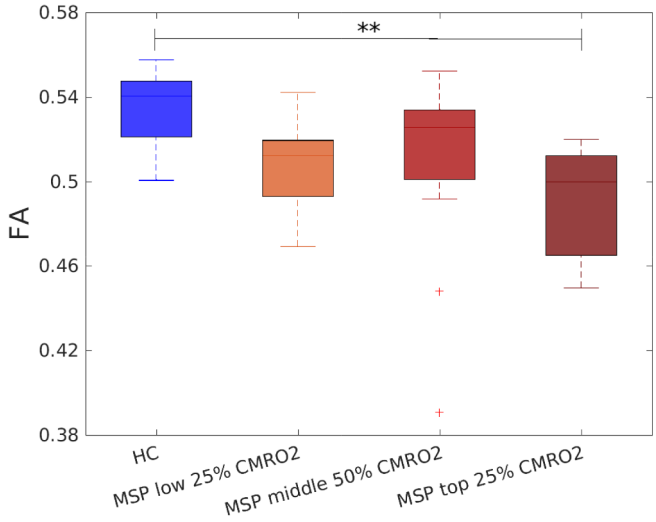

Fig. 2. Box and whisker plot of fractional anisotropy (FA) in (blue) HC, (orange) lowest quartile $CMRO_2$ MS patients, (red) middle 50% $CMRO_2$ MS patients, and (dark red) highest quartile $CMRO_2$ MS patients. ** = $p < 0.01$ from post-hoc Tukey t-test. (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article.)

Table 3
Mean (standard deviation) of physiologic parameters from MS patients (MS patients) based on $CMRO_2$ quartile ranges. p-value reflects ANOVA between HC and MS patient groups with bold text reflecting a significant difference from HC mean (post-hoc Tukey HSD). *$p < 0.05$, **$p < 0.01$, ***$p < 0.001$.

	Low $CMRO_2$ ($n = 8$)	Middle $CMRO_2$ ($n = 17$)	High $CMRO_2$ ($n = 8$)	*ANOVA* p-value
Imaging				
CBF (mL/100 g/min)	**42.29 (7.28)****	58.41 (12.57)	68.83 (7.08)	< 0.001
Y_v (%)	61.73 (4.12)	62.11 (5.24)	61.49 (3.17)	0.86
$CMRO_2$ (μmol O_2/100 g/min)	**124.93 (12.05)*****	165.52 (12.97)	207.44 (14.07)	< 0.001
BPF (%)	70.66 (4.76)	70.10 (4.44)	72.66 (5.45)	0.15
FA	0.508 (0.022)	0.513 (0.040)	**0.490 (0.027)****	**0.01**
Neuropsychological				
MFIS	30.75 (15.40)	**35.31 (17.11)****	**43.50 (17.95)*****	0.001
SDMT	55.38 (6.48)	53.00 (11.05)	52.13 (4.79)	0.23
BDI	7.88 (7.12)	6.71 (5.31)	**13.63 (9.23)****	0.002
Demographics				
Age (years)	49.50 (8.19)	46.53 (11.44)	43.50 (9.96)	0.43
Education (years)	16.25 (2.49)	16.71 (2.82)	15.14 (1.95)	0.39
Disease Duration (years)	11.38 (10.25)	10.00 (8.19)	5.88 (4.55)	0.36[a]
TLV (mL)	6.35 (8.67)	5.32 (6.27)	4.06 (2.46)	0.77[a]
EDSS	2.25 (0.65)	2.78 (2.29)	2.50 (1.34)	0.56[a]

[a] ANOVA between MS patients only.

patients had significantly increased depression ($p = 0.001$) compared to HC. There were no significant differences in any other parameters (*see* Table 3) but the high $CMRO_2$ MS patients had higher BPF and had lower disease duration, age, and TLV compared to low and middle $CMRO_2$ MS patients.

5. Discussion

The present study utilized MRI to non-invasively measure whole-brain $CMRO_2$ in HC and MS patients and investigated the relationship between $CMRO_2$, fatigue, and cognitive performance. Our study yielded 3 main findings. First, MS patients have lower $CMRO_2$ compared to HC, confirming previous cerebral hypometabolism findings. Second, after controlling for demographic and MS disease characteristics, $CMRO_2$ predicts increased fatigue, via the MFIS scale, and reduced cognitive performance via SDMT. Finally, MS patients with higher $CMRO_2$ have reduced FA in normal-appearing white matter. Altogether, these results suggest that increased $CMRO_2$ reflects ongoing demyelination and autoimmune activity that plays an important role in both fatigue and cognitive dysfunction. Thus, they have important implications for the clinical utility of $CMRO_2$ monitoring in disease management.

Our results display generalized cerebral hypometabolism in MS patients, consistent with previous work in MS patients utilizing PET (D'haeseleer et al., 2015; Schiepers et al., 1997; Sun et al., 1998) and MRI (Ge et al., 2012). While our study replicates cerebral hypometabolism in MS patients compared to HC, possibly due to overall atrophy, there is a wide range of $CMRO_2$ amongst MS patients (e.g., 110.14–230.91 μmol O_2/100 g/min; Ge et al., 2012; Schiepers et al., 1997). One possible explanation for this variability is that it reflects sub-clinical, immune-mediated disease activity versus stable disease. For example, Schiepers et al observed significant reductions in *N*-acetylaspartate (NAA) in normal-appearing white matter (NAWM) and hypermetabolism in stable MS patients with lesions < 1.5 mL. These NAWM alterations suggest active inflammatory disease with dispersed neuronal damage that is not yet detectable via MRI. Additionally, they observed hypometabolism and normal NAA in NAWM of MS patients with apparent lesions, suggesting more stable disease potentially after functional neuronal recovery. Further, pathological studies have suggested that inflammation and demyelination lead to increased sodium channel density, probably necessitating increased oxygen metabolism (Paling et al., 2011). Our results support this hypothesis insofar as high $CMRO_2$ MS patients exhibit significantly lower FA in NAWM (i.e., diffuse neuronal damage) and the lowest TLV. Altogether, these findings suggest that $CMRO_2$ may reflect increased oxygen metabolism due to sub-clinical inflammatory and demyelinating disease states.

In the present study, we observed a positive relationship between measures of fatigue and $CMRO_2$ in MS patients independent of age, EDSS, and TLV. Together with the diffusion imaging results described above, fatigue may be driven by metabolically active processes of lesion repair and cortical reorganization (Reddy et al., 2000; Tartaglia et al., 2004). Myelin biosynthesis (i.e., remyelination) requires metabolically demanding ATP production via mitochondrial oxidative phosphorylation (i.e., high $CMRO_2$; Sivakolundu et al., 2019a). Thus, the capability for reorganization and repair may lead to increased $CMRO_2$ and could induce fatigue in these patients. While this hypothesis effectively describes those patients with the highest fatigue scores and $CMRO_2$, many MS patients have elevated fatigue compared to HC. In these MS patients, fatigue may be driven by chronic CNS dysfunction due to atrophy and the perception of increased effort to perform a task (Bakshi, 2003). While more work is certainly necessary, $CMRO_2$ can add significant information about fatigue pathology and provide a potential technique to monitor fatigue physiology in MS.

In addition to fatigue, we observed a relationship between increased $CMRO_2$ and reduced cognitive performance in MS patients beyond age, education, EDSS, or TLV. Previous work has also shown reduced cognitive performance on SDMT related to acute relapses in MS patients (Benedict et al., 2014). However, other work has shown a relationship between reduced $CMRO_2$ and cognitive decline (Sun et al., 1998) associated with cerebral atrophy (Benedict et al., 2004). In MS, atrophy and inflammation are two key players; thus, without broad atrophy, an active inflammatory response could explain these oxygen metabolism differences. Because the high $CMRO_2$ MS patients do not display significant atrophy (see Table 2), increased oxygen metabolism might reflect immune-mediated disease activity more than atrophy. Additionally, as a whole, our MS patients did not exhibit significant cognitive decline, possibly due to the protective effects of education (Benedict et al., 2010).

Altogether, our results suggest a physiologic mechanism of $CMRO_2$ as a resource affording cognitive abilities (Scholey et al., 1999). For example, in a healthy system, during periods of cognitive demand, physiologic responses lead to increased oxygen delivery and metabolism in neural tissue, allowing efficient cognitive performance (Attwell et al., 2010; Cauli and Hamel, 2010). However, in MS patients with ongoing inflammation, lesion repair, and/or cortical reorganization, this resource of increased oxygen delivery and metabolism would be consumed and thus unavailable during cognitive demand, leading to dysfunction. Additionally, we did not observe any association between fatigue and cognitive performance, that might reflect a dissociation between perceived fatigue and cognitive impairment (Krupp and Elkins, 2000). Overall, our results suggest that $CMRO_2$ can provide further insight into the pathophysiology of cognitive dysfunction beyond structural changes and provide further monitoring in MS patients.

As a measure of oxygen utilization, $CMRO_2$ may provide a more sophisticated assessment of disease processes (e.g., virtual hypoxia, energy failure) and the ability of currently approved and future disease-modifying therapies (DMT) to address these processes. For example, with the addition of measures of regional perfusion and spectroscopic metabolites, $CMRO_2$ could provide a comprehensive picture of oxygen supply and demand with or without DMT (Paling et al., 2011). While, more work is certainly necessary to understand $CMRO_2$ changes in MS, the ability to monitor patients' inflammatory activity (e.g., indexed by $CMRO_2$) in response to therapy, especially early in the disease course, would provide valuable therapeutic information. Future studies should longitudinally follow patients before, during, and post-relapse and throughout their disease course to validate the inflammation-biomarker potential of $CMRO_2$ and assess its relationships to fatigue and cognitive impairment.

Compared to other techniques (e.g., PET, calibrated fMRI) the present technique is limited to whole brain metrics without regional data. Thus, we cannot delineate changes in $CMRO_2$ specific to gray matter, white matter, or lesions. In contrast, the present technique is much faster (~90 s) and simpler to perform. We utilized the superior sagittal sinus to interpret global measures of CBF and $CMRO_2$ which does not incorporate all draining veins of the brain. However, we corrected for this and observed similar CBF and $CMRO_2$ compared to previous studies using the four feeding arteries to measure CBF (De Vis et al., 2018; Ge et al., 2012). We were also limited by the use of self-reports to assess fatigue, which results in subjective metrics that may differ between patients based on personal perceptions of fatigue.

6. Conclusion

Overall, this study displays the potential for these quick MRI scans to monitor global $CMRO_2$ non-invasively. MS patients overall exhibit reduced $CMRO_2$ compared to HC. Higher $CMRO_2$ in MS patients predicts increased fatigue, worse cognitive performance, and reduced FA. While more work is certainly necessary, these results support the hypothesis that increased $CMRO_2$ may reflect on-going sub-clinical disease activity leading to MS-related symptomology. These simple, brief (e.g., 90 s) resting scans used to measure $CMRO_2$ could provide insight into the pathophysiology of fatigue and cognition, distinguish inflammatory from stable disease, and provide outcome measures for

symptomatic therapies.

7. Data availability

The datasets generated and analyzed during this study are available from the corresponding author upon request.

CRediT authorship contribution statement

Kl. West: Conceptualization, Methodology, Formal analysis, Investigation, Data curation, Writing - original draft. **Dk. Sivakolundu:** Conceptualization, Methodology, Investigation, Data curation, Writing - review & editing. **Gb. Maruthy:** Methodology, Investigation, Data curation, Writing - review & editing. **Md. Zuppichini:** Methodology, Investigation, Data curation, Writing - review & editing. **P. Liu:** Software, Resources, Writing - review & editing. **Bp. Thomas:** Software, Resources, Writing - review & editing. **Js. Spence:** Formal analysis, Writing - review & editing. **H. Lu:** Conceptualization, Methodology, Supervision, Funding acquisition. **Dt. Okuda:** Conceptualization, Methodology, Supervision, Funding acquisition. **B. Rypma:** Conceptualization, Methodology, Supervision, Funding acquisition.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.nicl.2020.102281.

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