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Effect of Evobrutinib on Slowly Expanding Lesion Volume in Relapsing Multiple Sclerosis

A Post Hoc Analysis of a Phase 2 Trial

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Abstract

Background and Objectives

Chronic active lesions (CALs) are demyelinated multiple sclerosis (MS) lesions with ongoing microglia/macrophage activity, resulting in irreversible neuronal damage and axonal loss. Evobrutinib is a highly selective, covalent, CNS-penetrant, Bruton tyrosine kinase inhibitor. This post hoc analysis evaluated the effect of evobrutinib on slowly expanding lesion (SEL) volume, an MRI marker of CALs, assessed baseline–week 48 in a phase 2, double-blind, randomized trial (NCT02975349) in relapsing MS (RMS).

Methods

In the 48-week, double-blind trial, adult patients received evobrutinib (25 mg once daily [QD], 75 mg QD, or 75 mg twice daily [BID]), placebo (switched to evobrutinib 25 mg QD after week 24), or open-label dimethyl fumarate (DMF) 240 mg BID. SELs were defined as slowly and consistently radially expanding areas of preexisting T2 lesions of ≥10 contiguous voxels (~30 mm^3) over time. SELs were identified by MRI and assessed by the Jacobian determinant of the nonlinear deformation from baseline to week 48. SEL volume analysis, stratified by baseline T2 lesion volume tertiles, was based on week 48/end-of-treatment status (completers/non-completers). Treatment effect was analyzed using the stratified Hodges-Lehmann estimate of shift in distribution and stratified Wilcoxon rank-sum test. Comparisons of evobrutinib and DMF vs placebo/evobrutinib 25 mg QD were made. Subgroup analyses used pooled treatment groups (evobrutinib high dose [75 mg QD/BID] vs low dose [placebo/evobrutinib 25 mg QD]).

Results

The SEL analysis set included 223 patients (mean [SD] age: 42.4 [10.7] years; 69.3% female; 87.4% relapsing/remitting MS). Mean (SD) SEL volume was 2,099 (2,981.0) mm^3 with evobrutinib 75 mg BID vs 2,681 (3,624.2) mm^3 with placebo/evobrutinib 25 mg QD. Median number of SELs/patient ranged from 7 to 11 across treatments. SEL volume decreased with increasing evobrutinib dose vs placebo/evobrutinib 25 mg QD, and no difference with DMF vs placebo/evobrutinib 25 mg QD was noted. SEL volume significantly decreased with evobrutinib 75 mg BID vs placebo/evobrutinib 25 mg QD (−474.5 mm^3 [−1,098.0 to −3.0], $p = 0.047$) and vs DMF (−711.6 [−1,290.0 to −149.0], $p = 0.011$). SEL volume was significantly reduced for evobrutinib high vs low dose within baseline Expanded Disability Status Scale ≥3.5 and longer disease duration (≥8.5 years) subgroups.

Discussion

Evobrutinib reduced SEL volume in a dose-dependent manner in RMS, with a significant reduction with evobrutinib 75 mg BID. This is evident that evobrutinib affects brain lesions associated with chronic inflammation and tissue loss.

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Glossary

BID = twice daily; **BTK** = Bruton tyrosine kinase; **CAL** = chronic active lesion; **DMF** = dimethyl fumarate; **EAE** = encephalomyelitis; **EDSS** = Expanded Disability Status Scale; **EOT** = end of treatment; **Gd^+** = gadolinium-enhancing; **HDA** = high disease activity; **mITT** = modified intention-to-treat; **MS** = multiple sclerosis; **PRL** = paramagnetic rim lesion; **QD** = once daily; **RMS** = relapsing MS; **RRMS** = relapsing-remitting MS; **SEL** = slowly expanding lesion; **SPMS** = secondary-progressive MS; **SWI** = susceptibility weighted imaging.

Trial Registration Information

ClinicalTrials.gov number: NCT02975349. Submitted to ClinicalTrials.gov on November 29, 2016. First patient enrolled: March 7, 2017.

Classification of Evidence

This study provides Class II evidence that evobrutinib reduces the volume of SELs assessed on MRI comparing baseline with week 48, in patients with RMS.

Introduction

Multiple sclerosis (MS) is an inflammatory and immune-mediated progressive neurodegenerative disease of the CNS, characterized by demyelinated lesions in the brain and spinal cord that lead to neuronal loss and accumulation of neurologic disability.[1-3] The inflammatory processes in MS involve multiple cell types in the periphery and CNS, with CNS-compartmentalized inflammation being most relevant to the progressive neurodegeneration observed across all clinically defined stages of this disease.[4-6]

Chronic active (mixed active/inactive or smoldering) lesions identified on histopathology are chronically demyelinating MS lesions, likely driven by sustained microglia and/or macrophage activity, resulting in the progressive accumulation of irreversible neural tissue damage and axonal loss.[7-14] Slowly expanding lesions (SELs) identified on MRI are areas within preexisting T2 lesions that show gradual, radial expansion over time.[7] This expansion identifies areas of accumulating tissue damage within chronic lesions.[7,9,15,16] Thus, SELs are considered MRI markers of chronic active lesions (CALs) in vivo.

SELs correlate independently with clinical outcomes, predict long-term disability, and may represent an imaging marker of MS progression.[10,17] In a study of 52 patients with relapsing-remitting MS (RRMS), a higher proportion of SELs among baseline lesions was associated with worsening disability and MS progression over 9 years.[9] Similarly, in a study of 135 patients with RRMS, increased disability was independently associated with increasing SEL volume. In addition, increasing SEL volume was associated with up to a 5-times higher risk of confirmed disability progression.[17] In patients with primary progressive MS and secondary progressive MS (SPMS)—in the ORATORIO[10] and MS-SMART trials,[16] respectively—SELs were associated with higher disease activity and an increased risk of disability progression.

Evobrutinib, a highly selective, CNS-penetrant, covalent, Bruton tyrosine kinase (BTK) inhibitor, targets B cells, macrophages, and microglia.[18,19] In a phase 2 trial (NCT02975349) in patients with relapsing MS (RMS), evobrutinib 75 mg once daily (QD) and twice daily (BID) reduced T1 gadolinium-enhancing (Gd^+) and T2 lesions vs placebo (week 24) and the annualized relapse rate (week 48) vs placebo/evobrutinib 25 mg QD.[20] Evobrutinib was well-tolerated with the most common adverse events of any grade being nasopharyngitis and increased levels of lipase and asymptomatic, reversible alanine liver aminotransaminases.[20] Results from the ongoing open-label extension indicate that the efficacy and safety of evobrutinib are maintained after >4.5 years of treatment.[21] The evobrutinib 75 mg BID fasted dose used in this phase 2 trial is predicted to be comparable, with respect to exposure and BTK occupancy, with the 45 mg BID fed dose currently being used in the ongoing phase 3 trials (NCT04338022 and NCT04338061).[22,23] In patients with RMS, evobrutinib is present in the CSF at concentrations that overlapped the free plasma concentrations, which resulted in high levels of BTK occupancy, suggesting that evobrutinib may be able to exert a treatment effect within the CNS, targeting central immunopathology.[24] To investigate the potential treatment effect of evobrutinib within the CNS, the aim of our post hoc study was to evaluate the effect of evobrutinib treatment (at various doses for 48 weeks) vs placebo/evobrutinib 25 mg QD (placebo for 24 weeks, followed by evobrutinib 25 mg QD for 24 weeks) on SEL volume, assessed based on MRI from baseline to week 48, in a phase 2 RMS trial.

Methods

Trial Design

These post hoc SEL analyses were performed on data from a phase 2, randomized, placebo-controlled trial, comprising a 48-week double-blind period with a parallel, open-label, dimethyl fumarate (DMF) reference group. Detailed descriptions of the

protocol, methods, and primary results have been published previously.[20] Additional information can also be found at ClinicalTrials.gov using clinical study number NCT02975349. Patient enrollment occurred between March and September 2017.

In brief, eligible trial participants were 18–65 years of age, diagnosed with RRMS or SPMS with superimposed relapses, had ≥1 documented relapse(s) within 2 years before screening (either 1 relapse within 1 year before randomization or ≥1 T1 Gd^+ MRI lesion within 6 months before randomization), and had an Expanded Disability Status Scale (EDSS) score of 0–6 at baseline.

Patients were randomized 1:1:1:1:1 to receive evobrutinib 25 mg QD, evobrutinib 75 mg QD, evobrutinib 75 mg BID, placebo (this group was then switched to evobrutinib 25 mg QD after week 24; blinding was preserved at the time of switching), or DMF 240 mg BID (open-label reference group). For simplicity and accuracy, the placebo treatment arm will be described as placebo/evobrutinib 25 mg QD. Evobrutinib and DMF were administered while fasting (i.e., taken >1 hour before meal or >2 hours after meal).

MRI scans were performed, using a standardized imaging protocol, at screening and at weeks 12, 16, 20, 24, 48, and end of treatment (EOT). Axial T1-weighted slices were acquired with 3-dimensional spoiled gradient echo (repetition time = 28–30 milliseconds, echo time = 5–11 milliseconds, flip angle = 27–30°, and resolution 1 × 1 × 3 mm). Axial T2-weighted slices were acquired with 2-dimensional fast spin-echo (repetition time = 4,500–6,200 milliseconds, echo time = 66–91 milliseconds, and resolution 1 × 1 × 3 mm). Both T1-weighted and T2-weighted images were used for detection of SELs.

All patients from the modified intention-to-treat (mITT) analysis set were investigated in these SEL analyses. The mITT analysis set consisted of all randomized patients who received at least 1 dose of the trial treatment and who have at least 1 baseline and 1 post-baseline MRI assessment.

Standard Protocol Approvals, Registrations, and Patient Consents

The trial is registered with ClinicalTrials.gov (NCT02975349) and was conducted in accordance with the Declaration of Helsinki and the International Conference on Harmonisation Good Clinical Practice Guidelines. Written informed consent was obtained from each patient before any trial-related activities were performed.

Identification of SELs

The SEL identification process using precontrast T1-weighted and T2-weighted MRI images simultaneously has been described previously.[7] In brief, SELs were identified, based on MRI scans over 48 weeks, as areas of preexisting T2 lesions of at least 10 contiguous voxels (size ~30 mm^3) showing gradual and constant concentric expansion over time. Local expansion was determined using Jacobian analysis, a technique based on nonlinear registration that is typically used to measure subtle volume changes in specific brain structures. T1-weighted and T2-weighted images were used together to drive the nonlinear registration. By subsequently taking the Jacobian determinant of the resultant deformation field, the rate of local volume change between 2 time points could be quantified at each voxel. SEL identification was performed as a 2-step process. First, contiguous regions of T2 lesions preexisting at baseline that underwent a minimum local volume expansion were identified as SEL candidates. For the experiments performed in this study, the initial threshold for SEL identification was a minimum expansion of 12.5% per year and SEL boundaries were then refined based on a minimum expansion of 4% per year, where expansion was determined by the Jacobian determinant.[7] SEL candidates were subsequently scored to favor those undergoing gradual and constant expansion over time and those with concentric centrifugal (inside-out) radial expansion. SEL identification was performed by an independent reading center (NeuroRx Research) who remained blinded to all trial patient and treatment assignment information.

Statistical Analyses

The SEL volume analysis was based on all patients with available SEL measurements from the mITT analysis set (SEL analysis set). Both the absolute SEL volume and SEL volume as a percentage of baseline T2 lesion volume were investigated as follows. Two analyses, stratified by baseline T2 lesion volume tertiles, of SEL volume (absolute and percentage) were performed. The primary analysis included all patients with SEL volume values regardless of whether it was a week 48 or EOT value (i.e., treatment completers and non-completers). The sensitivity analysis included only those patients with week 48 SEL volume values (i.e., treatment completers). The baseline T2 lesion volume tertiles were as follows: tertile 1, ≤8,000 mm^3 (≤8 cm^3); tertile 2, 8,000–19,000 mm^3 (8–19 cm^3); and tertile 3, ≥19,000 mm^3 (≥19 cm^3). Treatment effects between the evobrutinib and DMF treatment groups vs placebo/evobrutinib 25 mg QD were analyzed using a stratified Hodges-Lehmann estimate of shift in SEL volume distribution and stratified Wilcoxon rank-sum test. The abovementioned analyses were repeated for subgroups based on pooled evobrutinib treatment groups (evobrutinib high dose [75 mg QD and BID] vs low dose [placebo/evobrutinib 25 mg QD and evobrutinib 25 mg QD]). Pooled treatment groups were used to analyze the subgroups because of the small patient numbers across the treatment arms, as well as the lack of effect of the placebo/evobrutinib 25 mg QD and evobrutinib 25 mg QD treatment arms on MRI and clinical end points during the 48-week phase 2 trial.[20] The following subgroups were analyzed: baseline EDSS ≤3.0 vs ≥3.5; non-high disease activity (HDA; ≤1 relapse in 2 years before randomization) vs HDA (≥2 relapse in 2 years before randomization); recent disease onset (<8.5 years) vs protracted disease onset (≥8.5 years); and RRMS vs SPMS. The cutoffs used for EDSS and disease onset

Table 1 Baseline Characteristics

	Placebo/evobrutinib 25 mg QD (n = 53)	Evobrutinib 25 mg QD (n = 50)	Evobrutinib 75 mg QD (n = 51)	Evobrutinib 75 mg BID (n = 53)	DMF 240 mg BID (n = 54)
Sex, n (%)					
Male	14 (26.4)	18 (36.0)	16 (31.4)	17 (32.1)	15 (27.8)
Female	39 (73.6)	32 (64.0)	35 (68.6)	36 (67.9)	39 (72.2)
Age, y, mean ± SD	41.6 ± 10.8	42.4 ± 9.4	42.9 ± 10.1	42.2 ± 11.5	42.8 ± 11.7
Time since MS onset, y, n (%)					
<8.5 y	32 (60.4)	26 (52.0)	20 (39.2)	23 (43.4)	29 (53.7)
≥8.5 y	21 (39.6)	23 (46.0)	31 (60.8)	30 (56.6)	25 (46.3)
Type of MS, n (%)					
RRMS	47 (88.7)	42 (84.0)	43 (84.3)	47 (88.7)	49 (90.7)
SPMS	6 (11.3)	8 (16.0)	8 (15.7)	6 (11.3)	5 (9.3)
No. of relapses in 2 y before randomization, n (%)					
≤1 relapse (non-HDA)	26 (49.1)	27 (54.0)	18 (35.3)	25 (47.2)	20 (37.0)
≥2 relapses (HDA)	27 (50.9)	23 (46.0)	33 (64.7)	28 (52.8)	34 (63.0)
EDSS score, n (%)					
≤3	27 (50.9)	28 (56.0)	22 (43.1)	28 (52.8)	35 (64.8)
≥3.5	26 (49.1)	22 (44.0)	29 (56.9)	25 (47.2)	19 (35.2)
T1 Gd^+ lesions					
Patients with lesions, n (%)	24 (45.3)	19 (38.0)	18 (35.3)	23 (43.4)	19 (35.2)
Mean ± SEM	1.2 ± 0.3	0.9 ± 0.3	1.7 ± 0.8	1.7 ± 0.5	2.2 ± 0.9
T2 lesion volume, cm^3					
Mean ± SD	15.9 ± 12.6	13.8 ± 11.7	14.0 ± 12.2	19.0 ± 13.5	18.8 ± 17.7
Median	12.9	10.5	9.4	16.2	15.3

Abbreviations: BID = twice daily; DMF = dimethyl fumarate; EDSS = Expanded Disability Status Scale; HDA = high disease activity; mITT = modified intention-to-treat; MS = multiple sclerosis; QD = once daily; RRMS = relapsing-remitting MS; SPMS = secondary progressive MS.
mITT analysis set.

corresponded with the median observed in the mITT population to ensure the subgroups were balanced.

Data Availability

Data are available on reasonable request. Any requests for data by qualified scientific and medical researchers for legitimate research purposes will be subject to the Data Sharing Policy of the health care business of Merck KGaA, Darmstadt, Germany. All requests should be submitted in writing to the data sharing portal of the health care business of Merck KGaA, Darmstadt, Germany, emdgroup.com/en/research/our-approach-to-research-and-development/healthcare/clinical-trials/commitment-responsible-data-sharing.html. When the health care business of Merck KGaA, Darmstadt, Germany, has a co-research, co-development, or co-marketing or co-promotion agreement or when the product has been out-licensed, the responsibility for disclosure might be dependent on the agreement between parties. Under these circumstances, the health care business of Merck KGaA, Darmstadt, Germany, will endeavor to gain agreement to share data in response to requests.

Results

The demographics and baseline characteristics of the patients included in this analysis are summarized in Table 1. Overall, mean (SD) SEL volume was 2,099 (2,981.0) mm^3 with evobrutinib 75 mg BID vs 2,681 (3,624.2) mm^3 with placebo/evobrutinib 25 mg QD (Table 2). The median SEL volume, measured as a percentage of BL T2 lesion volume, was 8.9% with evobrutinib 75 mg BID vs 10.4% with placebo/evobrutinib 25 mg QD (Table 2). The median number of SELs per patient was similar across treatment groups

Table 2 SEL Volume

	Placebo/evobrutinib 25 mg QD	Evobrutinib 25 mg QD	Evobrutinib 75 mg QD	Evobrutinib 75 mg BID	DMF 240 mg BID
No. of SELs per patient					
All patients					
N	42	42	46	43	50
Median (min, max)	8 (0, 32)	7 (0, 45)	8.5 (0, 42)	11 (0, 41)	10 (0, 57)
Completers (week 48)					
N	38	39	42	42	50
Median (min, max)	7 (0, 32)	8 (0, 45)	8.5 (0, 42)	11.5 (0, 41)	10 (0, 57)
Absolute SEL volume					
All patients, mm^3					
N	42	42	46	43	50
Mean ± SD	2,681 ± 3,624.2	2,043 ± 2,692.0	1,920 ± 2,288.1	2,099 ± 2,981.0	2,866 ± 4,042.9
Completers (week 48), mm^3					
N	38	39	42	42	50
Mean ± SD	2,493 ± 3,602.8	2,196 ± 2,735.3	1,887 ± 2,315.0	2,109 ± 3,016.3	2,866 ± 4,042.9
SEL volume as a percentage of baseline T2 lesion volume					
All patients, %					
N	42	42	46	43	50
Median	10.4	11.0	8.2	8.9	9.5

Abbreviations: BID = twice daily; DMF = dimethyl fumarate; mITT = modified intention-to-treat; QD = once daily; SEL = slowly expanding lesion.
SEL analysis set.

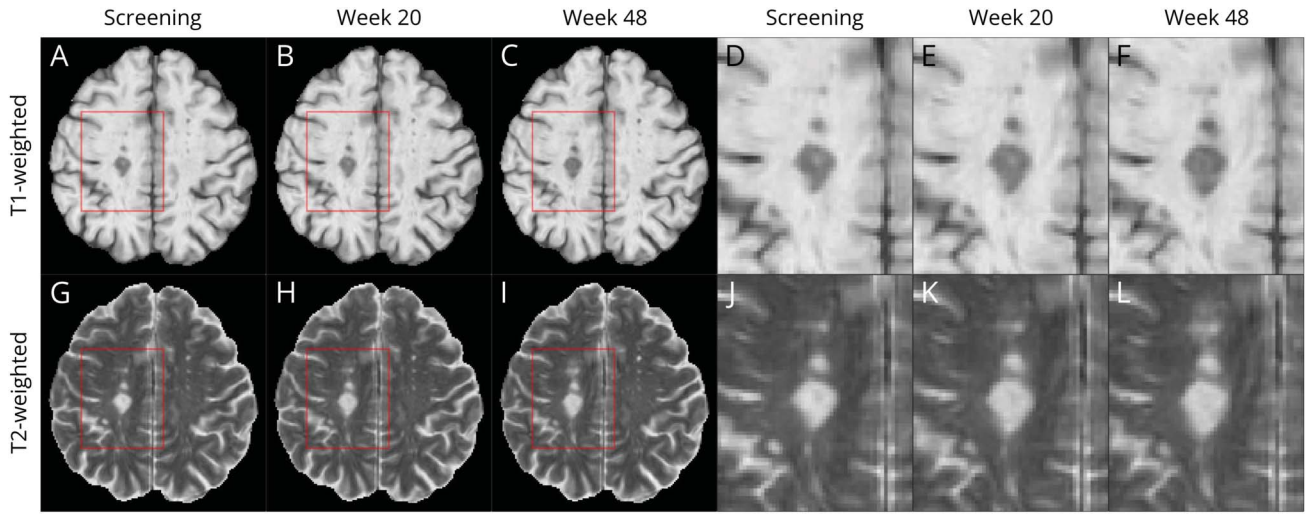
(placebo/evobrutinib 25 mg QD: 8; evobrutinib 25 mg QD: 7; evobrutinib 75 mg QD: 8.5; evobrutinib 75 mg BID: 11; DMF 240 mg BID: 10; Table 2). Details of clinical relapses and MRI outcomes over the 48-week double-blind trial have been published previously.[20] An example of a SEL at 3 time points over the 48-week trial is shown in Figure 1. An animated version of this figure is more appropriate for data visualization and is available in Video 1 along with 2 additional animated examples (Videos 2 and 3).

Relative to placebo/evobrutinib 25 mg QD, the absolute SEL volume decreased numerically with an increasing evobrutinib dose in both the all-patient and completer analyses (Figure 2A); the reduction was significant for evobrutinib 75 mg BID in the all-patient analysis (location shift −474.5 mm^3 [95% CI −1,098.0 to −3.0], $p = 0.047$). Similar results were observed for SEL volume as a percentage of baseline T2 lesion volume (Figure 2B), where significant reductions in SEL volume were seen in both the all-patient and completer analyses for evobrutinib 75 mg QD (location shift −3.26% [−6.18 to −0.08], $p = 0.040$ and −3.48% [−6.53 to −0.50], $p = 0.022$) and 75 mg BID (location shift −4.34% [−7.02 to −1.46], $p = 0.003$ and −4.11% [−6.84 to −1.23], $p = 0.005$). The nonstratified test results (data not shown) indicated that most of the adjustment for baseline T2 lesion volume is accomplished using the percent volume end point with additional adjustment accomplished using stratification.

There was no difference in SEL volume (absolute or percent) with DMF 240 mg BID compared with placebo/evobrutinib 25 mg QD (Figure 2, A and B). However, there was a significant reduction in absolute SEL volume for evobrutinib 75 mg BID vs DMF 240 mg BID in both the all-patient (location shift −711.6 mm^3 [95% CI −1,290.0 to −149.0], $p = 0.011$) and completer (location shift −736.1 mm^3 [95% CI −1,335.5 to −157.0], $p = 0.009$; Figure 3, A and B) analyses.

When evaluated by baseline T2 lesion volume, the greatest SEL volume (absolute and percent) and greatest treatment effect were in those patients with the highest T2 lesion volume in tertile 3 (≥19,000 mm^3; Figure 4, A and B). With DMF 240 mg BID, SEL volume was less than (percent volume) or similar (absolute volume) to placebo/evobrutinib 25 mg QD

Figure 1 Example of a SEL Shown at 3 Time Points Over 48 Weeks



(A–C) and (G–I) show the same axial slice on T1-weighted and T2-weighted images, respectively, at screening, week 20, and week 48. (D–F) and (J–L) shows a zoomed-in region of interest corresponding to the red box in (A–C) and (G–I) highlighting the SEL. An animated version of this figure is more appropriate for data visualization and is available in Video 1. SEL = slowly expanding lesion.

in the overall population and when analyzed by tertiles of baseline T2 lesion volume.

Overall, there was a greater effect with a high evobrutinib dose (75 mg QD and BID) vs a lower dose (placebo/evobrutinib 25 mg QD and evobrutinib 25 mg QD) on absolute SEL volume in patients with more advanced disease as seen in those subgroups with higher baseline EDSS scores (≥3.5), high disease activity (≥2 relapses in 2 years before randomization), and protracted disease onset (≥8.5 years; eFigure 1, links.lww.com/WNL/D410). There were similar results for SEL volume as a percentage of baseline T2 lesion volume (eFigure 2, links.lww.com/WNL/D411). The RRMS (absolute and percent) and SPMS (absolute completer analysis and percent) subgroups also demonstrated a reduction in SEL volume with a high evobrutinib dose vs a lower dose.

Classification of Evidence

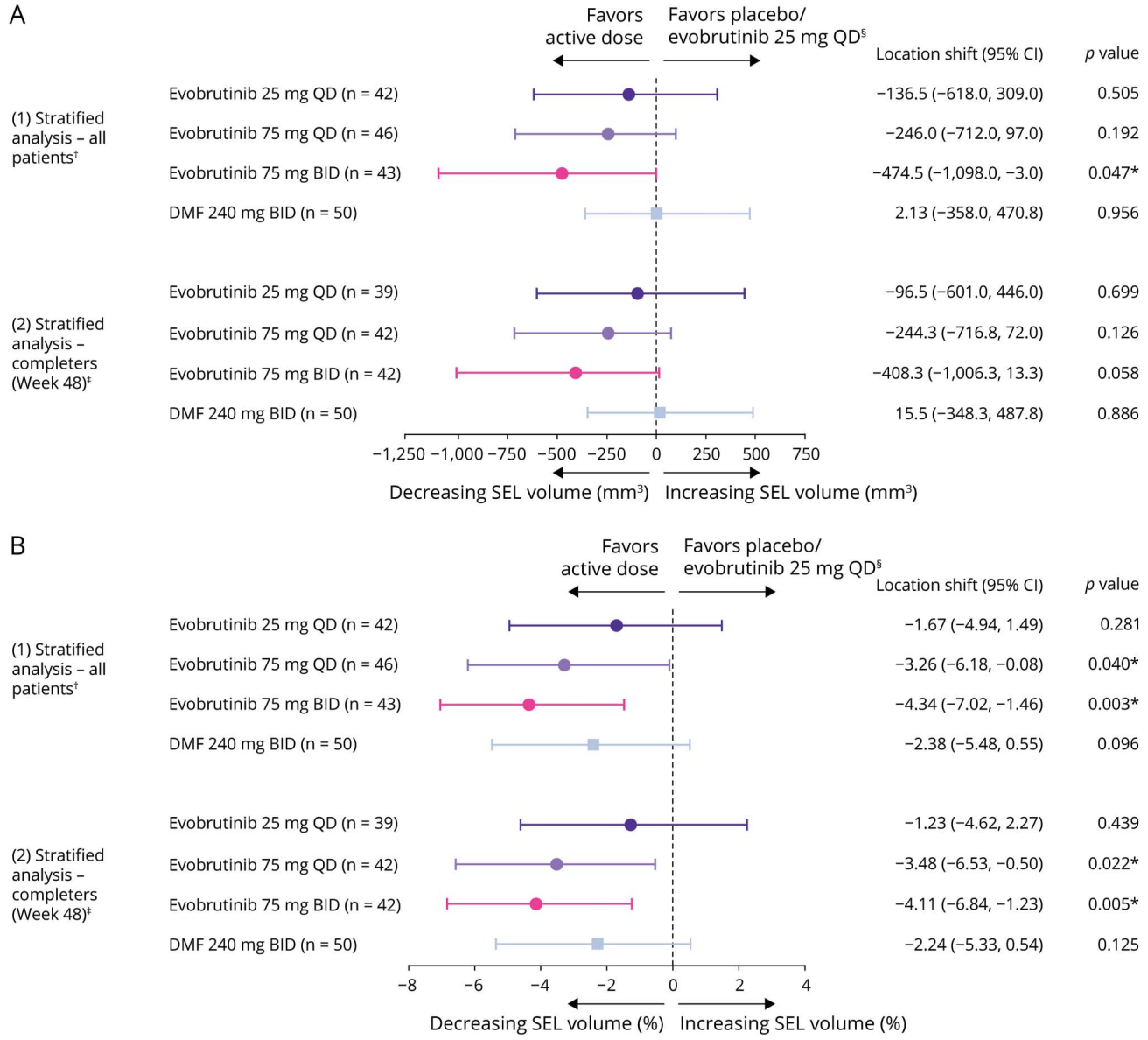
This study provides Class II evidence that evobrutinib reduces the volume of SEL assessed on MRI comparing baseline with week 48, in patients with RMS.

Discussion

SELs are an MRI marker of CALs in vivo that correlate independently with clinical outcomes including long-term disability.[10,17] This study observed that evobrutinib significantly reduces SEL volume (both absolute and percent) in a dose-dependent manner in RMS, particularly in patients treated with evobrutinib 75 mg BID (fasted dose—predicted to be comparable, with respect to exposure and BTK occupancy, with the 45 mg BID fed dose that is being evaluated in phase 3).[22,23] CALs have a thin border of inflammatory macrophages and microglia that have heterogeneous roles in MS, exerting both beneficial and detrimental effects, including driving a proinflammatory environment.[1,3,25-29] Preclinical data have demonstrated that evobrutinib has a direct effect on proinflammatory microglia and macrophages in mouse models.[30,31] This direct effect on proinflammatory microglia/macrophages,[30,31] cell populations that are linked to CNS-compartmentalized pathologic inflammation and progressive neurodegeneration, is of particular relevance considering the evidence supporting the ability of evobrutinib to exert a central effect by penetrating the CNS.[24,32,33] Evobrutinib has been detected in the plasma and brains of experimental autoimmune encephalomyelitis (EAE) mice[32] and the CSF of patients with RMS.[24] In both B-cell–dependent and independent models of experimental CNS autoimmunity, evobrutinib limited CNS-compartmentalized inflammation, demyelination, and disease severity.[32,34] Evobrutinib has also been shown to significantly reduce neuroinflammation, demyelination, and axonal pathology both in the brain and spinal cord in an in vivo EAE mouse model.[33] Taken together with the significant reduction in SEL volume, these data suggest that evobrutinib 75 mg BID can achieve high levels of BTK occupancy and directly inhibit/limit crucial CNS-inflammatory pathways.

Some MS therapies directed at the adaptive immune system have been shown to have a modest effect on both the number and volume of SELs.[10,35-37] However, it remains unclear whether this is a direct effect on the microglia within CALs or an indirect effect through the suppression of the acute inflammatory environment in the brain. It is expected that the effect of most MS drugs, including monoclonal antibodies, on CALs, is not through direct effects on microglia, but because of their effects on peripheral immune cells.[3,25,38,39] Monoclonal

Figure 2 Evobrutinib Treatment Groups vs Placebo/Evobrutinib 25 mg QD or DMF Treatment Group vs Placebo/Evobrutinib 25 mg QD



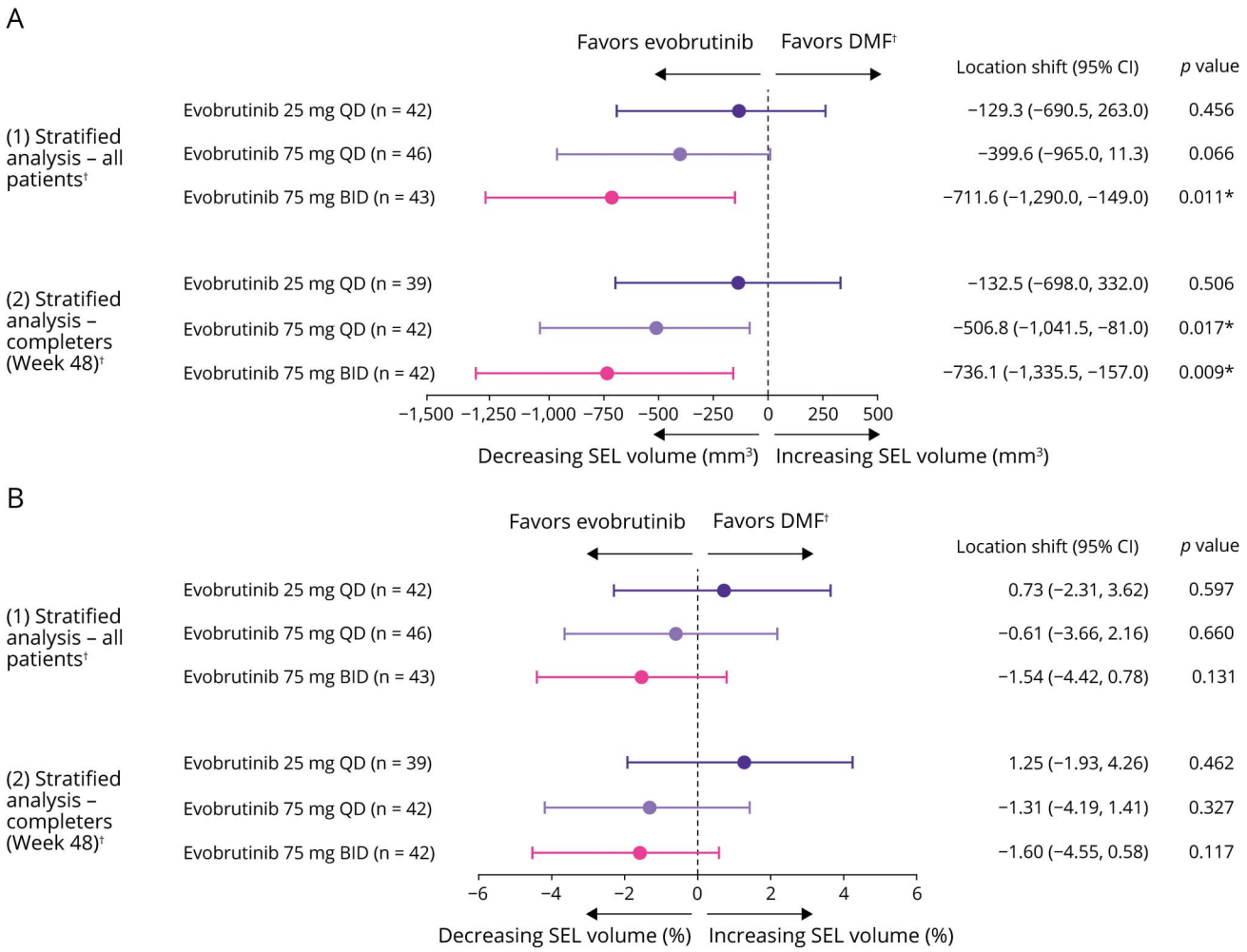
(A) Absolute SEL volume. (B) SEL volume as a percentage of baseline T2 lesion volume (stratified analyses). BID = twice daily; DMF = dimethyl fumarate; QD = once daily; SEL = slowly expanding lesion. *p value <0.05. †Evobrutinib or DMF treatment groups vs placebo/evobrutinib 25 mg QD (n = 42). ‡Evobrutinib or DMF treatment groups vs placebo/evobrutinib 25 mg QD (n = 38). §Patients switched from placebo to evobrutinib 25 mg QD for the second 24-week treatment period. SEL analysis set.

antibodies, including anti-CD20 s and natalizumab, because of their large size, have limited presence in the CNS.[40]

With ocrelizumab, a modest treatment effect vs placebo on SEL volume was observed in the ORATORIO trial in patients with progressive MS; when SELs were identified over 120 weeks, median SEL volume as a percentage of baseline T2 lesion volume was 2.5% with ocrelizumab vs 3.4% with placebo.[10] In the ASCEND trial in patients with SPMS, when SELs were identified over 108 weeks, median SEL volume as a percentage of baseline T2 lesion volume was 2.7% with natalizumab vs 5.0% with placebo.[36] Both of these trials used the same SEL identification methodology as in our study.

In our study of patients with RMS, with SELs identified over 48 weeks, median SEL volume as a percentage was 8.9% with evobrutinib 75 mg BID vs 10.4% with placebo/evobrutinib 25 mg QD or 8.7% with a high evobrutinib dose vs 10.6% (9.9) with a lower evobrutinib dose. The greater volume of SELs seen in our study compared with the ORATORIO trial likely reflects, at least in part, the different intervals over which SELs were measured; larger SEL volumes are observed over shorter intervals, possibly

Figure 3 Evobrutinib Treatment Groups vs DMF



(A) Absolute SEL volume. (B) SEL volume as a percentage of baseline T2 lesion volume (stratified analyses). BID = twice daily; DMF = dimethyl fumarate; QD = once daily; SEL = slowly expanding lesion. †Evobrutinib treatment groups vs DMF 240 mg BID (n = 50). SEL analysis set.

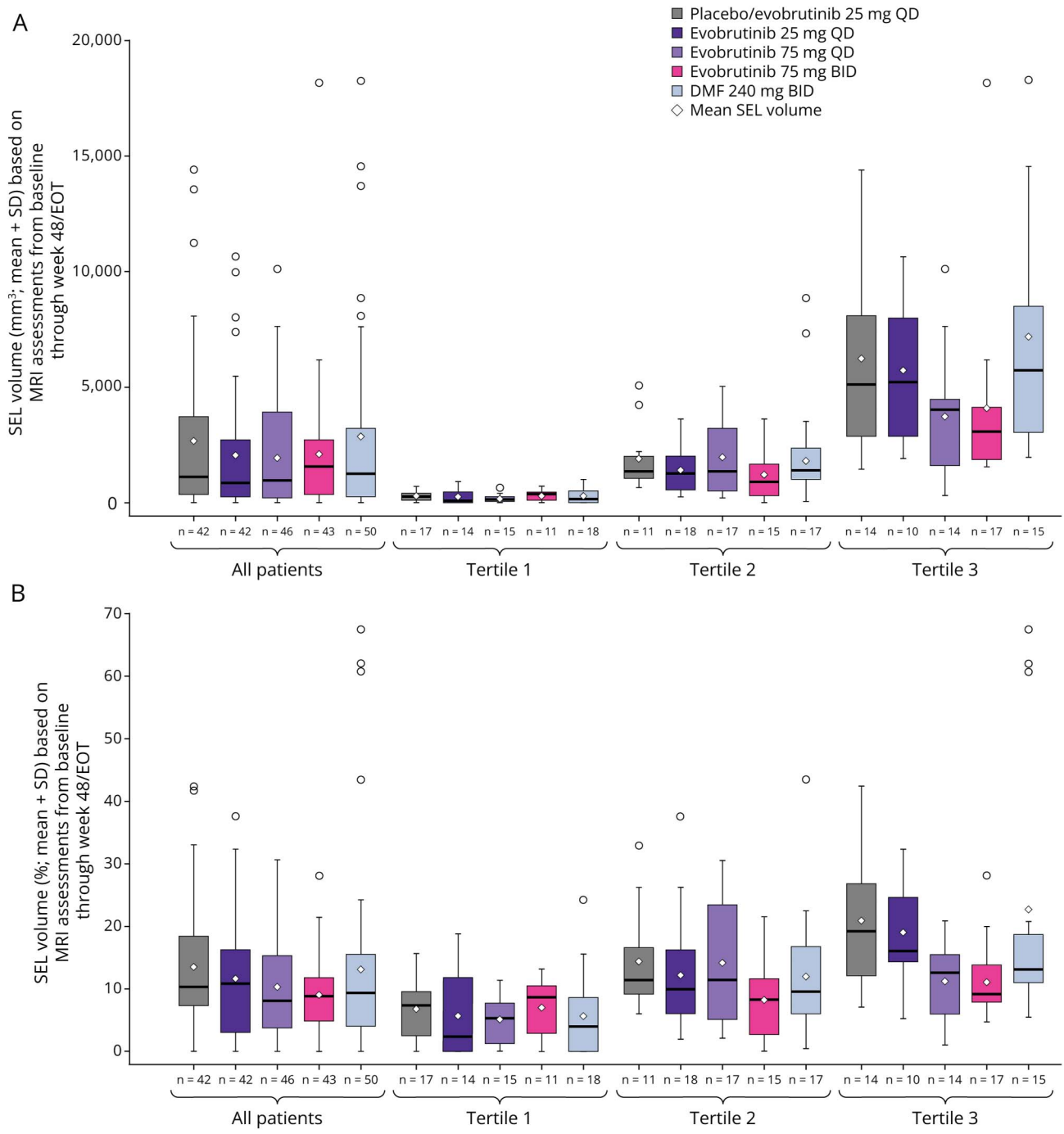
because of increased noise in the SEL measurement and because fewer chronic lesions may show constant expansion over longer intervals.[41] This noise would be expected to dilute the observed treatment effect. For this reason, and because the patient populations in the progressive MS trials referenced above were more disabled with longer disease duration, it is difficult to directly compare the effects of evobrutinib and the monoclonal antibodies used in the abovementioned trials.

Our data demonstrate a dose-dependent decrease in SEL volume with the greatest effect observed in those patients with more advanced disease (including the RRMS and SPMS subgroups) and greater T2 lesion volume. This is of particular importance because patients with more active or advanced disease typically have the most SELs and greatest SEL volume.[7,10,36] Data from a recent study have indicated that 86%–99% of patients with RRMS had SELs, suggesting that SELs (and progressive biology) are present from early stages of MS and are not just a marker of a later disease stage.[17] It should be noted that the number of SELs per patient was similar across treatment groups. As SELs are identified from the baseline unenhancing T2 lesion volume, the fact that the number of lesions does not differ between treatment groups, but the SEL volume does, suggests that treatment with evobrutinib reduces the volume of SELs that is expanding, possibly by slowing the rate of expansion of SELs, without stopping it completely within the 48-week period of this study.

To date, the effects of treatment on SELs have been investigated for another BTK inhibitor, tolebrutinib, in a phase 2 trial of patients with RMS.[42] In this trial with tolebrutinib, placebo was administered for only 4 weeks, and SEL detection was performed for multiple dose groups over only 16 weeks, which is a very short interval to detect slow expansion.[42] Thus, this trial design was suboptimal for SEL analysis, and current data only report the SEL volume by treatment dosing group with no comparison with placebo or stratification by baseline T2 lesion volume.

The findings presented here should be considered in the context of certain study limitations. The correlation of SELs to disability and progression is well established; however, while SELs are a

Figure 4 SEL Volume by Tertiles of Baseline T2 Lesion Volume



(A) Absolute SEL volume. (B) SEL volume as a percentage of baseline T2 lesion volume. BID = twice daily; DMF = dimethyl fumarate; EOT = end of treatment; QD = once daily; SEL = slowly expanding lesion. SEL analysis set. Tertiles of baseline T2 lesion volume in overall population—tertile 1: ≤8,000 mm^3 (≤8 cm^3); tertile 2: 8,000–19,000 mm^3 (8–19 cm^3); and tertile 3: ≥19,000 mm^3 (≥19 cm^3). SEL volume based on MRI assessments from baseline through week 48/EOT.

marker of chronic lesion activity, there is limited pathologic validation of SELs as a marker of CALs. Future studies are warranted to investigate this point. While it is possible that a pseudoatrophy effect could contribute to the greater effect of evobrutinib 75 mg BID on SEL volume, pseudoatrophy has not been observed with evobrutinib 75 mg BID. This SEL volume analysis from a phase 2 trial was not prespecified. In addition, the overall small sample size in phase 2 trials meant that the analysis was not powered to detect SEL volume differences between high-dose and low-dose groups within various subgroups. In particular, the SPMS subgroup was relatively small compared with the RRMS subgroup. However, directionally, we did observe that higher doses of evobrutinib had a positive effect on SEL volume compared with lower doses. Finally, paramagnetic rim lesions (PRLs) or iron-rim lesions are another MRI marker of CALs based on susceptibility-weighted imaging (SWI) and the presence of iron at the lesion edge.[8,13,43,44] PRLs

were not considered in this study because SWI was not part of the imaging protocol for the trial. However, future evaluation of SELs and PRLs may help to clarify the biological underpinnings of SELs and other MRI lesion subtypes.

Evobrutinib dose-dependently reduced the volume of SELs, a marker of ongoing tissue loss within chronic lesions and associated with long-term disability accumulation. The reduction in SEL volume was most notable in patients treated with evobrutinib 75 mg BID and in patients with more advanced disease and greater T2 lesion volume at baseline. Overall, this is the most comprehensive evidence that a BTK inhibitor affects brain lesions associated with chronic inflammation and tissue loss.

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Disclosure

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Colm Elliott, PhD	NeuroRx Research, Montreal, Quebec, Canada	Drafting/revision of the manuscript for content, including medical writing for content; study concept or design; analysis or interpretation of data
Emily C. Martin, PhD	EMD Serono, Billerica, MA	Drafting/revision of the manuscript for content, including medical writing for content; study concept or design; analysis or interpretation of data
Yann Hyvert, PhD	The Healthcare Business of Merck KGaA, Darmstadt, Germany	Drafting/revision of the manuscript for content, including medical writing for content; study concept or design; analysis or interpretation of data
Davorka Tomic, MD	Ares Trading SA, Eysins, Switzerland, an affiliate of Merck KGaA, Darmstadt, Germany	Drafting/revision of the manuscript for content, including medical writing for content; analysis or interpretation of data
Xavier Montalban, MD, PhD	Centre d'Esclerosi Múltiple de Catalunya (Cemcat), Hospital Universitario Vall d'Hebron, Barcelona, Spain	Drafting/revision of the manuscript for content, including medical writing for content; major role in the acquisition of data; analysis or interpretation of data

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