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Gray matter SWI-filtered phase and atrophy are linked to disability in MS

Jesper Hagemeier[1], Bianca Weinstock-Guttman[2], Mari Heininen-Brown[1], Guy U. Poloni[1], Niels Bergsland[1], Claudiu Schirda[1], Christopher R. Magnano[1], Cheryl Kennedy[1], Ellen Carl[1], Michael G. Dwyer[1], Alireza Minagar[3], Robert Zivadinov[1,2]

[1]Buffalo Neuroimaging Analysis Center, University at Buffalo, Buffalo, NY, USA [2]The Jacobs Neurological Institute, University at Buffalo, Buffalo, NY, USA, [3]Department of Neurology, Louisiana State University Health Sciences Center, Shreveport, LA

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1. ABSTRACT

The association between clinical outcomes and abnormal susceptibility-weighted imaging (SWI)-filtered phase, indicative of increased iron content, as well as atrophy, was investigated in the subcortical deep-gray matter (SDGM) of multiple sclerosis (MS) patients. 149 relapsing-remitting (RR) and 61 secondary-progressive (SP) MS patients underwent SWI on a 3T scanner. Mean phase of the abnormal phase tissue (MP-APT) and normalized volumes were determined for the total and region-specific SDGM structures. In an age- and gender-adjusted regression model, total SDGM volume was the strongest predictor of Expanded Disability Status Scale (EDSS) (beta = -.224, p <.001), followed by total SDGM MP-APT (beta = -.168, p <.019). This model accounted for 30.4% of the variance in EDSS. Only SDGM MP-APT added additional variance in predicting EDSS, compared to conventional MRI metrics. Caudate and red nucleus MP-APT and amygdala volume were associated with EDSS. Our findings suggest that disability in MS patients is associated better with SDGM pathology, as indicated by increased iron content and atrophy, than with lesion burden or white matter and cortical volumes

2. INTRODUCTION

Although multiple sclerosis (MS) is considered historically as a white matter (WM) disease, abnormalities in the gray matter (GM) are consistently reported in both the cerebral cortex and subcortical deep GM (SDGM) brain structures. (1) Mounting evidence suggests that GM pathology may play a more important role in predicting clinical outcomes in MS patients than WM damage. (2, 3)

Over the last decade, efforts have been made to develop novel MRI techniques that are able to quantitatively measure a wide spectrum of GM pathology. Histopathological and MRI studies have found increased iron deposition in the SDGM of individuals with any of several neurodegenerative disorders, including MS. (4-8) Studies have shown a relationship between increased level of iron content and clinical progression, cognitive impairment, and brain atrophy in MS patients. (5, 9-12) The underlying pathological mechanisms of iron deposition in MS patients are unknown; however, it is thought that iron may be derived from myelin/oligodendrocyte debris, destroyed macrophages, or it can be the product of hemorrhaging from damaged brain vessels. (8, 13, 14)

The most frequent approach for visualizing iron deposition *in vivo* is to measure the level of hypointensities on T2-weighted imaging (WI) in SDGM. It is thought that increased iron content causes shortening of T2-relaxation time. (5, 7, 15) Studies using a T2 hypointensity approach have found increased iron content in SDGM areas, such as the caudate, thalamus, putamen, globus pallidus, dentate and red nucleus, and an association between elevated levels of iron with disease duration, disability and cognitive dysfunction. (5, 10, 11, 16, 17) However, the main drawback of T2-based approaches is insufficient sensitivity to detect changes in paramagnetic substances and water content. (8, 9, 14)

Paramagnetic substances such as deoxyhemoglobin, ferritin and hemosiderin have the ability to change the local magnetic field, thereby influencing the frequency, or phase, of proton spin. (7) By using the phase component of MRI acquisition, valuable information is gained from the contents of paramagnetic substances in voxels. Tissues differ in their susceptibility to phase effects, giving rise to a new form of contrast enhancement. Recently, susceptibility-weighted imaging (SWI) has been used to identify the characteristics of iron depositions in MS patients. (18-21) The SWI-filtered phase approach (7, 21) employs a complex-space high-pass filter in order to retain only localized phase shifts – the type most likely caused by iron deposition. (22)

In this study, we used the SWI-filtered phase and atrophy approach to investigate characteristics of SDGM pathology in large cohort of MS patients. We sought to investigate the relationship between abnormal SDGM phase, indicative of increased iron content, and atrophy, with clinical outcomes. We aimed to determine whether SDGM pathology, as determined by abnormal phase and atrophy can add independent variance in predicting disability, compared to lesion burden or white matter and cortical volumes.

3. METHODS

3.1. Subjects

Two-hundred ten (210) consecutive MS patients ([149] relapsing-remitting [RR] and 61 secondary-progressive [SP]) were consecutively enrolled. Subjects underwent full neurological assessment, including determination of the Expanded Disability Status Scale (EDSS) status. (23) Inclusion criteria were: age 18-65 years, RR or SPMS disease course, (24) and EDSS between 0 and 6.5. Participants were excluded if they had a relapse or were treated with steroids within the month preceding study entry, were pregnant, or had any pre-existing medical conditions known to be associated with brain pathology (i.e., cerebrovascular disease).

This study was approved by the internal Institutional Review Board and written informed consent was obtained from all participants.

3.2. Image acquisition

Subjects were examined using a 3T GE Signa Excite HD 12.0 Twin Speed 8-channel scanner (General Electric, Milwaukee, WI). SWI data was collected using a 3-Dimensional (3D) flow-compensated GRE (Gradient Recalled Echo) sequence with the following parameters: 64 locs/slab, 2mm thick, a 512x192 matrix, FOV=25.6cmx 19.2cm (512x256 matrix with PhaseFOV=0.75), for an in-plane resolution of 0.5mm x 1mm. Flip angle, echo-, and repetition times were: FA=12, TE=22ms, and TR=40ms, for a total acquisition time of AT=8:46. Raw (k-space) data was transferred to an offline Linux workstation for post-processing using in-house developed software written in Matlab (MathWorks Inc., Natick, MA, USA).

Additionally, the following sequences were acquired: 2D multi-planar dual fast spin-echo (FSE) proton density (PD) T2-WI (TE1/TE2/TR=9/98/5300ms; FA=90; ETL=14); Fluid-Attenuated Inversion-Recovery (FLAIR) (TE/TI/TR=120/2 100/8 500 ms; FA=90; ETL=24); 3D high resolution (HIRES) T1-WI using a fast spoiled gradient echo (FSPGR) with magnetization-prepared inversion recovery (IR) pulse (TE/TI/TR=2.8/900/5.9 ms, FA=10); and spin echo (SE) T1-WI (TE/TR=16/600 ms, FA=90). In plane resolution of all scans, with the exception of SWI, was 1mm x 1mm. For All 2D scans (PD/T2, FLAIR and SE T1), 48 slices were collected, with a thickness of 3mm, and no gap between slices.

3.3. Image analyses

Analyses were performed by operators who were unaware of the participants' disease subtypes. SDGM structures were segmented using a combination of semi-automated edge-contouring and FMRIB's fMRI integrated registration and segmentation tool (FIRST) on 3D T1-WI. (25) Specifically, the thalamus, caudate, putamen, globus pallidus, hippocampus, amygdala, and nucleus accumbens were identified in this way. (26) Structures not identifiable by FIRST, such as the red nucleus, pulvinar nucleus of the thalamus, and substantia nigra, were identified semi-automatically using JIM5 (Xinapse Systems Ltd., Northamptonshire, UK) on the most representative slice for each subject. (26)

A detailed overview of SWI processing, analysis approach and validation is provided elsewhere. (21) Abnormal phase was defined as a measure of the degree of phase decrease for voxels with mean phase values at least two standard deviations below the mean value found in the same regions of healthy control subjects, as previously described. (21) Mean phase values of sub-threshold voxels indicative of abnormal iron content was calculated to yield the mean phase of abnormal phase tissue (MP-APT). Lower (more negative) MP-APT values represent increased iron content within a region. Mean values are presented in radians.

Global and regional brain atrophy was determined with corrections for T1-hypointensity misclassification using an in-house developed in-painting program, on the 3D T1 weighted scans, as described previously. (27) Normalized cortical volume (NCV) and normalized white-matter volume (NWMV) were calculated with SIENAX version 2.6. (28) Normalized volumes were obtained for all SDGM structures with FIRST. (26)

T2 hyperintense and T1 hypointense lesion volumes (LVs) were outlined using a semi-automated edge-detection contouring-thresholding technique. (29)

3.4. Statistical analysis

Analyses were conducted using PASW Statistics version 18.0.2 (IBM Corp., Somers, NY, USA). Differences between disease types on demographic, clinical and conventional MRI characteristics were assessed using the chi-square test, Student's *t*-test and Mann-Whitney U test, as appropriate.

Comparisons between RR and SPMS patients on SDGM MP-APT and normalized volumes were performed using the Mann-Whitney U test.

To decrease the number of variables and amount of multiple comparisons in subsequent analyses, SDGM structures were consolidated into the total SDGM variable. To determine the relationship between MRI measures and clinical outcomes (EDSS and disease duration), we employed Spearman's rank correlation analyses. MRI measures that significantly related to clinical outcome measures in univariate analyses ($p<.05$) were added to a backward stepwise regression model. This was done to determine the relative predictive value of the variables on clinical outcomes. All models were adjusted for age and gender, and repeated for RR and SPMS patients separately. Additionally, predictor variables were tested for multi-collinearity. T1- and T2-LVs were strongly correlated (Spearman rho=.84). However, both variables were retained in all models, because the variance inflation factor, a measure of multi-collinearity, remained < 4 in all regression models. Furthermore, T1- and T2-LVs were not normally distributed as determined by the Shapiro-Wilks test ($p<.001$) and were, therefore, logarithmically transformed to improve normality. Variables were stepwise-excluded until remaining variables displayed significance at the $p<.1$ level.

In additional backward stepwise regression analyses, conventional MRI measures (T1-LV, T2-LV, NCV, and NWMV) were forced into the model, whereas SDGM MP-APT and normalized volumes could be excluded readily if non-significant. The purpose of this model was to test whether SDGM MP-APT and normalized volumes could significantly improve on a model containing all conventional MR measures, thereby explaining additional variance compared to those conventional measures. Similar to the aforementioned regression models, age and gender were added as covariates. Subsequently, analyses were repeated separately for RR and SPMS groups.

To investigate, in an exploratory fashion, which of the SDGM structures could best predict clinical outcome, we carried out additional stepwise regression models, entering all SDGM structures as predictor variables while including age and gender as covariates.

All findings were tested using two-tailed tests.

4. RESULTS

4.1. Demographic and clinical characteristics

As shown in Table 1, RR and SPMS patients had an equal proportion of males to females ($p=.93$), and age at onset ($p=.286$). Median EDSS was 3.0 and mean disease duration and age were 14 and 47 years, respectively. Females accounted for the majority of subjects (71%). As expected, SP patients were older, had longer disease duration and higher EDSS ($p<.0001$). Lesion volumes (T1-LV, T2-LV) were higher among SP patients ($p<.01$). In addition, progressive MS patients had significantly decreased NCV ($p<.001$), but not NWMV ($p=.052$). Of the 210 MS patients, 179 (85.6%) were on disease-modifying therapy.

4.2 Differences between disease subtypes

Increased MP-APT was observed in SP patients in total SDGM ($p<.001$), putamen ($p<.001$), caudate ($p=.002$) and the red nucleus ($p=.01$), as shown in Table 2. Decreased volumes in several SDGM structures were noticeably more extensive in progressive MS patients (Table 2). In particular, the caudate, thalamus, pulvinar nucleus of thalamus, amygdala, nucleus accumbens, and hippocampus were all significantly lower in SP patients at the $p<.01$ level.

4.3. Relationship between MRI measures and clinical outcomes

Significant relationships were found in MS patients between the total SDGM MP-APT and volume, NCV, NWMV, T2- and T1-LVs ($p<.01$) with EDSS and disease duration. Correlations were stronger for RR compared to SPMS patients.

4.4. Regression analyses

Backward stepwise regression analysis showed that, after adjusting for age and gender, none of the MRI variables were included in the model, and could predict disease duration (Table 3). Total SDGM volume was the strongest predictor of EDSS (beta = -.224, p <.001), followed by total SDGM MP-APT (beta = -.168, p <.019). This model accounted for 30.4% (adjusted $R^2 = .304$) of the variance in EDSS. Subsequent analyses according to disease type showed that in RR patients, total SDGM MP-APT was retained in the model predicting disease duration. Total SDGM volume, MP-APT and NCV were included in the model predicting EDSS. However, in SP patients none of the MRI measures were preserved.

Additional backward stepwise regression analyses were performed to determine whether total SDGM MP-APT or volume can explain additional variance in predicting clinical outcomes when compared to conventional MRI measures (Table 4). T1- and T2-LVs, NCV and NWMV were forced in the model regardless of significance, whereas the total SDGM variables were only included if significant at the $p<.1$ level. In predicting disease duration, neither total SDGM MP-APT nor volume were retained in the model. However, SDGM MP-APT was retained in the model predicting EDSS (beta = -.175, p = .016). The addition of this variable to the model caused an

Table 1. Demographic, clinical and conventional MRI characteristics in MS patients

	MS (n=210)	**RR (n=149)**	**SP (n=61)**	***p*-value**
Females, n (%)	149 (71%)	106 (71.1%)	43 (70.5%)	.93
Age in years, mean (SD)	47.1 (10.3)	43.9 (9.5)	54.9 (7.9)	<.001
Age of onset, mean (SD)	32.6 (9.2)	32.1 (8.8)	33.6 (10.3)	.286
Disease duration in years, mean (SD)	14.5 (9.8)	11.7 (7.5)	21.2 (11.5)	<.001
Expanded Disability Status Scale, mean (SD) median	3.5 (2.1) 3	2.6 (1.6) 2	5.6 (1.5) 6	<.001
T2 lesion volume, mean (SD)	17.2 (18.4)	15 (16.5)	22.5 (21.5)	.004
T1 lesion volume, mean (SD)	3.3 (6.6)	2.3 (4)	5.5 (10)	.003
Normalized cortical volume, mean (SD)	660 (55.9)	671.7 (54.6)	631.7 (48.7)	<.001
Normalized white matter volume, mean (SD)	738 (58)	743.8 (58.5)	722.8 (54.7)	.052

Abbreviations: MS: multiple sclerosis; RR: relapsing-remitting; SP: secondary progressive; SD: standard deviation. All lesion and brain volumes are expressed in milliliters. Differences between RR and SPMS groups were tested using the chi-square test, Student's *t*-test and the Mann-Whitney U test, as appropriate

Table 2. Comparison of SDGM MP-APT and volume structures between RR and SPMS patients

	MP-APT			**SDGM volume**		
	RR (n=149)	**SP (n-61)**	***p*-value**	**RR (n=149)**	**SP (n-61)**	***p*-value**
Total SDGM	-.156 (.02)	-.172 (.031)	< .001	42.71 (4.89)	39.74 (5.15)	< .001
Caudate	-.178 (.019)	-.186 (.02)	.002	6.57 (1.01)	6.08 (.84)	.002
Putamen	-.186 (.032)	-.209 (.044)	< .001	9.02 (1.27)	8.45 (1.31)	.012
Globus Pallidus	-.196 (.035)	-.204 (.038)	.178	3.29 (.49)	3.1 (.58)	.034
Thalamus	-.099 (.014)	-.102 (.018)	.662	13.93 (1.7)	12.85 (1.89)	<.001
Pulvinar nucleus of thalamus	-.157 (.03)	-.152 (.035)	.095	.37 (.12)	.33 (09)	.004
Amygdala	-.207 (.064)	-.242 (.117)	.099	2.34 (.36)	2.16 (.45)	.009
Nucleus Accumbens	-.75 (.226)	-.809 (.282)	.242	.74 (.21)	.63 (.18)	.001
Red Nucleus	-.238 (.033)	-.252 (.038)	.010	.17 (.05)	.16 (.03)	.029
Substania Nigra	-.322 (.039)	-.328 (.042)	.331	.29 (.09)	.27 (.04)	.013
Hippocampus	-.159 (.036)	-.175 (.054)	.149	6.79 (.81)	6.44 (.83)	.003

Abbreviations: MP-APT: mean phase of the abnormal phase tissue; SDGM: subcortical deep gray matter; RR: relapsing-remitting; SP: secondary progressive. MP-APT is expressed in radians. All normalized deep GM volumes are expressed in milliliters. Differences between groups were assessed using the Mann-Whitney U test

Table 3. Backward stepwise regression between MRI variables and clinical outcomes

	Disease duration[1]		**EDSS[2]**	
Variable	Beta	*p*-value	Beta	*p*-value
Age	.577	< .001	.370	<.001
Gender	---	---	---	---
Total SDGM MP-APT	---	---	-.168	.019
Total SDGM volume	---	---	-.224	.001
T2-LV	---	---	---	---
T1-LV	---	---	---	---
NCV	---	---	---	---
NWMV	---	---	---	---

Abbreviations: EDSS: Expanded Disability Status Scale; SDGM: subcortical deep gray matter; MP-APT: mean phase of the abnormal phase tissue; LV: lesion volume; NCV: normalized cortical volume; NWMV: normalized white matter volume. Effect sizes representing the variance explained by the outcome variables: [1] adjusted R^2 = .329, [2] adjusted R^2 = .304. The models were adjusted for age and gender

Table 4. Backward stepwise regression between total SDGM variables and clinical outcomes, when conventional MRI measures are added to the model

	Disease duration[1]		**EDSS[2]**	
Variable	Beta	*p*-value	Beta	*p*-value
Age	.562	< .001	.297	< .001
Gender	---	---	---	---
Total SDGM MP-APT	---	---	-.175	.016
Total SDGM volume	---	---	---	---
T2-LV	.144	.241	.019	.883
T1-LV	-.138	.269	.124	.334
NCV	.007	.928	-.133	.110
NWMV	-.102	.146	-.066	.356

Abbreviations: EDSS: expanded disability status scale; SDGM: subcortical deep gray matter; MP-APT: mean phase of the abnormal phase tissue; LV: lesion volume; NCV: normalized cortical volume; NWMV: normalized white matter volume. Effect sizes representing the variance explained by the outcome variables: [1] adjusted R^2 = .328, [2] adjusted R^2 = .326, Increase in effect size compared to models including only conventional MRI measures: [1] adjusted R^2 increase = ---, [2] adjusted R^2 increase = .049. The models were adjusted for age and gender

Table 5. Backward stepwise regression between region-specific SDGM structures and clinical outcomes

Variable	Measurement	Disease duration[1]		EDSS[2]	
		Beta	*p*-value	Beta	*p*-value
Age		.522	< .001	.450	< .001
Amygdala	MP-APT	-.142	.012	---	---
Thalamus	Volume	-.130	.027	---	---
Caudate	MP-APT	---	---	-.133	.026
Amygdala	Volume	---	---	-.270	< .001
Red nucleus	MP-APT	---	---	-.131	.023

Abbreviations: EDSS: Expanded Disability Status Scale; MP-APT: mean phase of abnormal phase tissue. Effect sizes representing the variance explained by the outcome variables: [1] adjusted R^2 = .371, [1] adjusted R^2 = .354, The models were adjusted for age and gender

increase in the explained variance of 4.9% (adjusted R^2 = .049) for EDSS. Analyses were repeated for disease subtypes separately. In RR patients, both variables were retained in predicting disease duration, with total SDGM volume being the strongest predictor. Total SDGM volume was the only MRI variable included in the model predicting EDSS. Neither total SDGM MP-APT nor volume was retained either model predicting the clinical outcome among SP patients.

Additional analyses shed more light on which SDGM structures predicted clinical outcomes (Table 5). After adjusting for age and gender, amygdala (beta = -142, p = .012) MP-APT and thalamus (beta = -.130, p = .027) volumes were able to predict disease duration, while caudate (beta = -.133, p = .026) and red nucleus (beta = -.131, p = .023) MP-APT, and amygdala (beta = -.270, p < .001) volumes significantly predicted EDSS.

5. DISCUSSION

In this study, we investigated how the pathology of SDGM, as measured by two different MRI techniques, is associated with clinical outcomes in a large cohort of RR and SPMS patients. Using SWI-filtered phase data, we computed the MP-APT as a biomarker of abnormal iron content. (21) One of the key findings of this study is that SDGM pathology, as measured by increased abnormal phase and volume, was better associated with disability, as measured by EDSS, than conventional MRI metrics. Furthermore, MP-APT explained additional variance in predicting EDSS, compared to conventional MRI measures. We showed that both the MP-APT and volume of various SDGM structures were significantly lower in SP than in RRMS patients, indicating that apparent increased iron content and advanced atrophy occur at a faster rate in progressive MS patients.

Increased iron deposition has been previously reported in SDGM of MS patients by using histopathological techniques, (4, 6) as well as by using iron-sensitive MRI sequences like T2 relaxometry, (12) T2 hypointensity, (10, 16, 30) magnetic field correlation (31) and SWI. (18-21) More severe atrophy of the SDGM structures was also consistently found in MS patients. (26, 32-36)

However, to the best of our knowledge, this is one of the first studies performed with a large cohort of MS patients that investigated how SDGM pathology is related to clinical outcomes, when adjusting for other cortical and WM volumes, and lesion burden measures. Moreover, we are not aware of a study that used both iron measures and atrophy of SDGM to explore a relationship with clinical outcomes. Our results indicate that SDGM abnormal phase and decreased volume explained physical disability, as measured by EDSS, better than WM or cortical volumes, as well as T2- or T1-LVs. Previous studies have mostly emphasized the importance of global or cortical GM atrophy in MS. (2, 3) However, our findings suggest that even though cortical GM damage appears important, SDGM pathology may be even more relevant for the clinical manifestation of MS. It is important to note that SDGM MP-APT was able to explain additional variance in EDSS, over and above conventional lesion burden and GM or WM atrophy measures (Table 4). This suggests that abnormal phase, indicative of increased iron content, in SDGM is clinically relevant. Therefore, measurement of iron deposition in SDGM structures of MS patients on SWI-filtered phase images may become an important biomarker for monitoring of the disease process and therapeutic responses. (21) Abnormal phase, as determined using the novel SWI-filtered phase imaging technique, can potentially capture iron content and predict the extent of SDGM atrophy more accurately than other available imaging techniques such as T2 hypointensity, which have several drawbacks, such as being sensitive to a range of tissue changes other than iron. (7, 21)

Additional stepwise regression analyses were carried out to explore which specific SDGM structures may contribute to more severe clinical outcomes. These analyses showed that both MP-APT and volume measures of various SDGM structures were retained by the model and could predict disease duration and EDSS (Table 5). The caudate, thalamus, red nucleus and amygdala accounted for most of the variance. Damage and iron deposition of these SDGM structures have often been observed in MS, and appear to be related to fatigue, disease duration, physical disability, and cognitive and memory impairment. (5, 10, 11, 16, 17, 37, 38) However, it remains unknown to which extent associations with clinical outcomes are causal.

Whether iron deposition is an epiphenomenon of the MS disease process or may play a primary role in triggering inflammation and disease development remains unclear at this time, and should be studied at the early stages of disease pathogenesis. (39) One hypothesis postulates that iron accumulation in the brain can lead to neurodegeneration by the formation of toxic free radicals,

resulting in oxidative stress. (8, 14, 40) Another plausible explanation is that iron is merely a marker of past breakdown of myelin and oligodendrocytes, which are known to have high iron concentrations. (41) Demyelination and axonal loss are known to be most extensive in the progressive stages of MS, which is in line with our findings, as we found significantly increased SDGM abnormal phase and decreased volumes in SP compared to RRMS patients.

Brain atrophy, especially GM atrophy, correlates better with physical disability than conventional MRI measures. (1-3) Our findings support the view that GM atrophy is more associated with physical disability than lesion measures. (1) However, by separating SDGM from cortical GM we were able to better examine the contribution of GM damage to physical disability. In fact, this is one of the first studies showing that SDGM pathology is associated more closely with physical disability than lesion or cortical volume measurements. Nevertheless, use of more specific MRI sequences, like double inversion recovery, can increase the ability to capture part of lesion GM pathology *in vivo*. (1) The NWMV was not retained in regression models, confirming findings from previous studies. (2, 3)

A potential limitation is in the identification of the SDGM structures. We employed both FIRST and JIM5 to segment these structures, as has been previously reported and validated. (21) However, using two automated and semi-automated approach of segmenting the SDGM may influence data heterogeneity with respect to the precision of volume measurements in relation to red nucleus, substantia nigra and the pulvinar nucleus of thalamus. The previous study showed similar reproducibility between the FIRST and JIM5 segmented SDGM structures. (21) As expected, more severe SDGM abnormal phase and atrophy were found in the SP group, but more robust association models with clinical outcomes were detected in RRMS patients. Age and sex were included as confounders in all regression models. This result could be explained by the fact that in progressive MS patients more severe SDGM abnormal phase and atrophy are more uniformly distributed, given little regression variation with respect to clinical outcomes.

In conclusion, our results showed that, in a large sample of RR and SPMS patients, SDGM pathology, as measured by increased iron deposition and more advanced atrophy, explains physical disability better than conventional MRI measures. Further longitudinal studies are warranted to determine the value of SDGM pathology, as a potential biomarker in MS clinical trials.

6. ACKNOWLEDGMENTS

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Abbreviations: MS: multiple sclerosis; WM: white matter; GM: gray matter; SDGM: subcortical deep gray matter; WI: weighted imaging; SWI: susceptibility weighted imaging; EDSS: expanded disability status scale; 3D: 3-Dimensional; GRE: gradient recalled echo; FSE: fast spin-echo; PD: proton density; FLAIR: Fluid-Attenuated Inversion-Recovery;, HIRES: high resolution; FSPGR: fast spoiled gradient echo; IR: inversion recovery; SE: spin echo; FIRST: fMRI integrated registration and segmentation tool; MP-APT: mean phase of abnormal phase tissue; NCV: normalized cortical volume; NWMV: normalized white-matter volume; LV: lesion volume

Key Words: Multiple sclerosis, Abnormal phase, Iron deposition, Susceptibility-weighted imaging filtered phase, Brain atrophy, Disability

Send correspondence to: Robert Zivadinov, Department of Neurology, School of Medicine and Biomedical Sciences, Buffalo Neuroimaging Analysis Center, 100 High Street, Buffalo, NY 14203, USA, Tel: 716 859 7031, Fax: 716 859 4005, E-mail: rzivadinov@bnac.net