Title Page

CLOCK-MS Protocol

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Title Page

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1. Protocol Summary

1.1. Synopsis

Protocol Title: CLadribine Tablets: Collaborative Study to Evaluate the Impact On Central Nervous System BiomarKers in Multiple Sclerosis (CLOCK-MS)

Short Title: Cladribine Tablets MoA Study

Rationale: The purpose of this exploratory study is to better understand the mechanism of action (MoA) of cladribine tablets by exploring the effect on central nervous system (CNS) biomarkers relevant in the relapsing forms of multiple sclerosis (RMS; to include relapsing-remitting MS [RRMS] and active secondary progressive MS). The study is designed to generate hypotheses regarding the impact and relevance of cladribine tablet activity in the CNS by assessing the cerebrospinal fluid (CSF) levels of lymphocyte subsets, other immune cells, neuronal injury markers, and soluble immunological markers in study participants with RMS before and during treatment with cladribine tablets, and the association of these CSF markers with corresponding blood markers and with clinical outcomes.

Objectives and Endpoints

Objectives	Endpoints (Outcome Measures)
Primary	
• To evaluate changes in the CSF levels of lymphocyte subtypes (CD3+ T lymphocytes, CD19+ B lymphocytes) and markers of neuronal injury (NfL) during treatment with cladribine tablets in patients with RMS	• Change in CSF levels of CD3+ T lymphocytes, CD19+ B lymphocytes, and NfL from baseline to second LP (end of Week 5, end of Week 10, or end of Year 1, and/or end of Year 2[a])
Secondary	
• To evaluate changes in the CSF levels of other lymphocyte subtypes and immune cells during treatment with cladribine tablets	• Change in CSF levels of CD4+ and CD8+ cells, plasmablasts, NK cells, Tregs, and CD14+ cells from baseline to second LP (end of Week 5, end of Week 10, or end of Year 1, and/or end of Year 2[a])
• To assess cladribine CSF and serum levels during treatment with cladribine tablets	• Cladribine CSF and serum concentrations at baseline and second LP (end of Week 5 time point only)

Objectives	Endpoints (Outcome Measures)
• To assess the peripheral blood levels of lymphocyte subtypes, other immune cells, and soluble immunological and neuronal injury markers during treatment with cladribine tablets	• Change in peripheral blood levels of CD3+, CD4+ CD8+, CD19+, Th1, Th2, and Th17 cells, Tregs, plasmablasts, NK cells, CD14+ cells, and NfL levels from baseline to second LP (end of Week 5, end of Week 10, or end of Year 1, and/or end of Year 2[a])
• To assess clinical outcomes, based on the ARR and EDSS, during treatment with cladribine tablets	• ARR over 24 months of treatment with cladribine tablets • Change in EDSS scores from baseline at Month 12 and Month 24 • 6-month confirmed EDSS progression
• To evaluate MRI findings during treatment with cladribine tablets, using: o Standard sequences to evaluate T1 Gd+ and new or enlarging T2 lesions, and o Quantitative gradient echo contrast imaging (R2t*) of cerebral cortex, deep gray matter, and white matter to measure tissue integrity	• MRI findings at Month 12 and Month 24 compared to baseline: o Proportion of participants with no Gd+ lesions o Proportion of participants with no new or newly enlarging T2 lesions o Mean number of Gd+ lesions per participant o Mean number of new or newly enlarging T2 lesions per participant • Change in R2t* in white matter and gray matter
• To collect safety data (adverse events, hematology) during treatment with cladribine tablets	• Occurrence of TEAEs, SAEs, and TEAEs leading to treatment discontinuation • Absolute lymphocyte count assessed per USPI • Complete blood cell count as assessed per routine clinical practice

[a] Data from any third, optional end of Year 2 LP will be included with the data from protocol-required end of Year 2 LPs.ARR=annualized relapse rate, CD=cluster of differentiation, CSF=cerebrospinal fluid, EDSS=Expanded Disability Status Scale, Gd+=gadolinium-enhancing, LP=lumbar puncture, NfL=neurofilament light chain, MRI=magnetic resonance imaging, NK cells=natural killer cells, Tregs= regulatory T cells, RMS=relapsing forms of multiple sclerosis, SAE=serious adverse event, TEAE=treatment-emergent adverse event, Th cell=T helper cell, USPI=United States Prescribing information.

Overall Design

This will be an open-label, randomized, multicenter collaborative research Phase 4 biomarker study, designed to generate hypotheses to better understand the MoA of cladribine tablets in RMS (to include RRMS and active secondary progressive MS). Approximately 50 study participants with RMS who meet all eligibility criteria, and who are willing and able to receive lumbar punctures (LPs), are planned to be enrolled at 5 centers in the United States. Participants are required to be naïve to treatment with cladribine tablets.

All study participants will be assigned to treatment with cladribine 10 mg tablets according to the approved United States Prescribing Information (USPI; 2 yearly treatment courses). Study participants will be randomized 1:2:2:1 to receive a total of 2 LPs according to 1 of 4 different schedules as described below. All study participants will have LP-aligned blood samples taken at baseline (on the same day as the baseline LP), and post-baseline at the end of Week 5, Week 10, Year 1, and Year 2).

Aliquots of the CSF- and the LP-aligned blood samples will be sent to the central laboratory for cladribine concentration measurement and for soluble biomarker assessments; cellular biomarkers will be assessed locally.

Clinical outcomes including relapses will be assessed at baseline, Week 5, Week 10, Months 6, 9, and 12 during Year 1, and at Months 14, 18, 21, and 24 during Year 2.

Optional, exploratory single cell RNA sequencing of CSF and blood cells may be performed locally at each site if sufficient CSF and blood sample material is available. In addition, RNA samples will be stored for future analysis from all study participants who consent to an optional RNA sample banking.

If the start of cladribine tablets Year 2 treatment needs to be delayed (e.g. to allow for lymphocyte recovery to ≥800 cells/µL), all following visits and procedures will be postponed by the same period of time (for up to 6 months), to maintain the protocol-specified time interval between start of cladribine tablets Year 2 treatment and all following assessments.

Disclosure Statement: This is a single-treatment group, open-label, randomized (different time schedules of LPs), basic science, multicenter collaborative research Phase 4 study.

Number of Participants: Approximately 50 participants will be randomly assigned (1:2:2:1) to 4 different groups with different LP schedules.

Intervention Groups and Duration: The total duration of the study will be 2 years for each participant. After an initial 3-week screening period, all enrolled study participants will undergo an initial LP and then initiate treatment with cladribine 10 mg tablets at baseline (Day 1). All participants will receive cladribine 10 mg tablets at a cumulative dose of 3.5 mg/kg divided into 2 treatment courses per the USPI (1.75 mg/kg per treatment course; Year 1 and Year 2 treatment). Any treatment delays will follow the guidance from the approved USPI. On Day 1, patients will be randomized 1:2:2:1 (approximately 9:16:16:9 participants planned) to receive a total of 2 LPs according to 1 of the 4 following schedules:

- Group 1: Baseline and end of Week 5 (optimal time point for assessing cladribine concentrations in CSF; LP to take place same day following intake of the last Week 5 cladribine tablet)
- Group 2: Baseline and end of Week 10 (expected "nadir" time for lymphocyte and monocyte quantities in CSF)
- Group 3: Baseline and end of Year 1 (at least 43 weeks after the last dose of cladribine tablets Year 1 treatment, but must occur prior to next treatment course)
- Group 4: Baseline and end of Year 2 (at least 43 weeks after the last dose of cladribine tablets Year 2 treatment) (Year 1 and Year 2 time points needed to assess if cladribine tablets effects on CSF markers are maintained at the end of the treatment cycle).

  Note: If the start of cladribine tablets Year 2 treatment needs to be delayed (e.g. to allow for lymphocyte recovery to ≥800 cells/µL), the end of Year 2 LP will be postponed by the same period of time (for up to 6 months), to maintain the protocol-specified time interval between start of cladribine tablets Year 2 treatment and second LP.

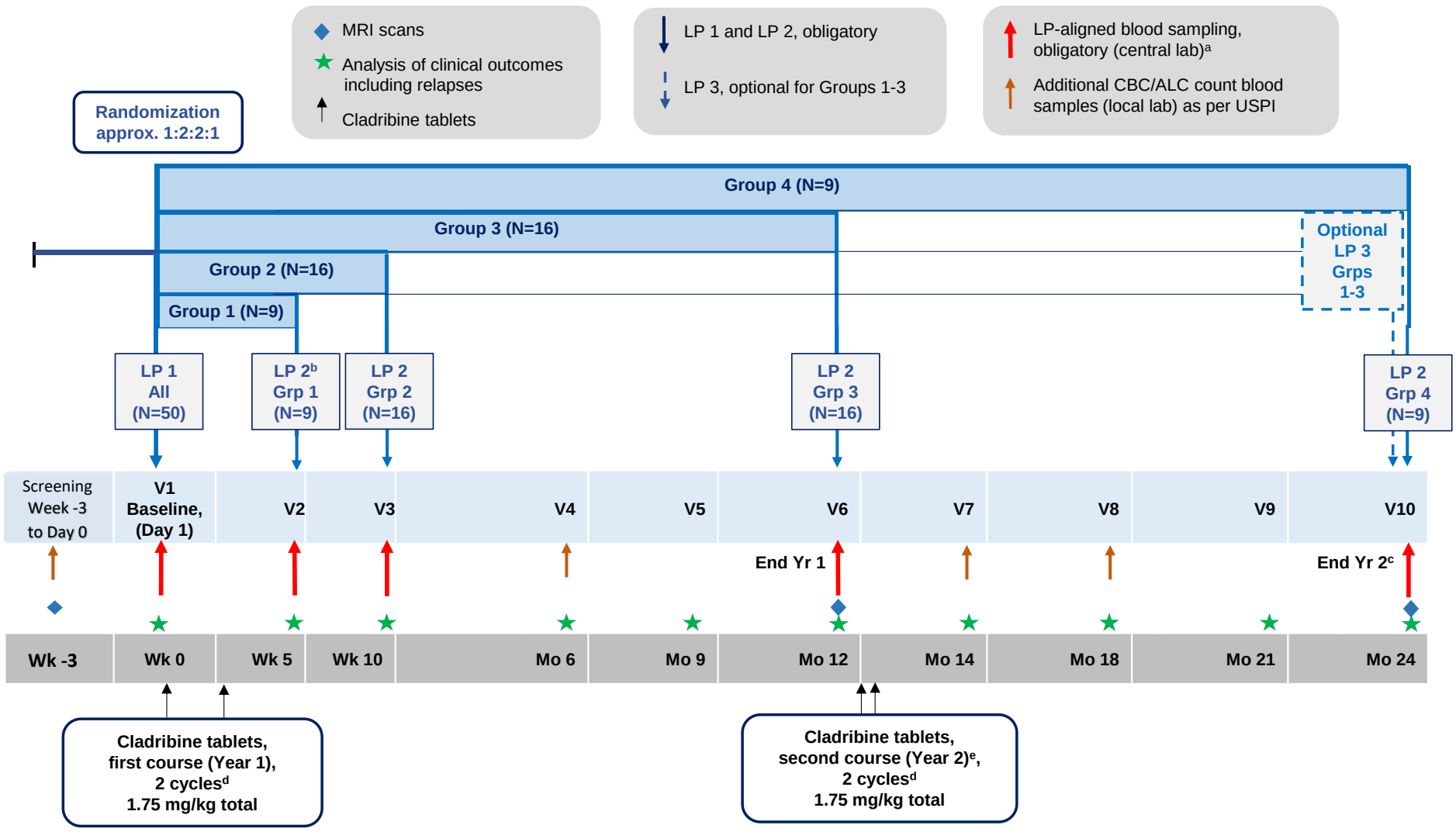
  In addition, all patients randomized to Groups 1-3 will have the option to volunteer for a third, optional LP at the end of Year 2, the timing of which would be aligned with the second LP of Group 4.

Data Monitoring Committee: Yes

1.2. Schema

Figure 1 presents a schematic of the exploratory study design. A detailed Schedule of Activities (SoA) is provided in Section 1.3.

Figure 1. Study Schema



Note: Optional, exploratory single cell RNA sequencing of CSF and blood cells may be performed locally at each site using CSF samples and LP-aligned blood samples obtained together with the samples for central assessment.

ALC=absolute lymphocyte count, CBC=complete blood count, CSF=cerebrospinal fluid, grp=patient group, LP=lumbar puncture, Mo=month, MRI=magnetic resonance imaging, N=number of participants, USPI=United States Prescribing Information, V=visit, Wk=week, Yr=year.

[a] For patients assigned to an LP, the LP-aligned blood sample needs to be obtained on the same day as the LP.

[b] For patients in Group 1, the LP and aligned blood samples need to be obtained on the same day of and following the last intake of the Week 5 cladribine tablet treatment course.

[c] If the start of the second treatment course needs to be delayed (e.g. to allow for lymphocyte recovery to ≥800 cells/µL), to maintain the protocol-specified time interval between start of cladribine tablets Year 2 treatment and all following assessments, Year 2 visits and procedures for this participant will be delayed accordingly (for up to 6 months).

[d] Each cycle consists of daily treatment over 4-5 consecutive days. The second cycle is administered 23 to 27 days after the last dose of the first course.

[e] The second treatment course is administered at least 43 weeks after the last dose of the first course, and can be delayed for up to 6 months.

1.3. Schedule of Activities (SoA)

Study Period	**Screening**	**Baseline**	**Treatment Course 1 (Year 1)**					**Treatment Course 2 (Year 2)**				
Visit	**Week –3 to Day 0**	**Day 1**	**Week 5[a]**	**Week 10**	**Month 6**	**Month 9**	**End Yr 1[b]**	**Month 14[c]**	**Month 18[c]**	**Month 21[c]**	**End Yr 2[c,d]**	**ED**
Visit Windows		**-**	**-**	**±7 d**	**± 14 d**	**± 14 d**	**-**	**±14 d**	**±14 d**	**±14 d**	**-**	
Informed consent	X											
Inclusion/exclusion crit.	X	X										
Demographics	X											
Medical history (incl. previous MS treatment)	X											
Questionnaire-Disease characteristics & symptoms	X											
Local safety laboratory: CBC with differential (ALC)[e]	X			[X][f]	X		X	X	X			
Check of all other eligibility criteria for cladribine tablets treatment[g]	X						X					
Body weight[h]		X					X					
Cladribine tablets dispensed		X					X					
Randomization to LP		X										
LP for CSF sampling[i,j]		X	[X][i,j]	[X][i]			[X][i]				[X][i]	
Cladribine concentration (central laboratory)		X	[X][i,j]									
Lymphocyte total and subsets, other immune cells (flow cytometry; local laboratory)		X	[X][i,j]	[X][i]			[X][i]				[X][i]	
Soluble biomarkers: NfL + exploratory (central laboratory)		X	[X][i,j]	[X][i]			[X][i]				[X][i]	
Single cell RNA sequencing (optional, local laboratory)		X	[X][i,j]	[X][i]			[X][i]				[X][i]	
CSF RNA central sample banking (optional)		X	[X][i,j]	[X][i]			[X][i]				[X][i]	

Study Period	**Screening**	**Baseline**	**Treatment Course 1 (Year 1)**					**Treatment Course 2 (Year 2)**				
Visit	**Week –3 to Day 0**	**Day 1**	**Week 5[a]**	**Week 10**	**Month 6**	**Month 9**	**End Yr 1[b]**	**Month 14[c]**	**Month 18[c]**	**Month 21[c]**	**End Yr 2[c,d]**	**ED**
Visit Windows		**-**	**-**	**±7 d**	**± 14 d**	**± 14 d**	**-**	**±14 d**	**±14 d**	**±14 d**	**-**	
LP-aligned blood sampling[j]		X	X[k]	X			X				X	
Cladribine concentration (central laboratory)		X	X[k]									
Lymphocyte total and subsets, other immune cells (flow cytometry; local laboratory		X	X[k]	X			X				X	
Soluble biomarkers: NfL + exploratory (central laboratory)		X	X[k]	X			X				X	
Single cell RNA sequencing (optional, local laboratory)		X	X[k]	X			X				X	
Peripheral blood RNA sample banking (optional)		X	X[k]	X			X				X	
Relapse evaluation[l]		X	X	X	X	X	X	X	X	X	X	X
Physical/neurological exam.	X	X			X		X		X		X	X
EDSS		X	X	X	X	X	X	X	X	X	X	X
MRI[m]	X						X				X	
Concomitant medication	**X**	X	X	X	X	X	X	X	X	X	X	X
Concomitant diseases/Medical history	**X**	X	X	X	X	X	X	X	X	X	X	X
Adverse events		X	X	X	X	X	X	X	X	X	X	X

Study Period	Screening	Baseline	Treatment Course 1 (Year 1)					Treatment Course 2 (Year 2)				
Visit	Week –3 to Day 0	Day 1	Week 5[a]	Week 10	Month 6	Month 9	End Yr 1[b]	Month 14[c]	Month 18[c]	Month 21[c]	End Yr 2[c,d]	ED
Visit Windows		-	-	±7 d	± 14 d	± 14 d	-	±14 d	±14 d	±14 d	-	

ALC=absolute lymphocyte count, CBC=complete blood count, CSF=cerebrospinal fluid, d=days, ED=early discontinuation visit, EDSS=Expanded Disability Status Scale, Gd+=gadolinium-enhancing, IWRS=Interactive Web Response System, LP=lumbar puncture, MRI=magnetic resonance imaging, MS=multiple sclerosis, USPI=United States Prescribing Information, Yr=Year.

[a] Visit to take place on the day of the last intake of cladribine tablets of the second cycle of Year 1 cladribine tablets treatment.

[b] End Year 1 Visit (with assigned LP and LP-aligned blood draw) to take place at least 43 weeks, but no later than 47 weeks, after the last dose of cladribine tablets Year 1 treatment.

[c] If the start of cladribine tablets Year 2 treatment needs to be delayed (e.g. to allow for lymphocyte recovery to ≥800 cells/µL), all following visits and procedures, including any LPs and LP-aligned blood samplings, will be postponed by the same period of time (for up to 6 months), to maintain the protocol-specified time interval between start of cladribine tablets Year 2 treatment and all following assessments.

[d] End Year 2 Visit (with assigned LP and LP-aligned blood draw) to take place at least 43 weeks, but no later than 47 weeks, after the last dose of cladribine Year 2 treatment.

[e] Safety hematology (see Appendix 2): CBC tests with differential to assess ALC and other hematologic parameters (local laboratory) are to be performed per the USPI. The ALC is to be assessed at least before initiating Year 1 and Year 2 treatment with cladribine tablets, and 2 and 6 months after the start of each treatment year. The ALC must be within normal limits of the local laboratory or assessed as normal by the investigator, before initiating treatment with cladribine tablets in Year 1, and need to be ≥800 cells/µL before initiating treatment in Year 2. If the lymphocyte count at Month 2 (Week 10) is <200 cells/µL, the ALC needs to be monitored monthly until Month 6.

[f] The Week 10 LP-aligned blood sample may be used for the USPI-required Month 2 safety hematology assessments.

[g] See Section 5.4 for required assessments. This includes safety biochemistry (see Appendix 2) and other laboratory screenings (e.g. for pregnancy, see Appendix 2).

[h] Body weight to be measured before each treatment course for calculating appropriate cladribine tablets dose.

[i] All study participants will receive an initial LP at baseline, and will then be randomized 1:2:2:1 to receive the second LP post-baseline at the end of Week 5, end of Week 10, end of Year 1, or end of Year 2. In addition, all study participants randomized to Groups 1-3 will have the option to volunteer for an optional third LP at the end of Year 2. Aliquots of CSF samples will be sent to local and central assessments.

[j] The Week 5 LP needs to take place on the same day following the intake of the last Week 5 cladribine tablet.

[k] For participants receiving an LP at the respective visit, the blood draws will take place on the same day. Aliquots of the blood samples will be distributed for central and local assessments.

[l] Procedure as outlined in Section 8.1.1.

[m] Includes standard sequences to evaluate T1 Gd+ and new or enlarging T2 lesions (central assessment) and R2t* echo contrast imaging of cerebral cortex, deep gray matter, and white matter to measure tissue integrity (centrally assessed at Washington University).

1.3.1. Schedule of Activities (SoA): Vaccine Sample (Optional)

Study Period	**Treatment Course (Year 1 or Year 2)**			
Vaccine Visit	**Week -3 to Day 0 Pre-Vaccine**[a,b]	**Day 1 Vaccine Obtained**	**Week 4 Post-Vaccine**	**Month 6 Post-Vaccine**
Visit Windows	**-21 d**	**-**	**±7 d**	±7 d
Blood Sampling[c] (optional, central laboratory)	X		X	X

d=days

a Pre-vaccine visit will occur within 21 days of obtaining a vaccine as part of standard of care

b Participants must have taken at least 1 dose of cladribine tablets prior to providing pre-vaccine blood sample

c Participants must consent to optional vaccination blood draw prior to providing blood samples

2. Introduction

Despite the recent approvals of several newer therapies, treatment and disease burden remain significant for patients with relapsing forms of multiple sclerosis (RMS). The ability to predict treatment response is limited, and there is an unmet need to identify potential predictive markers that might ultimately help to better predict clinical efficacy and safety outcomes of these new disease-modifying drugs (DMDs).

Cladribine is a synthetic chlorinated analog of the naturally occurring nucleoside deoxyadenosine. The oral formulation (cladribine 10 mg tablet; Mavenclad®) has been approved for the treatment of adult patients with highly active RMS in the European Union since August 2017. In the United States (US), the Food and Drug Administration (FDA) has recently approved cladribine tablets for the treatment of adult patients with RMS, to include relapsing-remitting MS (RRMS) and active secondary progressive disease. In the US, cladribine tablets are generally recommended for patients who have had an inadequate response to, or are unable to tolerate, an alternate drug indicated for the treatment of RMS. Based on individual patient assessment and shared decision-making, cladribine may be used as an initial treatment at the discretion of the investigator.

Complete information on the chemistry, pharmacology, efficacy, and safety of cladribine 10 mg tablets is available in the US Prescribing Information (USPI; EMD Serono, 2019).

2.1. Study Rationale

Increasing evidence suggests that in RMS, early signs of neuronal degeneration are dependent on inflammatory activity in the central nervous system (CNS). The levels of several biomarkers related to inflammation and neurodegeneration that are present in the cerebrospinal fluid (CSF) seem to increase with disease activity, and to decrease again towards normality during treatment with DMDs. Previous investigations of biomarkers have revealed effects of DMDs on the CSF profile of various immune cells and proteins that reflect the different aspects of the immunopathogenesis of multiple sclerosis (MS) (Novakova, 2017). For cladribine specifically, one study has shown that the proportion of MS patients expressing oligoclonal bands in the CSF was decreased after treatment with parenteral cladribine. The disappearance of the oligoclonal bands was associated with milder neurological disability after 10 years of follow-up (Rejdak, 2019). However, the effects of cladribine on any other CSF biomarkers relevant to RMS have not been evaluated to date.

The purpose of this exploratory, multicenter collaborative research Phase 4 study is to better understand the mechanism of action (MoA) of cladribine tablets by exploring their effect on CSF biomarkers relevant to RMS. The study is designed to generate hypotheses regarding the impact and relevance of cladribine tablet activity in the CSF. This will be done by assessing the CSF levels of lymphocyte subsets and other immune cells as well as the CSF levels of soluble immunological and neuronal injury markers in study participants with RMS before and during treatment with cladribine tablets. The association between CSF and peripheral (blood) levels of these markers, and their association with clinical outcomes, will be explored.

The study will be conducted at 5 selected sites throughout the US, based on the sites' experience of treating patients with RMS and conducting lumbar punctures (LPs). All study participants will

receive 2 yearly treatment courses with cladribine 10 mg tablets (Year 1 and Year 2; see Section 4.3).

Details on the rationale for the study design are provided in Section 4.2.

2.2. Background

Multiple Sclerosis

MS is a chronic, inflammatory, demyelinating CNS disease and is the most common cause of serious neurological disability in young adults (Przybek, 2015). With no definite cause established, MS has been hypothesized to result from a complex interaction between individual genetic susceptibility and environmental factors that act as triggers of a self-sustained autoimmune response (Noseworthy, 2000). With a peak of onset averaging at the age of 30 years in both genders, MS occurs predominantly in women with a female to male ratio of 3:1 (Kingwell, 2013). In the US, the incidence is estimated to be 3.2 per 100,000 patient years (Multiple Sclerosis International Federation, 2013). Based on a recent study by the US Multiple Sclerosis Prevalence Workgroup (MSPWG), an estimated 947,000 people in the US have MS (Wallin, 2017). Studies indicate that patients with MS show a nearly 3-fold increase in mortality relative to the general population, and about half of them are attributed to MS as the underlying cause of death (Scalfari, 2013).

MS is not characterized by a singular, well-defined clinical presentation common to all patients with MS. The symptoms, which include a combination of cognitive, sensory, and motor manifestations, vary among patients and throughout the disease course according to the affected CNS areas and the severity of the demyelinating attacks.

Approximately 85% of patients with MS initially present with RMS (Multiple Sclerosis International Federation, 2013), which is characterized by alternating periodic acute exacerbations of disease activity (relapses) and periods of remission, consisting of partial or complete recovery (Lublin, 2014).

Treatment and Management of RMS

Patients with RMS are recommended to receive treatment with DMDs to decrease the relapse rate and to slow down the accumulation of brain lesions made visible by magnetic resonance imaging (MRI). Numerous immunomodulatory agents with differing routes of administration and different MoA have been shown to have beneficial effects. FDA-approved medications for the treatment of RMS include the intravenous (IV) medications natalizumab, ocrelizumab, and alemtuzumab; the injected medications interferon (IFN) beta and glatiramer; and the oral medications dimethyl fumarate, teriflunomide, fingolimod, and cladribine tablets. Because they are not curative, most DMDs are typically continued indefinitely unless they are ineffective or not tolerated. There is currently no standard protocol to guide the choice of DMD for patients with MS. Rather, treatment decisions and medication choices are the result of a thorough risk-benefit analysis and consideration of disease activity, patient-specific factors, and drug-related factors (Pardo, 2017).

Cladribine Tablets

The purine analog cladribine was initially developed as a synthetic anti-neoplastic agent for the treatment of lymphoid malignancies. In cells with high levels of deoxycytidine kinase and low

levels of deoxynucleotidase (5'-NTase), in particular lymphocytes, cladribine prodrug is converted into its active triphosphate form, leading to selective depletion of dividing and non-dividing B and T cells.

Because of these actions on the immune system, a potential clinical utility in the treatment of MS was postulated, and initial clinical studies on MS were performed using a parenteral formulation. Based on observations from these, an oral cladribine formulation for the treatment of MS was developed by Serono, later Merck KGaA, Darmstadt, Germany. The unique (short) treatment schedule (2 treatment courses: Year 1 and Year 2) and oral route of cladribine, together with the efficacy benefits observed, represent a feasible treatment option for patients with RMS.

Three Phase 3 clinical trials for cladribine tablets have been completed (CLARITY, ORACLE MS and CLARITY EXTENSION), and data from these trials (Giovannoni, 2018; Leist, 2014; Giovannoni, 2010), as well as the interim long-term follow-up data from a prospective registry (the PREMIERE registry), have provided information on the efficacy and safety of cladribine tablets. In total, the follow-up consisted of >10,000 person-years of exposure for both cladribine tablets and placebo, with >8 years of follow-up for some patients.

In the CLARITY trial, patients with active RMS were randomized to placebo or cladribine tablets (3.5 or 5.25 mg/kg body weight) for 2 years; each dose showed significant benefits in rate of relapse, disability progression, and MRI measures (Giovannoni, 2010). Treatment with cladribine tablets in CLARITY produced efficacy improvements that were maintained in patients treated with placebo in the extension phase (Giovannoni, 2018). In clinical studies, 87% of participants treated with cladribine tablets experienced lymphopenia. The lowest absolute lymphocyte counts (ALCs) occurred approximately 2 to 3 months after the start of each treatment course and were lower with each additional treatment course. In patients treated with a cumulative dose of 3.5 mg per kg over 2 courses as monotherapy, 26% and 1% had nadir ALCs of <500 cells/µL and <200 cells/µL, respectively. At the end of the second treatment course, 2% of clinical study participants had an ALC of <500 cells/µL; the median time to recovery to ≥800 cells/µL was approximately 28 weeks. Herpes zoster infection is considered as an important identified risk of treatment with cladribine tablets. The observed incidence rates in the monotherapy oral cohort for herpes zoster infection were 0.86 adjusted adverse events (AEs) per 100 person-years for cladribine tablets 3.5 mg/kg vs. 0.20 for placebo, and for severe herpes zoster infection 0.9 vs. 0.5, respectively. The incidence of herpes zoster infection was higher during periods of Grade 3 or 4 lymphopenia compared to the time when the patients were not experiencing Grade 3 or 4 lymphopenia.

Incidence rates of the most common infections, including the most common severe infections, were similar between the placebo and the cladribine tablets exposed groups, with the exception of herpes zoster infection as described above. The incidence rates for infections, in general, were 24.93 vs. 27.05 adjusted AEs per 100 person-years (cladribine tablets 3.5 mg/kg monotherapy vs. placebo), and for severe infections 0.84 vs. 0.86, respectively. There was no evidence for an increased risk of opportunistic infections in patients treated with cladribine tablets (incidence rates were 1.08 and 1.17 for cladribine tablets 3.5 mg/kg monotherapy and placebo, respectively). However, tuberculosis has been defined as an important identified risk, with overall 3 cases observed in the cladribine tablets development program. All cases were reported prior to the implementation of mandatory pre-screening for tuberculosis; no cases occurred thereafter.

Treatment with cladribine tablets may increase the risk of malignancy. In controlled and extension clinical studies worldwide, malignancies occurred more frequently in cladribine-treated participants [10 events in 3,754 patient-years (0.27 events per 100 patient-years)], compared to placebo patients [3 events in 2,275 patient-years (0.13 events per 100 patient-years)] (EMD Serono, 2019).

A detailed description of the chemistry, pharmacology, efficacy, and safety of cladribine tablets is provided in the USPI (EMD Serono, 2019).

MS Biomarkers in the CSF

Intensive research is ongoing to look for cellular and soluble biomarkers in serum and CSF that are involved in the immunological and neurodegenerative processes of MS to better understand the pathogenesis of MS and the MoA of DMDs, with the ultimate goal to identify prognostic or predictive markers and the potential to guide optimal selection of immunomodulatory therapies in individual patients and monitor their efficacy.

Various types of immune cells related to the autoimmune-inflammatory processes occurring in MS have been assessed in the CSF of patients with MS and are discussed as potential biomarkers, including T- and B-lymphocytes, monocytes, and dendritic cells (Tumani, 2009). Assessing the levels of immune cells present in the CSF by flow cytometry is a potential tool to explore the biology of the inflammatory processes during MS directly in the intrathecal compartment. From such studies, it is known that patients with RRMS have increased CSF levels of B lymphocytes, activated T cells, plasmablasts and dendritic cells, and a decreased proportion of monocytes (Han, 2014).

Treatment with cladribine tablets is known to result in selective and transient reductions in cluster of differentiation (CD)19+ B lymphocytes and (CD3+) T lymphocytes within the peripheral circulation, including CD4+ T helper (Th) and CD8+ cytotoxic T cells (Comi, 2019). In this planned exploratory study, flow cytometry will be used to assess the levels of lymphocyte subsets and other immune cells present in the CSF before and at different time points during the treatment with cladribine tablets. Primary cellular markers assessed will be CD3+ T lymphocytes and CD19+ B lymphocytes. Secondary cellular markers will include T lymphocyte subsets such as CD4+ cells, CD8+ T cells, natural killer cells, plasmablasts, and other immune cells such as CD14+ cells.

Further, numerous soluble CSF markers of immunopathology, inflammation, glial function, and neurodegeneration (such as markers related to demyelination, oxidative stress, or axonal damage) are explored as potential biomarkers for MS (Alvermann, 2014; Avsar, 2012; Tumani, 2009). Of these, the neurofilament light chain (NfL) subunit of the neurofilament protein, which is a major component of the axonal cytoskeleton, has been extensively studied. NfL levels in the CSF seem to reflect the extent of neuroaxonal damage in MS patients and have been shown to strongly correlate with NfL serum levels (Kuhle, 2019; Novakova, 2017). NfL levels in both compartments are discussed as potential biomarkers for monitoring disease activity and treatment outcomes (Hyun, 2019; Kuhle, 2019; Cai, 2018). Elevated CSF and blood concentrations of NfL have been found to correlate with an increase in the number of relapses, disability worsening, MRI disactivity, and brain volume loss in MS (Kuhle, 2019). Of note, the DMD fingolimod was shown to significantly reduce NfL plasma levels of MS patients when compared to placebo (Kuhle, 2019).

For cladribine specifically, a single study has shown that the proportion of MS patients with oligoclonal bands expression in the CSF decreases after treatment with parenteral cladribine (Rejdak, 2019). The effects of cladribine tablets on other cellular or soluble CSF biomarkers has not been evaluated to date.

In the CLOCK-MS study, the NfL levels in the CSF assessed before and at different time points during treatment with cladribine tablets will therefore be the primary soluble marker. Other soluble CSF and serum markers related to inflammation (e.g. CSF levels of immunoglobulin [Ig] G and IgM, and oligoclonal bands) and neurodegeneration (e.g. serum levels of neurofilament heavy chain [NfH]) will be assessed as exploratory markers. Up to now, the search for potential biomarkers in MS has been largely hypothesis-driven. Single soluble molecules or sets of molecules have been examined, based on their presumed association with key aspects of MS disease pathophysiology. More recent technologies such as RNA profiling allow for an unbiased screen to identify new candidate biomarkers. Such candidate biomarkers can then be further investigated with hypothesis-driven approaches to explore their role in the pathogenesis of MS and if they might be used as surrogate endpoints for treatment efficacy. Therefore, this study also includes the option for single cell RNA profiling and for RNA sample banking.

2.3. Benefit/Risk Assessment

Treatment with Cladribine Tablets

All study participants will receive treatment with cladribine 10 mg tablets at the cumulative dose of 3.5 mg/kg, divided into 2 yearly treatment courses.

The uniquely short treatment schedule and oral route of cladribine have important implications for adherence to treatment. Considering the robust and sustained efficacy benefits observed, cladribine tablets represent a feasible treatment option for patients with RMS.

Cladribine tablets may increase the risk of malignancy. Severe (Grade ≥3) lymphopenia, herpes zoster infection, and tuberculosis are considered key identified risks of treatment with cladribine 10 mg tablets. Risk mitigation procedures will be followed as summarized in Table 1.

More detailed information about the known and expected benefits and risks and reasonably expected AEs of treatment with cladribine 10 mg tablets are provided in the Investigator's Brochure and the USPI (EMD Serono, 2019).

Lumbar Punctures

Each participant will be assigned to 2 protocol-required LPs. LPs are widely used to obtain CSF for diagnostic purposes, such as detection of infections, inflammation, malignancies involving the central and peripheral nervous systems, and other CNS disorders including Alzheimer's disease or MS (Doherty, 2014; Menéndez-González, 2014). When performed correctly, the procedure is well tolerated and accepted with a low complication rate (Engelborghs, 2017).

In this study, sites will be using the same LP procedure they routinely apply for diagnostic purposes (for example, use of the same anesthetics). The use of non-cutting needles is preferred to prevent post-LP headache. If an epidural CSF leak is suspected post-LP, an epidural blood patch is encouraged, which should be placed by a licensed and trained physician.

The most common adverse effects associated with LPs are post-LP headache and back pain (Engelborghs, 2017). In a large multicenter European study with 3868 participants (BIOMARKAPD), the overall AE rate was 31%, with 19% of patients reporting any headache and 17% of patients reporting any back pain (Duits, 2016). Other known adverse effects associated with LPs include dizziness or discomfort, and bleeding or bruising at the puncture site. Rare complications include brain or spinal hemorrhage, allergic reactions to the lidocaine used for local anesthesia, and infections. The risk for infections can be minimized by the use of sterile gloves and thorough disinfection of the lumbar region.

It has been discussed that many patients, in particular in the US, may perceive a planned LP as frightening and invasive (Howell, 2016; Menéndez-González, 2014). However, in a recent study in 237 younger and older adults from the US with limited first-hand LP experience, only 1 of 4 respondents viewed the LP as a frightening invasive procedure, and the majority of respondents (89%) were willing to undergo LP for medical reasons (Tsvetkova, 2017). The authors concluded that Americans (born in the US or abroad) are not resistant to LPs if they provide useful information to improve their health, although there may be more limited enthusiasm to undergo LPs solely for research purposes.

Several studies have evaluated the feasibility of LPs to evaluate CSF biomarkers in clinical studies, mainly in patients with Alzheimer's disease, and the procedure has generally been characterized as safe and acceptable. The authors of the large BIOMARKAPD study concluded that the overall risk of complications is relatively low if known LP risk factors are taken into account (Duits, 2016).

A similar study in 689 patients with Alzheimer's disease from Spain also concluded that LPs can be safely performed to study CSF biomarkers (Alcolea, 2014). Again, headache was the most frequent complication (24.8%), but was severe in only 2.5% of patients. Back pain was reported by 16.1% of patients. No major complications were reported.

Other Study Procedures

All other study procedures, e.g. MRI assessments and blood draws, are established procedures usually part of the routine practice when managing MS patients. Therefore, they do not pose any additional risk to the participants.

2.3.1. Risk Assessment

Table 1. Risk Assessment

Potential risk of clinical significance	Summary of data	Mitigation strategy
Study Intervention 1: Cladribine 10 mg tablets		
Severe (Grade ≥3) lymphopenia	Refer to: IB Section 5.2.1.8.1 USPI (EMD Serono, 2019)	All study participants will have their ALC monitored before initiating cladribine tablets in Year 1 and Year 2, as well as 2 and 6 months after the start of treatment in each treatment course, or as clinically indicated and in accordance with the USPI. If the ALC drops to <500 cells/µL, the study participant should be actively monitored until values increase.
Herpes zoster infection	Refer to: IB Section 5.2.1.8.2 USPI (EMD Serono, 2019)	Herpes zoster vaccination will be recommended for patients who demonstrate lack of antibody reactivity to varicella zoster virus prior to initiation of cladribine tablets. Anti-herpes prophylaxis will be considered during the time a patient is experiencing Grade 4 lymphopenia.
Tuberculosis	Refer to: IB Section 5.2.1.8.2 USPI (EMD Serono, 2019)	Study participants will be monitored for signs and symptoms of any infection and anti-infective treatment will be initiated as clinically indicated. Interruption or delay of cladribine tablets therapy may be considered until proper resolution of the infection.
Other infections	Refer to: IB Section 5.2.1.8.2 USPI (EMD Serono, 2019)	
Malignancies	Refer to: IB Section 5.2.1.8.3 USPI (EMD Serono, 2019)	Patients with current malignancies will be excluded. In case of prior or increased risk of malignancy, risks will be evaluated vs. expected benefit on an individual basis

Potential risk of clinical significance	Summary of data	Mitigation strategy
Study Intervention 2: Lumbar puncture		
Two LPs required for each participant	Refer to literature: Engelborghs, 2017 Tsvetkova, 2017 Duits, 2016 Howell, 2016 Alcolea, 2014	All LPs will be performed by licensed and trained physicians. Local sterilizing procedures per standard practice will be performed, and sterile gloves and drapes will be used, to minimize the risk of infections. Eligibility for LP will be checked prior to each LP. Use of non-cutting needles is preferred to prevent post-LP headache. If a post-LP epidural CSF leak is suspected, an epidural blood patch is encouraged, which should be placed by a licensed and trained physician.

ALC=absolute lymphocyte count, IB=Investigator's Brochure, LP=lumbar puncture, USPI=United States Prescribing Information.

2.3.2. Benefit Assessment

All study participants will receive the approved treatment regimen with cladribine 10 mg tablets, at a cumulative dose of 3.5 mg/kg divided into 2 yearly treatment courses. This regimen showed robust and sustained clinical benefits in patients with RMS (see Section 2.2). In addition, individual participants may benefit from the medical evaluations and assessments of their disease during the study, including the LPs, neurological examinations, and MRI evaluations per the SoA (Section 1.3.

Finally, the participants will contribute to the process of generating hypotheses regarding potential predictive CSF and serum markers that might help to better predict the clinical efficacy and safety of treatment with cladribine tablets for individual patients in the future.

2.3.3. Overall Benefit:Risk Conclusion

Taking into account the measures taken to minimize risk to participants participating in this study, the potential risks identified in association with cladribine tablets and the LP procedures are justified by the anticipated benefits that may be afforded to participants with RMS.

3. Objectives and Endpoints

Objectives	Endpoints (Outcome Measures)
Primary	
• To evaluate changes in the CSF levels of lymphocyte subtypes (CD3+ T lymphocytes, CD19+ B lymphocytes) and markers of neuronal injury (NfL) during treatment with cladribine tablets in patients with RMS	• Change in CSF levels of CD3+ T lymphocytes, CD19+ B lymphocytes, and NfL from baseline to second LP (end of Week 5, end of Week 10, or end of Year 1, and/or end of Year 2[a])
Secondary	
• To evaluate changes in the CSF levels of other lymphocyte subtypes and immune cells during treatment with cladribine tablets	• Change in CSF levels of CD4+ and CD8+ cells, plasmablasts, NK cells, Tregs, and CD14+ cells from baseline to second LP (end of Week 5, end of Week 10, or end of Year 1, and/or end of Year 2[a])
• To assess cladribine CSF and serum levels during treatment with cladribine tablets	• Cladribine CSF and serum concentrations at baseline and second LP (end of Week 5 time point only)
• To assess the peripheral blood levels of lymphocyte subtypes, other immune cells, and soluble immunological and neuronal injury markers during treatment with cladribine tablets	• Change in peripheral blood levels of CD3+, CD4+ CD8+, CD19+, Th1, Th2, and Th17 cells, Tregs, plasmablasts, NK cells, CD14+ cells, and NfL levels from baseline to second LP (end of Week 5, end of Week 10, or end of Year 1, and/or end of Year 2[a])
• To assess clinical outcomes, based on the ARR and EDSS, during treatment with cladribine tablets	• ARR over 24 months of treatment with cladribine tablets (accompanied by 95% CIs) (see below for the definition of relapse) • Change in EDSS scores from baseline at Month 12 and Month 24 • 6-month confirmed EDSS progression

Objectives	Endpoints (Outcome Measures)
• To evaluate MRI findings during treatment with cladribine tablets, using: o Standard sequences to evaluate T1 Gd+ and new or enlarging T2 lesions, and o Quantitative gradient echo contrast imaging (R2t*) of cerebral cortex, deep gray matter, and white matter to measure tissue integrity	• MRI findings at Month 12 and Month 24 compared to baseline: o Proportion of participants with no Gd+ lesions o Proportion of participants with no new or newly enlarging T2 lesions o Mean number of Gd+ lesions per participant o Mean number of new or newly enlarging T2 lesions per participant • Change in R2t* in white matter and gray matter
• To collect safety data (adverse events, hematology) during treatment with cladribine tablets	• Occurrence of TEAEs, SAEs, and TEAEs leading to treatment discontinuation • Absolute lymphocyte count assessed per USPI • Complete blood cell count as assessed per routine clinical practice
Tertiary/Exploratory	
• To evaluate changes in the CSF and peripheral blood levels of other cellular markers during treatment with cladribine tablets	• Change in CSF and serum levels of other cell types of interest (as identified during the course of the study) from baseline to second LP (end of Week 5, end of Week 10, or end of Year 1, and/or end of Year 2[a])
• To assess changes in antibody titers during treatment with cladribine tablets after receiving a vaccine.	• Change in anti-immunogen antibody titers at 1 month and 6 months post-immunization compared to pre-immunization

Objectives	Endpoints (Outcome Measures)
• To identify potentially predictive CSF or serum biomarkers, e.g. by: o Serial assessment of a broad spectrum of CSF and serum biomarkers related to inflammation (cytokines, chemokines, others) or neurodegeneration o Optional serial single cell RNA sequencing o Multifactorial modeling approaches to explore the association between changes in all primary, secondary, and exploratory markers assessed and clinical efficacy and safety outcomes	• Change in CSF and serum levels of the followinginflammation-related biomarkers compared to baseline: IgA, IgG, IgM, oligoclonal bands, CXCL12, CXCL13, CCL19, and CCL21 • Change from baseline in serum levels only of the following inflammation-related biomarkers: IFNγ, IL-10, IL-4, osteopontin, and complement levels • Change in serum levels of several biomarkers related to neuronal injury or degeneration, including, but not limited to NfH, tau, isoprostanes • Optional: characterization of single cell RNA sequences of blood and CSF cells

[a] Data from any third, optional end of Year 2 LP will be included with the data from protocol-required end of Year 2 LPs.

ARR=annualized relapse rate, CD=cluster of differentiation, CI=confidence interval, CSF=cerebrospinal fluid, EDSS=Expanded Disability Status Scale, Gd+=gadolinium-enhancing, IFN=interferon, Ig=immunoglobulin, IL=interleukin, LP=lumbar puncture, NfH=neurofilament heavy chain, NfL=neurofilament light chain, MRI=magnetic resonance imaging, NK cells=natural killer cells, Tregs= regulatory T cells, RMS=relapsing forms of multiple sclerosis, SAE=serious adverse event, TEAE=treatment-emergent adverse event, Th cell=T helper cell, USPI=United States Prescribing Information.

Definition of Clinical Relapse of MS

One of the secondary objectives of this exploratory study is to assess the annualized relapse rate (ARR). For this study, a clinical event will be defined as clinical relapse of MS if ALL 3 following criteria are met:

1. Neurological abnormality, either newly appearing or re-appearing, that
    a. Is separated by at least 30 days from onset of a preceding clinical event, AND
    b. Lasts for at least 24 hours
2. Absence of fever or known infection (fever defined as temperature >37.5ºC [99.5ºF] measured axillary, orally, or intrauricular)
3. Objective neurological impairment, correlating with the participant's reported symptoms, defined as either
    a. Increase of at least 1 of the functional system (FS) scores of the Expanded Disability Status Scale (EDSS), OR
    b. Increase of the total EDSS score.

Note: The occurrence of paresthesia, fatigue, mental symptoms, and/or vegetative symptoms without any additional symptom will not be classified as an MS clinical relapse.

Section 8.1.1 describes the procedure for relapse evaluation at each visit.

4. Study Design

4.1. Overall Design

This will be an open-label, randomized, multicenter collaborative research Phase 4 biomarker study, designed to generate hypotheses to better understand the MoA of cladribine tablets in RMS.

The total duration of this interventional study will be 2 years for each participant. Approximately 50 study participants with RMS who meet the criteria for treatment with cladribine tablets per the approved USPI, and who are willing and able to receive LPs, are planned to be enrolled and randomized at 5 centers in the US. All study participants will receive treatment with cladribine 10 mg tablets during Year 1 and Year 2 according to the approved USPI (EMD Serono, 2019). Cladribine 10 mg tablets will be provided as commercial material.

After an initial 3-week screening period, all eligible study participants will undergo an initial LP and then initiate treatment with cladribine tablets at baseline (Day 1). Study participants will be randomized 1:2:2:1 (approximately 9:16:16:9 participants planned) to receive a total of 2 LPs according to 1 of the 4 following schedules:

1. Group 1: Baseline (Day 1) and end of Week 5 (LP to take place same day following intake of the last cladribine tablet of the first treatment course), or
2. Group 2: Baseline and end of Week 10, or
3. Group 3: Baseline and end of Year 1 (at least 43 weeks, but no later than 47 weeks, after the last tablet of the second cycle of cladribine Year 1 treatment), or
4. Group 4: Baseline and end of Year 2 (at least 43 weeks, but no later than 47 weeks, after the last tablet of the second cycle of cladribine Year 2 treatment).

   Note: If the start of cladribine tablets Year 2 treatment needs to be delayed (e.g. to allow for lymphocyte recovery to ≥800 cells/µL), the end of Year 2 LP will be postponed by the same period of time (for up to 6 months), to maintain the protocol-specified time interval between start of cladribine tablets Year 2 treatment and the second LP.

   In addition, all study participants randomized to Groups 1-3 can volunteer for a third, optional LP at the end of Year 2, the timing of which would be aligned with the second LP of Group 4.

Blood sampling will be aligned with the timing of the LPs (see Figure 1 and SoA in Section 1.3). All study participants will have LP-aligned blood samples taken at baseline (on the same day as the baseline LP), and post-baseline at Week 5, end of Week 10, Year 1, and Year 2). If an LP is scheduled, the LP-aligned blood sampling will be done on the same day.

Samples of the CSF- and the LP-aligned total blood samples will be sent to the central laboratory for cladribine concentration measurement (Section 8.5) and for soluble biomarker assessments (Section 8.8.2); cellular biomarkers will be assessed locally (Section 8.8.1). All cellular markers and NfL levels will be assessed in both CSF and peripheral blood. Other soluble biomarkers will be assessed in CSF or serum only, as appropriate (Section 8.8.2).

In addition, physicians are expected to monitor participants' ALC in accordance with the cladribine tablets USPI and to follow any other recommended monitoring and guidance as specified in the USPI (EMD Serono, 2019). This requires additional routine blood samplings to

comply with the USPI, i.e. complete blood count (CBC) with differential at least at screening, Month 6, Month 14, and Month 18. The USPI-required Month 2 sample may be covered by the LP-aligned Week 10 sample. The additional routine blood samples can be evaluated locally for ALC and other CBC parameters per routine practice at the respective site.

Clinical outcomes (Sections 8.1.1 and 8.1.2) will be assessed at baseline, Week 5, Week 10, Months 6, 9, and 12 during Year 1, and at Months 14, 18, 21, and 24 during Year 2 (see SoA in Section 1.3).

MRI scans will be performed at screening, Month 12, and Month 24. All scans will be assessed centrally (Section 8.1.3).

Optional, exploratory single cell RNA sequencing of CSF and blood cells may be performed locally at each site, if sufficient CSF and blood sample material is available (Section 8.8.3).

In addition, RNA samples will be stored for future analysis from all study participants who consent to an optional RNA sample banking (Section 8.8.4).

Participants receiving a vaccine as standard of care and have started cladribine tablet treatment, will be given the opportunity to provide additional blood samples to examine antibody titers (see Section 8.9).

Visit schedules for all laboratory, efficacy, and safety assessments are detailed in the SoA (see Section 1.3). If the start of cladribine tablets Year 2 treatment needs to be delayed (e.g. to allow for lymphocyte recovery to ≥800 cells/µL), all following visits and procedures, including any LPs and LP-aligned blood samplings, will be postponed by the same period of time (for up to 6 months), to maintain the protocol-specified time interval between start of cladribine tablets Year 2 treatment and all following assessments.

4.2. Scientific Rationale for Study Design

The main rationale for this exploratory, multicenter collaborative research Phase 4 study is to better understand the MoA of cladribine tablets in association with CNS biomarkers in patients with RMS. The study is intended to generate hypotheses regarding the cladribine tablet activity within the CSF by assessing the CSF levels of lymphocyte subsets, neuronal injury markers, other immune cells and soluble immunological markers, and their association with the corresponding blood markers and the clinical outcomes.

All study participants will receive treatment with cladribine 10 mg tablets according to the approved regimen per the USPI (EMD Serono, 2019), and they will be followed up thoroughly during the study.

No placebo or active control arm is needed for this type of exploratory research. The sample size of approximately 50 study participants is based on the feasibility of enrollment (see Section 9.2), and no formal hypotheses will be tested.

A maximum CSF volume of 30 mL will be collected per LP (see Table 5). Per the current LP consensus guidelines, the collection of up to 30 mL of CSF is well tolerated and safe, and is advised as an acceptable maximum (Engelborghs, 2017).

Of the large set of cellular and soluble CSF biomarkers that will be assessed in this exploratory study, the 3 markers considered most important (CD3+ T lymphocytes, CD19+ B lymphocytes, and NfL level changes in the CSF) have been defined as primary endpoints.

To evaluate the changes for all the cellular and soluble markers assessed in the CSF is of key importance to obtain CSF markers before and at different post-baseline time points after the initiation of treatment with cladribine tablets.

The following post-baseline time points were considered as most important:

- End of Week 5: expected "maximum" concentrations of cladribine concentrations in the CSF (based on the serum pharmacokinetics)
- End of Week 10: expected "nadir" time for lymphocyte and monocyte quantities in CSF
- End of Year 1 and end of Year 2: time points needed to assess if the effects of cladribine tablets on CSF markers are maintained at the end of each treatment course.

For practical reasons, it was decided that individual study participants should not be required to undergo more than 2 LPs, an initial LP at baseline and a second LP at a pre-specified time point post-baseline. Study participants will be randomized to receive the second LP at 1 of the 4 important post-baseline LP time points (see Section 1.3). The 1:2:2:1 randomization ratio was chosen to have data from an appropriate number of study participants for those 2 LP time points with key importance for the primary objective (end of Week 10 and end of Year 1).

4.3. Justification for Dose

All study participants will receive treatment with cladribine 10 mg tablets at the recommended cumulative dose of 3.5 mg/kg, divided into 2 yearly treatment courses (1.75 mg/kg per treatment course). This regimen corresponds to the recommended dosage per the USPI (EMD Serono, 2019).

Each treatment course is divided into 2 treatment cycles:

- Administration of first treatment course (Year 1 treatment):
  - First cycle: starts on Day 1 of the study
  - Second cycle: administered 23 to 27 days after the last dose of first cycle.
- Administration of second treatment course (Year 2 treatment):
  - First cycle: administered at least 43 weeks after the last dose of Year 1 treatment
  - Second cycle: administered 23 to 27 days after the last dose of first cycle of Year 2 treatment.

The cycle dosage will be administered as 1 or 2 cladribine 10 mg tablets daily over 4 or 5 consecutive days.

Follow the guidance from the approved USPI, the treatment course in Year 2 can be delayed for up to 6 months, to allow for lymphocyte recovery. If lymphocyte recovery takes more than 6 months, the participant should not receive cladribine tablets anymore. In addition, Year 2 treatment should also be delayed in case of latent infections (in particular tuberculosis, hepatitis B or C) until the infection has been adequately treated.

Therefore, this study allows to delay the start of cladribine tablets Year 2 treatment for up to 6 months, to allow for lymphocyte recovery or treatment of latent infections. All following visits and procedures, including any LPs and LP-aligned blood samplings, will be postponed by the same period of time (for up to 6 months) for such participants, to maintain the protocol-specified time interval between start of cladribine tablets Year 2 treatment and all following assessments. If recovery/treatment is not complete after the 6 month delay, the participant will be discontinued from the study.

Refer to Section 6.6 for details on the distribution of the total dose and the number of tablets to be taken by weight range.

4.4. End of Study Definition

A participant has completed the study if he/she has completed all study parts, including the last visit and/or the last scheduled procedure at the end of Year 2 as shown in the SoA (Section 1.3).

The end of the study is defined as the date of the last participant's last visit and/or the last schedule procedure at the end of Year 2 as shown in the SoA (Section 1.3).

Once participants have concluded the study, they will continue to be treated as part of their usual practice and care.

5. Study Population

5.1. Inclusion Criteria

Participants are eligible to be included in the study only if all of the following criteria apply:

Age

1. Participant must be 18 to ≤65 years of age inclusive, at the time of signing informed consent.

Type of Participant and Disease Characteristics

2. Participants who
   - Have RMS (to include RRMS and active secondary progressive MS)
   - Are willing and able to receive at least 2 LPs
   - Have an EDSS of 0 to ≤ 5.5 during the screening period
   - Had at least 1 relapse or 1 gadolinium-enhancing (Gd+) or 1 new or enlarged T2 lesion in the last 12 months
   - Have ALCs within normal range of the local laboratory or assessed as normal by the investigator, within the 3-week screening period (repeated tests and re-screening allowed) and meet all other eligibility criteria for cladribine tablets treatment (see Section 5.4).

Sex

3. Male and female

   In females of reproductive potential, pregnancy should be excluded before initiation of cladribine tablets Year 1 and cladribine tablets Year 2 treatment (see Section 8.3.5).

   Contraceptive use by men or women should be consistent with US regulations regarding the methods of contraception for those participating in clinical studies, and with the approved cladribine tablets USPI (EMD Serono, 2019).

   Males and females of reproductive potential should use effective contraception during cladribine tablet dosing and for at least 6 months after the last dose in each treatment course. Women using systemically acting hormonal contraceptives should add a barrier method during treatment with cladribine tablets and for at least 4 weeks after the last dose in each treatment course.

   Women who become pregnant during treatment with cladribine tablets should discontinue treatment and will be discontinued from the study (see Section 8.3.5).

Informed Consent

4. Capable of giving signed informed consent as described in Appendix 1, which includes compliance with the requirements and restrictions listed in the Informed Consent Form (ICF) and in this protocol.

Prospective approval of protocol deviations to recruitment and enrollment criteria, also known as protocol waivers or exemptions, is not permitted.

Before performing any study assessments that are not part of the participant's routine medical care, the investigator will confirm that the participant or the participant's legal representative has provided written informed consent, as indicated in Appendix 1.

5.2. Exclusion Criteria

Participants are excluded from the study if any of the following criteria apply:

Medical Conditions

1. Have any contraindication for LP or any other comorbid conditions that preclude participation, including, but not limited to:
   - Skin infection at or near the site of the LP
   - Suspicion of increased intracranial pressure due to a cerebral mass
   - Uncorrected coagulopathy or concurrent use of anti-coagulants
   - Evidence of recent spine trauma
2. Have current malignancy
3. Are infected with human immunodeficiency virus (HIV)
4. Have active chronic infections (e.g. hepatitis or tuberculosis)
5. Have signs or symptoms suggestive of progressive multifocal leukoencephalopathy (PML) in MRI
6. Have a history of hypersensitivity to cladribine or to any of the excipients listed in the cladribine tablets USPI
7. Allergy or hypersensitivity to gadolinium and/or any other contraindication to perform an MRI
8. Have any other comorbid conditions that preclude participation.

Prior/Concomitant Therapy

9. Have been previously treated with cladribine in any dosing form (IV, subcutaneous, or oral)
10. Have previously been treated with rituximab, ocrelizumab, alemtuzumab, or daclizumab
11. Have received treatment with natalizumab during the last 6 months
12. Are currently receiving immunosuppressive or myelosuppressive therapy with e.g. methotrexate, cyclophosphamide, cyclosporine or azathioprine, or chronic treatment with systemic corticosteroids
13. Have received treatment with immunosuppressive or myelosuppressive therapy during the last 6 months
14. Have received chronic treatment with systemic corticosteroids during the last 4 weeks

Diagnostic Assessments

15. Have moderate or severe hepatic impairment (Child-Pugh score >6)
16. Have moderate or severe renal impairment (creatinine clearance <60 mL per minute).

Other Exclusions

17. Are pregnant or unwilling or unable to use effective contraception during cladribine tablets dosing and for 6 months after the last dose in each treatment course

18. Are intending to breastfeed on a cladribine tablet treatment day and/or during the 10 days after the last cladribine tablet dose.

5.3. Screen Failures

Screen failures are defined as participants who consent to participate in the clinical study ("enrolled") but are not subsequently randomly assigned to an LP schedule, e.g. because they do not meet all eligibility criteria for starting treatment with cladribine tablets.

A minimal set of screen failure information will be collected for screen failures to ensure transparent reporting of screen failure participants per the Consolidated Standards of Reporting Trials (CONSORT) publishing requirements, and to respond to queries from regulatory authorities. This minimal information includes demography, screen failure details, eligibility criteria, and any serious adverse event (SAE).

If a participant does not meet the laboratory requirements at the initial test during the screening period, the test can be repeated at any time during the screening period (this would not be considered as re-screening). In addition, re-screening is permitted for participants who do not meet the laboratory requirement at the end of the initial screening period. Re-screening can occur at ≥3 weeks after the initial screening. Of note, if a patient requires a repeated laboratory test, all other screening assessments used to confirm eligibility may need to be repeated if they have occurred more than 3 weeks prior to the scheduled cladribine tablets dosing visit. The need to repeat other screening assessments will be at the discretion of the Investigator.

Re-screened participants should be assigned the same participant number as used for the initial screening.

5.4. Criteria for Initiating and Continuing Treatment with Cladribine Tablets

All criteria for initiating the first and second course of cladribine tablets treatment (Year 1 and Year 2 treatment) have to be followed as outlined below.

Lymphocyte Counts

To initiate the first course of cladribine tablets (Year 1 treatment), and to continue treatment during Year 2, participants need to meet the following ALC criteria:

- The ALC has to be within normal range of the local laboratory or assessed as normal by the investigator, before initiating treatment in Year 1. Results must be obtained within 3 weeks of receiving cladribine tablets.
- The ALC has to be ≥800 cells/µL before initiating treatment in Year 2. If necessary, the start of cladribine tablets Year 2 treatment can be delayed for up to 6 months to allow for lymphocyte recovery. If this recovery takes more than 6 months, the participant will be discontinued from cladribine tablets treatment and from the study.
- If the ALC drops to values <500 cells/µL, the participant should be monitored for signs and symptoms suggestive of infections, including herpes infections. In case of infections, interruption or delay of cladribine tablets should be considered (see Section 6.6).

- If the ALC at Month 2 (end of Week 10 visit) drops to values <200 cells/µL (Grade 4 lymphopenia), the ALC needs to be monitored monthly until Month 6, and anti-herpes prophylaxis should be administered.

Infections

Screening tests for HIV, tuberculosis, and hepatitis B and C have to be performed before initiating Year 1 and Year 2 treatment with cladribine tablets, within 3 weeks of receiving the first dose for each treatment course. HIV infection, active tuberculosis, and active hepatitis must be excluded before initiation of each treatment course of cladribine tablets.

Participants also have to be evaluated for any acute infections. In case of an acute infection, the start of Year 2 treatment with cladribine tablets can be delayed (for up to 6 months in this study), until the infection is fully controlled. If this goal cannot be reached within 6 months, the participant should be discontinued from the study.

In case of latent infections (tuberculosis, hepatitis B, or hepatitis C), the start of cladribine tablets Year 2 treatment should be delayed until the infection has been adequately treated. If this goal cannot be reached within 6 months, the participant should be discontinued from the study.

For participants who have negative test results for antibodies to varicella zoster virus, vaccination is recommended prior to the initiation of treatment with cladribine tablets. Live-attenuated or live vaccines should be administered at least 4 to 6 weeks prior to starting cladribine tablets, to allow for the full effect of vaccination to occur.

Vaccinations

All immunizations should be administered according to immunization guidelines prior to starting treatment with cladribine tablets. Live-attenuated or live vaccines should be administered at least 4 to 6 weeks prior to starting treatment with cladribine tablets.

Hepatic Function

Treatment with cladribine tablets should not be initiated in participants with moderate to severe hepatic impairment. Serum aminotransferase, alkaline phosphatase, and total bilirubin levels should be evaluated within 3 weeks of initiating treatment in Year 1 and Year 2. Treatment should not be initiated in participants with laboratory values suggestive of moderate or severe hepatic impairment as determined by the investigator. Initial cladribine tablet treatment should not be initiated in participants with a Child-Pugh score > 6 (see Section 5.2).

If a participant develops clinical signs, including unexplained liver enzyme elevations or symptoms suggestive of hepatic dysfunction (e.g. unexplained nausea, vomiting, abdominal pain, fatigue, anorexia, or jaundice and/or dark urine), serum transaminases and total bilirubin should be measured promptly and treatment with cladribine tablets should be interrupted or discontinued, as appropriate.

Renal Function

Year 1 and Year 2 treatment with cladribine tablets should not be initiated in participants with moderate or severe renal impairment (creatinine clearance <60 mL per minute). Serum creatinine levels need to be assessed within 3 weeks prior to initiation of Year 1 and Year 2 treatment.

Other Criteria

In addition, the following criteria need to be met at the start of Year 1 and Year 2 treatment with cladribine tablets, within 3 weeks of initiating treatment:

- Standard cancer screening guidelines reviewed with participant; medical records reveal no current malignancy
- Baseline MRI reveal no signs and symptoms of PML
- Pregnancy is excluded, and will be prevented by the use of effective contraception during cladribine tablet dosing and for 6 months after the last dose in each treatment course.

5.5. Eligibility for Lumbar Punctures

Participants are NOT eligible to receive their assigned and protocol-required baseline or second LP or the third optional LP at the end of Year 2, if they meet any of the following exclusion criteria at the respective LP visit:

- Skin infection at or near the site of the LP
- Suspicion of increased intracranial pressure due to a cerebral mass
- Uncorrected coagulopathy or concurrent use of anti-coagulants
- Evidence of recent spine trauma
- Any other comorbid condition that precludes an LP.

  Coagulation status and platelet count should be checked before the LP, in patients at an increased risk of bleeding or with a significant bleeding history at the discretion of the investigator.

6. Study Intervention

Study intervention is defined as any investigational intervention(s), marketed product(s), placebo, or medical device(s) intended to be administered to a study participant according to the study protocol.

6.1. Study Intervention(s) Administered

All eligible study participants will receive treatment with cladribine 10 mg tablets during Year 1 and Year 2 (Intervention 1), and will be randomized at an approximate 1:2:2:1 ratio to receive a total of 2 LPs according to 1 of 4 different schedules (Intervention 2), as described in Table 2.

Table 2. Study Interventions

LP group	**1**	**2**	**3**	**4**
Intervention	**Cladribine tablets (Mavenclad®)**			
Dose formulation	Tablet			
Unit dose strength	10 mg tablet			
Dosage level	3.5 mg/kg divided into 2 yearly treatment courses (1.75 mg/kg per treatment course) (see Table 3), per the approved USPI			
Route of administration	Oral			
Dosing instructions	See Section 4.3			
Sourcing	Cladribine 10 mg tablets will be provided to the sites centrally by EMD Serono. Commercially available material will be used.			
Packaging and labeling	Cladribine 10 mg tablets will be provided in child-resistant day packs containing 1 or 2 tablets in blister cards. The tablets will be packaged and labeled per all regulatory requirements in the US.			
Intervention	**Lumbar punctures**			
Type	Procedure			
Schedule	Baseline and end of Week 5	Baseline and end of Week 10	Baseline and end of Year 1	Baseline and end of Year 2
	Optional additional third LP at the end of Year 2			
Use	Cladribine concentrations, cellular and soluble biomarker assessments			

Note: Refer to the SoA (Section 1.3) for the exact timing of the LPs.

LP=lumbar puncture, US=United States, USPI=United States Prescribing Information.

6.2. Preparation/Handling/Storage/Accountability

6.2.1. Cladribine Tablets

Cladribine 10 mg tablets will be provided by EMD Serono using commercial material. The tablets will be provided in boxes including child-resistant day packs. Each day pack will contain 1 or 2 tablets per blister card (Table 2). For the boxes, any commercially available pack size may be used.

On site, all cladribine tablet clinical trial material should be stored in a secure location. Only participants enrolled in the study may receive this clinical trial material, and only authorized site staff may supply or administer them.

Cladribine tablets for the first treatment course will be dispensed to the participants at the baseline visit. The first tablet will be taken directly at the site (AFTER the initial baseline LP). For participants assigned to LP Group 1 (second LP at the end of Week 5), the LP will be scheduled on the day of and after the last cladribine tablet intake. Cladribine tablets for the second treatment course will be dispensed at the end of Year 1 visit.

As the tablets are uncoated, they must be swallowed immediately once removed from the blister and not be left exposed on surfaces or handled for any period of time greater than that required for dosing. If a tablet is left on a surface or if it breaks and fragments fall from the blister, the area must be thoroughly washed. Hands must be dry when handling the tablets and washed thoroughly afterwards.

Cladribine tablets should be stored at controlled room temperature (20ºC to 25ºC [68ºF to 77ºF]; excursions permitted to 15ºC to 30ºC [59ºF to 86ºF]). Cladribine tablets should be stored in the original package to protect from moisture. Full instructions on handling and storage of cladribine tablets are provided in the USPI (EMD Serono, 2019).

The investigator is responsible for accountability, reconciliation, and record maintenance of the cladribine tablets clinical trial material. Dispensing and disposition of tablets will be recorded on appropriate accountability forms so that accurate records will be available for verification at each monitoring visit.

The investigator site will maintain records that adequately document that participants were provided the doses specified in this protocol, and all cladribine tablets provided were fully reconciled.

Unused cladribine tablets must not be discarded or used for any purpose other than the present study. Returned cladribine tablets should not be re-dispensed to a different participant.

A study monitor will document the adherence to drug administration.

Further guidance and information for the final disposition of unused cladribine tablets are provided in a separate manual.

6.2.2. Lumbar Punctures

Participants' LPs will be performed according to the processes established at the individual sites. The use of non-cutting needles is preferred. A maximum of 30 mL of CSF may be obtained per LP, in compliance with current LP consensus guidelines (Engelborghs, 2017). LPs are to be scheduled at each site at the timings outlined in the SoA (Section 1.3), as follows:

- For participants assigned to Group 1, the second LP at the end of Week 5 (and LP-aligned blood sampling) needs to take place on the same day of and after the intake of the last Week 5 cladribine tablet.
- For participants assigned to Group 2, the second LP at the end of Week 10 (and LP-aligned blood sampling) will have to be scheduled at the end of Week 10 (± 7 days).
- For participants assigned to Group 3, the second LP at the end of Year 1 (and LP-aligned blood sampling) will have to be scheduled at at least 43 weeks, but no later than 47 weeks after the last dose of cladribine tablets Year 1 treatment.
- For participants assigned to Group 4, the second LP at the end of Year 2 (and LP-aligned blood sampling) will have to be scheduled at least 43 weeks, but no later than 47 weeks after the last dose of cladribine tablets Year 2 treatment.

  Note: If the start of cladribine tablets Year 2 treatment needs to be delayed (e.g. to allow for lymphocyte recovery to ≥800 cells/µL), the end of Year 2 LP will be postponed by the same period of time (for up to 6 months), to maintain the protocol-specified time interval between start of cladribine tablets Year 2 treatment and the second LP.

  In addition, all study participants randomized to Groups 1-3 can volunteer for a third, optional LP at the end of Year 2, the timing of which would be aligned with the second LP of Group 4.

6.3. Measures to Minimize Bias: Randomization and Blinding

This is an open-label study; participants will be randomized 1:2:2:1 to one of 4 different schedules of LP timings as described in Table 2. Participants will be assigned using the REDCap randomization module. The central randomization procedure is used to reduce potential bias.

The randomization will be accessible in REDCap in a specific randomization eCRF. Once the participant is eligible, the study coordinator will be able to randomize the participant. prior to the start of treatment with cladribine tablets.Access to this form can be user specific and coordinators will be provided instructions for conducting the randomization.

6.4. Study Intervention Compliance (Cladribine Tablets)

Cladribine tablets for Year 1 treatment will be distributed at baseline visit, and cladribine tablets for Year 2 treatment will be distributed at the End of Year 1 visit (see Section 6.2.1).

The date and time of each dose will be recorded in the source documents and in the eCRF.

Participants' compliance with cladribine treatment will be assessed at each visit and will be documented in the source documents and eCRF. Deviation(s) from the prescribed dosage regimen should also be recorded in the eCRF.

A record of the number of tablets dispensed to and taken by each participant must be maintained and reconciled with LPs scheduled and compliance records. All cladribine tablet intake dates, including delayed intakes and/or (unallowed) dose reductions, will also be recorded in the eCRF.

6.5. Concomitant Therapy

It is recommended that administration of any other oral medicinal product be separated from that of cladribine tablets by at least 3 hours during the limited number of days of cladribine tablets administration.

Any concomitant therapies (medicines or non-drug interventions) used from the time the participant signs the ICF until completion of the study, including any changes, will be recorded in the eCRF.

Any concomitant medications (including prescription and over-the-counter medicines, vaccines, vitamins, and herbal supplements) that the participant is receiving at the time of enrollment or receives during the study must be recorded along with:

- Reason for use
- Dates of administration including start and end dates
- Dosage information including dose and frequency.

The Sponsor should be contacted if there are any questions regarding concomitant or prior therapy.

6.5.1. Rescue Medication

No specific rescue medication, i.e. any other DMD, is planned for this study, and any rescue medication required is at the discretion of the investigator (not provided by the sponsor).

Any participant who will require rescue medication will be discontinued from treatment with cladribine tablets, and will be discontinued from the study after completing a 30-day safety follow-up after taking the last cladribine tablets.

The SoA (Section 1.3) specifies the data to be collected if a participant discontinues the study.

6.5.2. Permitted Concomitant Medication

Acute short-term therapy of MS relapses with oral or IV corticosteroids at any dose will be permitted at certain times during the study, at the discretion of the investigator. Any such concomitant steroid therapy needs to be discontinued at least 4 weeks before any scheduled LP. If the participant requires oral or IV corticosteroid treatment within 4 weeks before a scheduled LP, it will be at the discretion of the investigator if the participant will be discontinued from the study.

If possible, scheduled MRI scans should be performed before administration of steroids, or at least 7 days after the last dose of steroids.

Any other medications not specifically excluded by the protocol and considered necessary for the participant’s welfare, and that would not interfere with cladribine tables, may be provided at the discretion of the investigator.

Since this will be an outpatient trial, special care should be taken to question the participants regarding any self-medication.

6.5.3. Prohibited Concomitant Medication

Participants may not use any of the following investigational drugs or therapies concomitantly with cladribine tablets:

- Any immunomodulatory therapy (including but not limited to IFN, dimethyl fumarate, glatiramer acetate, natalizumab, teriflunomide, ocrelizumab, daclizumab, fingolimod or other S1P receptor modulators, or oral DMDs)
- Any immunosuppressive or myelosuppressive therapy (including but not limited to cyclophosphamide, mitoxantrone, cyclosporin, methotrexate, and azathioprine) or chronic treatment with systemic corticosteroids
- Total lymphoid irradiation, alemtuzumab, IV Ig, or plasmapheresis
- Cytokine or anti-cytokine therapy.

Any participant who takes one of these prohibited medications will need to be discontinued from the study at the start of prohibited medication (data obtained until discontinuation will be included in the analyses).

Vaccination with live or attenuated live vaccine is not allowed during the study. After the study, such vaccinations should be avoided as long as the participant's white blood cell counts are not within normal limits (EMD Serono, 2019).

In addition, the concomitant use of compounds that require intracellular phosphorylation to become active (e.g. lamivudine, zalcitabine, ribavirin, stavudine, and zidovudine) should be avoided, because they could interfere with the intracellular phosphorylation and activity of cladribine (EMD Serono, 2019).

6.6. Dose Selection and Modification

All participants will receive 10 mg cladribine tablets at the recommended cumulative dosage of 3.5 mg per kg body weight, divided into 2 yearly treatment courses (1.75 mg per kg per treatment course, as outlined in Section 4.3).

The distribution of the cumulative dose over the 2 yearly treatment courses, based on the individual participant's body weight, is detailed in Table 3. The cycle dosage will be administered as 1 or 2 tablets daily over 4 or 5 consecutive days (Table 4). No more than 2 tablets should be administered daily. If a daily dose consists of 2 tablets, both tablets should be taken together as a single dose.

Full dosing and administration guidance can be found in the USPI (EMD Serono, 2019).

Table 3. Dose of Cladribine Tablets per Cycle by Participant Weight in Each Treatment Course (Year 1 and Year 2)

Body weight range (kg)	Dose in mg (number of 10 mg tablets) per treatment cycle	
	First cycle	Second cycle
40[a] to <50	40 mg (4 tablets)	40 mg (4 tablets)
50 to <60	50 mg (5 tablets)	50 mg (5 tablets)
60 to <70	60 mg (6 tablets)	60 mg (6 tablets)
70 to <80	70 mg (7 tablets)	70 mg (7 tablets)
80 to <90	80 mg (8 tablets)	70 mg (7 tablets)
90 to <100	90 mg (9 tablets)	80 mg (8 tablets)
100 to <110	100 mg (10 tablets)	90 mg (9 tablets)
≥110	100 mg (10 tablets)	100 mg (10 tablets)

[a] The use of cladribine tablets in patients weighing <40 kg has not been investigated.

Table 4. Cladribine 10 mg Tablets per Week Day

Total number of tablets per treatment week	Day 1	Day 2	Day 3	Day 4	Day 5
4	1	1	1	1	0
5	1	1	1	1	1
6	2	1	1	1	1
7	2	2	1	1	1
8	2	2	2	1	1
9	2	2	2	2	1
10	2	2	2	2	2

Dosing Errors

If a dose is missed, participants should not take double or extra doses.

If a dose is not taken on the scheduled day, then the participant must take the missed dose on the following day, and extend the number of days in that treatment cycle. If 2 consecutive doses are missed, the treatment cycle is extended by 2 days.

Dose Reductions

No dose reductions of cladribine tablets are planned for this study.

Treatment Interruptions and Delays

Lymphocytes must be ≥800 cells/µL before initiating cladribine tablets Year 2 treatment. If necessary, the start of Year 2 treatment can be delayed for up to 6 months to allow for recovery of lymphocytes to ≥800 cells/µL. If this recovery takes more than 6 months, the participant should not receive further treatment with cladribine tablets and will be discontinued from the study.

Participants with an ALC of <500 cells/µL should be monitored for signs and symptoms suggestive of infections, including herpes infections. If such signs and symptoms occur, anti-infective treatment should be initiated as clinically indicated, and interruption or delay of cladribine treatment until resolution of the infection should be considered (EMD Serono, 2019). The investigator should inform the sponsor and agree with the sponsor, if a delay is required. If cladribine treatment needs to be delayed for more than 6 months, the participant will be discontinued from the study.

6.7. Intervention after the End of the Study

No further interventions will be performed after the end of study. Participants who have completed the 2 cladribine tablet treatment courses should not receive additional cladribine tablet treatment during the next 2 years. Participants who have completed the study or discontinued the study will be treated for MS as part of their usual practice and care.

6.8. Special Precautions

All warnings and precautions per the cladribine tablets USPI (EMD Serono, 2019) should be followed.

7. Discontinuation of Study Intervention and Participant Discontinuation/Withdrawal

7.1. Discontinuation of Study Intervention

7.1.1. Discontinuation of Treatment with Cladribine Tablets

Discontinuation of cladribine tablets is mandatory in the following cases:

- Failure to meet all eligibility criteria for continuing cladribine tablet treatment (Section 5.4)
- Treatment with live or live attenuated vaccine at any time during the trial
- Initiation of any other prohibited treatment (Section 6.5.3)
- Occurrence of an exclusion criterion that is clinically relevant and affects the participant's safety, or any contraindication of cladribine tablets per the current USPI
- Any events that unacceptably endanger the safety of the participant.

If treatment with cladribine tablets is definitively discontinued, the participant will NOT remain in the study, but will be discontinued after he/she has been followed up for safety until 30 days after intake of the last cladribine tablet. Data up to study discontinuation will be included in the analysis.

The SoA (Section 1.3) specifies the data to be collected if a participant discontinues study treatment with cladribine tablets.

7.1.2. Post-Baseline Lumbar Puncture Not Conducted

If the eligibility criteria to receive the second or the optional third LP are not met (see Section 5.5), the participant will NOT remain in the study, but will be discontinued from the study after completing any ongoing cladribine tablets Year 1 or Year 2 treatment course, including a 30-day follow-up after the last intake of cladribine tablets and including at least 30 days of follow-up after the initial LP. Data up to study discontinuation will be included in the analysis.

The SoA (Section 1.3) specifies the data to be collected if a participant discontinues before the second LP has been performed.

7.1.3. Temporary Discontinuation

Not applicable.

7.1.4. Rechallenge

Not applicable.

7.2. Participant Discontinuation from the Study

Participants will be discontinued from the study in the following situations (the participant will be permanently discontinued both from the study interventions and from the study; data collected up to discontinuation can be used for analysis):

- Eligibility criteria for starting cladribine tablets Year 2 treatment not met (see Section 5.4)
- Second LP cannot be performed (see Section 5.5)
- Investigator decides that the participant should be discontinued for safety, behavioral compliance, or administrative reasons.

Further, participants or their caregiver / legal representative may withdraw consent to participate in the study at any time and without giving reasons. In these cases, the participant will be permanently discontinued from the study.

- Further, participants or their caregiver / legal representative may withdraw consent for the optional single cell RNA sequencing, the optional RNA sample banking, or an optional third LP at any time. This does not preclude the participant from continuing in the study and completing the other assessments.
- If a participant withdraws consent for disclosure of future information, the sponsor may retain and continue to use any data collected before such a withdrawal of consent.
- If a participant withdraws from the study, he/she may request destruction of any samples taken and not tested, and the investigator must document this in the site study records.

The SoA specifies the data to collect at study discontinuation and follow-up, and any additional evaluations that need to be completed.

The participant will be informed that any data collected from the signature of the ICF until the point of withdrawal will be maintained and included in the analysis. The investigator should make every attempt to clarify the level of withdrawal: e.g. if the participant can still be contacted by phone for checking his/her status (e.g. alive or not). In case of withdrawal from the study, the appropriate eCRF Section must be completed.

If a participant withdraws consent from the optional single cell RNA sequencing before the samples have been analyzed, optional vaccine collection, or from the optional RNA sample banking, the participant’s samples will be destroyed without being analyzed. In addition, data collected will be excluded from the analysis. If a participant withdraws from the optional single cell RNA sequencing or optional vaccine collection after the participant’s sample has been analyzed, any information learned from the analysis will remain part of the study database and may not be removed.

Participants or their caregiver/legal representative will be informed that they have the right to withdraw from the study at any time, without prejudice to their medical care, and that they are not obliged to state their reasons. Any withdrawal must be fully documented in the eCRF and source documents and should be followed up by the investigator.

Participants who are discontinued / withdrawn from the study will not be replaced.

7.3. Lost to Follow-up

A participant will be considered lost to follow-up if he or she repeatedly fails to return for scheduled visits and is unable to be contacted by the study site.

The following actions must be taken if a participant fails to return to the clinic for a required study visit:

- The site must attempt to contact the participant and reschedule the missed visit as soon as possible and counsel the participant on the importance of maintaining the assigned visit schedule and ascertain whether or not the participant wishes to and/or should continue in the study.
- Before a participant is deemed lost to follow-up, the investigator or designee must make every effort to regain contact with the participant (where possible, 3 telephone calls and, if necessary, a certified letter to the participant's last known mailing address or local equivalent methods). These contact attempts should be documented in the participant's medical record.
- Should the participant continue to be unreachable, he/she will be considered to have withdrawn from the study.

8. Study Assessments and Procedures

All screening procedures, and all study procedures and their timing, are summarized in the SoA (Section 1.3), including all baseline data collected.

- Protocol waivers or exemptions are not allowed.
- Immediate safety concerns should be discussed with the sponsor immediately upon occurrence or awareness to determine if the participant should continue or discontinue study intervention.
- Adherence to the study design requirements, including those specified in the SoA, is essential and required for study conduct.
- All screening evaluations must be completed and reviewed to confirm that potential participants meet all eligibility criteria. For all participants who signed informed consent, the investigator will maintain a screening log to record details of all screened participants and to confirm eligibility or record reasons for screening failure, as applicable.
- The investigator will ensure that each participant has provided written informed consent prior to any study procedures.
- Procedures conducted as part of the participant's routine clinical management (e.g. pregnancy test, routine blood tests, and assessment of renal and hepatic function) and obtained before signing of the ICF may be utilized for screening or baseline purposes, provided the procedures met the protocol-specified criteria and were performed within the time frame defined in the SoA.

CSF Sampling

Total CSF volumes of up to 30 mL (minimum required volume 7.5 mL) per LP will be collected from each participant at each LP (required LP at baseline, required second LP as randomly assigned, and optional third LP at the end of Year 2 for participants from LP Groups 1-3; as scheduled in the SoA in Section 1.3).

Three required samples will be obtained per total CSF sample, using the volumes shown in Table 5:

- Cellular marker sample for the assessment of lymphocyte subtypes and other cell types (local assessment; see Section 8.8.1)
- Biomarker sample for the assessment of soluble biomarkers (NfL, secondary and exploratory biomarkers; central assessment, see Section 8.8.2)
- Cladribine assay sample for the assessment of cladribine concentration (for LPs at the end of Week 5 only); central assessment; see Section 8.5).

After the 3 required samples have been obtained, any remaining CSF volume will be used to obtain the optional samples for RNA single cell sequencing and RNA sample banking (only from participants who consented; see Section 8.8.3 and Section 8.8.4). Any remaining volume after all required and optional samples have been taken will then be stored at the sponsor site (Washington University), with an option to evaluate biomarker, safety, or efficacy aspects of interest that might arise during or after the study.

The maximum amount of CSF required from each participant over the duration of the study will thus not exceed 60 mL (Table 5).

Table 5. Estimated CSF Volume Collected from Each Participant (N=50) During Required Study Procedures

	mL per LP		mL total required[a]	
	Minimum required	Estimated maximum obtained	Minimum required	Estimated maximum obtained
Total CSF volume	7.5	30	15	60
1 cellular marker sample	5	NA	10	NA
1 soluble biomarker sample	1.5	NA	3	NA
1 cladribine assay sample	1	NA	2	NA

[a] Two LPs required for each participant. The optional third LP (separate consent) is not considered.

CSF=cerebrospinal fluid, LP=lumbar puncture, N=approximate number of subjects planned to be enrolled, NA=not applicable.

Blood Sampling

Protocol-required blood sampling will be aligned with the timing of the LPs.

Optional vaccine blood sampling will be aligned with the timing of vaccines received as standard of care for participants in treatment with cladribine tablets.

Total volumes of up to 60 mL blood (minimum required volume 16 mL) will be collected from each participant at each LP-aligned blood sampling at baseline (on the same day as the baseline LP), and post-baseline at Week 5, Week 10, Year 1 and Year 2 (per the SoA in Section 1.3). If an LP is scheduled, the LP-aligned blood sampling will be done on the same day. Repeat blood samples may be taken for technical issues.Four required samples will be obtained per blood draw, using the volumes shown in Table 6:

- Two cellular marker samples for the assessment of lymphocyte subtypes and other cell types (local assessment; see Section 8.8.1)
- Biomarker sample for the assessment of soluble biomarkers (NfL, secondary and exploratory biomarkers; central assessment, see Section 8.8.2)
- Cladribine assay sample for the assessment of cladribine concentration (central assessment; see Section 8.5).

After the 4 required samples have been obtained, any remaining blood volume will be used to obtain the optional blood samples for RNA single cell sequencing and RNA sample banking (only from participants who consented; see Section 8.8.3 and Section 8.8.4). Any remaining volume after all required and optional samples have been taken, including the supernatant from the cellular marker samples after separation of cellular components, will then be stored at Washington University, with an option to evaluate other biomarker, safety, or efficacy aspects of interest that might arise during or after the study.

The maximum amount of blood obtained from each participant for required procedures over the duration of the study will thus not exceed 300 mL (Table 6).

Optional: Total volumes of up to 10mL will be collected from each participant that consents to the vaccine blood collection at each sampling at pre-vaccination, and post-vaccination visits at 4 Weeks and 6 Months (per the SoA in Section 1.3.1).

One sample will be obtained per blood draw for the optional vaccine collection, using the volumes shown in Table 6:

- One sample for the assessment of vaccine antibodies (central assessment, see Section 8.9)

The maximum amount of blood obtained for the optional vaccine collection from each participant is 30 mL (Table 6).

Table 6. Estimated Blood Volume Collected from Each Participant (N=50) During Required Study Procedures (Routine Safety Assessments Excluded)

	mL per visit		mL total required[a,c]	
	Minimum required	Estimated maximum obtained	Minimum required	Estimated maximum obtained
Total blood volume	16	up to 60[b]	80	300[b]
2 cellular marker samples	2x 5	NA	50	NA
1 soluble biomarker sample	5	NA	25	NA
1 cladribine assay sample	1	NA	5	NA
1 vaccine antibody sample (optional)	2	10	6	30

[a] Five LP-aligned blood samplings will be performed for each participant. Blood sampling for routine safety assessments is not considered.

[b] Actual volume may vary depending on the individual site procedures.

[c] Optional: 3 vaccine aligned samplings will be performed for each consenting patient that is obtaining a standard of care vaccine and has started cladribine tablet treatment. LP=lumbar puncture, N=approximate number of subjects planned to be enrolled, NA=not applicable.

Instructions for the collection, handling, and processing of the different CSF and blood samples will be provided separately. The actual date and time (24-hour clock time) of each sample will be recorded. Retention time and possible analyses of the samples after the end of study are specified in the respective ICF.

8.1. Efficacy Assessments

This exploratory study will primarily assess a set of cellular and soluble biomarkers in the CSF and in peripheral blood over 2 years of treatment with cladribine tablets. These assessments are discussed in Section 8.8.1 (cellular biomarkers) and Section 8.8.2 (soluble biomarkers).

Clinical efficacy outcomes (relapse evaluation, EDSS, MRI assessments) are secondary endpoints of the study, the timings of these assessments are provided in the SoA (Section 1.3).

8.1.1. Relapse Evaluation

Participants will be instructed to contact the investigator immediately if they develop new or re-occurring or worsening neurological symptoms. At each scheduled visit, the participant will be asked whether any such symptoms have occurred.

If a participant reports symptoms indicative of a relapse (for relapse definition see Section 3), the investigator will assess whether the symptoms occur in the presence of fever or infection (in case of an unscheduled phone-contact, the participant can simply be asked). If fever or infection is excluded, the investigator must arrange for a neurological examination as soon as possible, at the latest within 7 days following the reporting of the event. If fever or infection cannot be excluded, the neurological examination by the investigator will have to be postponed until the fever or the infection have ceased (provided that the symptoms indicative of a relapse are still present).

Based on the respective EDSS scores, in conjunction with the results from previous examinations, and based on the results of the neurological examination, the investigator will assess whether the EDSS criterion and the neurological criteria for a relapse (Section 3) are fulfilled.

The investigator will document any relapses (per definition in Section 3) in the relapse module of the eCRF, including the following data if available: EDSS and body temperature assessments; relapse onset date and stabilization/resolution date; estimated relapse duration; major systems affected; treatment required; and outcome of relapse.

Relapse evaluation data will be used to calculate the ARR (secondary endpoint) during the analysis of the study data.

8.1.2. Expanded Disability Status Scale (EDSS)

The EDSS (Kurtzke, 1983) is a standard, validated and reliable neurological assessment to describe the clinical severity and functional deficits of MS. The EDSS is the internationally accepted and most widely used instrument to assess long-term disability progression and occurrence of MS relapses in clinical trials (Meyer-Moock, 2014).

The EDSS consists of an ordinal rating system ranging from 0 (normal neurological status) to 10 (death due to MS) in 0.5 increment intervals, with 6 being the critical point at which an individual requires ambulatory assistance. The lower scale values of the EDSS measure impairments based on the neurological examination, while the upper range of the scale (EDSS >6) measures handicaps of patients with MS. The determination of EDSS 4-6 is heavily dependent on aspects of walking ability (Meyer-Moock, 2014).

For each participant, EDSS assessments are scheduled as described in the SoA (Section 1.3). Additional assessments may be required in case of a potential relapse (see Section 8.1.1). Sites will be instructed, whenever possible, to use the same evaluating physician to perform EDSS assessments for a given study participant.

8.1.3. Magnetic Resonance Imaging (MRI)

For each participant, the baseline MRI scan can be performed at any time during the 3-week screening period. Post-baseline MRIs will be obtained at the end of Year 1 and end of Year 2, as scheduled in the SoA (Section 1.3). MRIs can be obtained up to 3 weeks prior to dosing in Year 1. Two different types of MRI scans will be performed at each assessment:

- Standard sequences to evaluate T1 Gd+ and new or enlarging T2 lesions
- Quantitative gradient echo contrast imaging (R2t*) of cerebral cortex, deep gray matter, and white matter to measure tissue integrity.

All scans will be performed according to a standard procedure detailed in a separate manual. Each participant should be scanned using the same machine throughout the trial.

All scans will be assessed centrally by blinded assessors, the standard scans (Gd+ and T2 lesions) by a pre-specified provider for central MRI reading, the echo contrast images by a central reader at the site of the sponsor.

The following parameters associated with MS disease activity will be assessed and documented centrally for all participants for each scan obtained during the study:

- Standard scans:
  - Total number of Gd+ lesions
  - Total number of new T2 lesions
  - Total number of newly enlarged T2 lesions
- Quantitative gradient echo contrast imaging (R2t*)
  - Any changes in R2t* in white matter and gray matter.

8.2. Safety Assessments

The safety profile of this study's interventions will be assessed through the recording, reporting, and analysis of medical conditions, AEs, physical examination findings, and safety laboratory assessments. Assessments and time points of safety laboratory assessments will comply with all requirements in the USPI (EMD Serono, 2019).

A comprehensive assessment of any potential AE/UP experienced by each participant will be conducted starting when the participants have signed the ICF and throughout the study. The investigator will report any AE/UP, whether observed by the investigator or reported by the participant; the reporting period is specified in Section 8.3.1.

A Data Monitoring Committee (DMC) will be established that will monitor the participants' safety (for details see Section 9.6).

All clinically significant abnormalities occurring before signature of the ICF should be recorded in the Medical History Section and/or Disease History; all abnormalities occurring or worsening after signature of the ICF should be recorded in the AE/UP section.

Planned time points for all safety assessments, including safety laboratory assessments, are provided in the SoA (Section 1.3).

8.2.1. Physical Examinations

A physical and neurological examination per standard MS care at the respective site will be performed as scheduled in the SoA (Section 1.3). Investigators should pay special attention to clinical signs related to previous serious illnesses, and to a potential MS relapse.

The participant's body weight has to be assessed and documented on the eCRF at least at baseline and at the End of Year 1 visit, to identify the required number of cladribine tablets to be taken as described in Section 6.6.

The investigator will have to document any clinically relevant changes as an AE.

8.2.2. Vital Signs

Vital sign data will not be collected during this study. The investigator will monitor each participant for vital signs per routine clinical practice, and any clinically relevant changes will be documented as an AE.

8.2.3. Electrocardiograms

Electrocardiogram (ECG) data will not be collected during this study. The investigator will monitor participants' ECGs per routine clinical practice, and any clinically relevant changes will be documented as an AE.

8.2.4. Clinical Safety Laboratory Assessments

All participants will have their ALC routinely monitored for safety by the local laboratory according to the schedule described in the SoA (Section 1.3). In addition, the CBC will be assessed by the local laboratory, according to the schedule described in the SoA (Section 1.3).

For the following hematologic parameters, all scheduled and unscheduled assessments obtained will be collected on the eCRF and will be evaluated as part of the safety assessment:

- ALC
- White blood cells
- Hemoglobin
- Platelets
- Hematocrit.

Laboratory assessments of lymphocyte subsets and other immune cells as part of the biomarker evaluations are discussed in Section 8.8.1.

In addition, participants' renal and hepatic function will be assessed at Screening and at the End of Year 1 visit (i.e. before the start of the second year of treatment with cladribine tablets), per standard of care and consistent with the cladribine tablets USPI (EMD Serono, 2019). Lab results

need to be obtained within 3 weeks of cladribine tablets treatment in Year 1 and Year 2. Any clinically relevant changes in creatinine clearance or hepatic function, when compared with the pre-treatment assessment at screening, will be documented as an AE.

A list of all clinical safety laboratory tests to be performed is included in Appendix 2.

The investigator must review all laboratory reports and document this review. Abnormal laboratory findings should NOT be routinely captured and reported as AEs, as they will be collected and analyzed separately. However, abnormal laboratory findings or other objective measurements that meet the criteria for an SAE, result in discontinuation of cladribine tablets treatment, require medical intervention, or are judged by the investigator to be clinically significant changes from baseline values should be captured and reported on the AE Section of the eCRF.

8.2.5. Suicidal Ideation and Behavior Risk Monitoring

Not applicable.

8.3. Adverse Events and Serious Adverse Events

The definitions of an AE and SAE are provided in Appendix 3.

The investigator and any qualified designees are responsible for detecting, documenting, and recording events that meet the definition of an AE or SAE.The investigator remains responsible for following up participants that have AEs that are serious, considered related to the study interventions or study procedures, or that caused the participant to discontinue the study (see Section 7).

8.3.1. Time Period and Frequency for Collecting AE and SAE Information

The recording period for AEs begins when the participant is initially included in the study (date of signature of first ICF) and continues at least to the end of the mandatory safety follow-up period (i.e. up to 24 months in most participants) or up to 30 months in participants with delayed start of Year 2 treatment.

AEs are recorded and assessed continually throughout the trial and are assessed for final outcome at the final visit.

Any SAE assessed as related to cladribine tablets must be reported whenever it occurs, irrespective of the time elapsed since the last administration of cladribine tablets.

All SAEs will be recorded and reported to the sponsor or its designee immediately and under no circumstance should this exceed 1 working day, as indicated in Appendix 3. The investigator will submit any updated SAE data to the sponsor or its designee within 1 working day of it being available.

Any medical occurrences that begin before the participant has taken the first cladribine tablet and before the baseline LP, but after obtaining informed consent, will be analyzed as History/Current Medical Conditions, not as treatment-emergent AEs (TEAEs).

Investigators are not obligated to actively seek AEs or SAEs after conclusion of the study participation. However, if the investigator learns of any SAE, including a death, at any time after

a participant has been discharged from the study, and he/she considers the event to be reasonably related to the study intervention or study participation, the investigator must promptly notify the sponsor.

The detailed methods of recording, evaluating, and assessing causality of an AE and SAE and the procedures for completing and transmitting SAE reports are provided in Appendix 3.

8.3.2. Method of Detecting AEs and SAEs

At each study visit, the participant will be queried on changes in his or her condition. During the AE reporting period, any unfavorable changes in the participant's condition will be recorded as AEs, regardless if reported by the participant or their caregiver/legal representative or observed by the investigator.

Care will be taken not to introduce bias when detecting AEs and/or SAEs. Complete, accurate, and consistent data on all AEs experienced for the duration of the reporting period (defined above) will be reported on an ongoing basis in the appropriate Section of the eCRF. All SAEs must be additionally documented and reported using the appropriate Report Form as specified in Appendix 3.

Due to the interventional nature of the LPs that are performed specifically for the purpose of this study, the study-specific procedure of AE and SAE data collection and reporting will also be followed for any AEs occurring in association with the LP.

In this protocol, symptoms and signs of relapse or worsening of MS will be captured in the context of the efficacy assessment, and recorded on the relapse module of the eCRF. Therefore, symptoms, relapses, or worsening of MS will not be considered as AEs nor captured on the AE module of the eCRF, unless considered possibly or probably related to treatment with cladribine tablets or LP procedures (i.e. worsening is not consistent with the anticipated natural progression of the disease), and/or unless the outcome is fatal.

8.3.3. Follow-Up of AEs and SAEs

After the initial AE/SAE report, the investigator is required to proactively follow each participant at subsequent visits/contacts. All SAEs will be followed until resolution, stabilization, the event is otherwise explained, or the participant is lost to follow-up (as defined in Section 7.3). Further information on follow-up procedures is provided in Appendix 3.

8.3.4. Safety Reporting to Health Authorities, Independent Ethics Committees/Institutional Review Boards, and Investigators

Complete data onadverse drug reactions (ADRs), i.e. all AEs considered related to the study treatment, SAE reports and reports of special situations (e.g. overdose, medication error etc.) including safety laboratory data, vital signs, and efficacy data necessary to assess and monitor the safety of cladribine tablets shall be collected and provided to EMD Serono by the sponsor of this study as specified in the collaboration contract.

Sponsor will send appropriate safety notifications to Health Authorities in accordance with applicable laws and regulations.

The investigator must comply with any applicable site-specific requirements related to the reporting of ADRs/SAEs (particularly deaths) involving study participants to the Independent Ethics Committee/Institutional Review Board (IEC/IRB) that approved the study.

In accordance with International Council for Harmonisation of Technical Requirements for Pharmaceuticals for Human Use (ICH) Good Clinical Practice (GCP), the sponsor/designee will inform the investigators of "findings that could adversely affect the safety of participants, impact the conduct of the study or alter the IEC's/IRB's approval/favorable opinion to continue the study." In particular and in line with respective regulations, the sponsor/designee will inform the investigators of AEs that are both serious and unexpected and are considered to be related to the administered product ("suspected unexpected serious adverse reactions" or SUSARs). The investigator should place copies of safety reports in the Investigator Site File. National regulations with regard to safety report notifications to investigators will be taken into account.

When specifically required by regulations and guidelines, the sponsor/designee will provide appropriate safety reports directly to the concerned lead IEC/IRB and will maintain records of these notifications. When direct reporting is not clearly defined by national or site-specific regulations, the investigator will be responsible for promptly notifying the concerned IEC/IRB of any safety reports provided by the sponsor/designee and of filing copies of all related correspondence in the Investigator Site File.

8.3.5. Pregnancy and In Utero Exposure

Cladribine tablets are contraindicated in pregnant women and in females and males of reproductive potential who do not plan to use effective contraception during treatment with cladribine tablets and for 6 months after the last dose in each treatment course. Pregnancy should be excluded before the initiation of each treatment course (i.e. before initiation of Year 1 and Year 2 treatment with cladribine tablets). Pregnancies should be prevented by the use of effective contraception during cladribine tablet dosing and for at least 6 months after the last dose of each treatment course.

Only pregnancies considered by the investigator to be related to study treatment (for example, resulting from a drug interaction with a contraceptive medication) are considered to be AEs. However, all pregnancies with an estimated conception date during the period as defined must be recorded by convention in the AE page/Section of the eCRF. The same rule applies to pregnancies in female participants and to pregnancies in female partners of male participants. The investigator must notify the sponsor/designee in an expedited manner of any pregnancy using the Pregnancy Report Form, which must be transmitted via an email immediately (within a maximum of 1 working day after becoming aware of the event), following specific completion instructions.

Investigators must actively follow up, document, and report on the outcome of all these pregnancies, even if the participants are withdrawn from the study.

Investigators must notify the sponsor/designee of these outcomes using the Pregnancy Report Form. If an abnormal outcome occurs, the SAE Report Form will be used if the participant sustains an event and the Parent-Child/Fetus Adverse Event Report Form if the child/fetus sustains an event.

Any abnormal outcome (e.g. spontaneous abortion, fetal death, stillbirth, congenital anomalies, or ectopic pregnancy) must be reported in an expedited manner as described, while normal outcomes must be reported within 45 days after delivery.

In the event of a pregnancy in a participant occurring during the course of the trial, the participant must be discontinued from cladribine tablets immediately. The sponsor/designee must be notified without delay and the participant must be followed as mentioned above.

8.3.6. Unanticipated Problems

Any event that meets the definition of an Unanticipated Problem (UP), i.e. unexpected, related to study participation and suggests that the research places the participant or others at greater risk of harm than was previously recognized, should be recorded and reported to the sponsor or its designee immediately and under no circumstances should this exceed 1 working day, as indicated in Appendix 3. The investigator will submit any updated UP data to the sponsor or its designee within 1 working day of it being available.

The investigator must comply with any applicable site-specific requirements related to the reporting of UPs involving study participants to the Independent Ethics Committee/Institutional Review Board (IEC/IRB) that approved the study.

8.4. Treatment of Overdose

An overdose is defined as any dose greater than the highest daily dose included in a clinical trial protocol or planned for an individual participant enrolled in the trial. For this study, the determination if a participant had an overdose will be left to the discretion of the investigator, based on the quantity of overdose, emergence of any clinical signs and symptoms suggestive of a toxic administration, as well as his/her own judgment as it applies to each individual case.

The approved USPI of cladribine tablets (EMD Serono, 2019) does not recommend specific treatment for an overdose.

Treatment consists of careful observation and initiation of appropriate supportive measures. Because of the rapid and extensive intracellular and tissue distribution, hemodialysis is unlikely to eliminate cladribine to a significant extent.

Discontinuation of cladribine tablets may need to be considered. Decisions regarding dose interruptions or treatment discontinuation will be made by the investigator in consultation with the Sponsor, based on the clinical evaluation of the participant.

8.5. Pharmacokinetics

No longitudinal pharmacokinetic profiles will be obtained after administration of cladribine tablets. However, the analysis of cladribine concentrations in the CSF and serum samples, to explore the correlation between CSF and serum concentrations and clinical outcomes, is a required part of the study. Smaller samples (approximately 1 mL) will be obtained from the CSF- and LP-aligned blood sample obtained at baseline and at the end of Week 5 (see SoA in Section 1.3) that will be used to assess the cladribine CSF and serum concentrations.

The quantification of cladribine in CSF and serum will be performed by a central vendor. The serum assay has been validated; validation of the CSF assay will be performed by a central, qualified vendor.

Precise details for the preparation and shipment of the CSF and blood (serum) samples will be provided by the central vendor as a separate user's manual.

After testing is complete, the central vendor will ship any remaining samples to the sponsor site (Washington University in St. Louis). The central vendor and the sponsor will store the CSF and serum samples in a secure storage space with adequate measures to protect confidentiality.

Retention time and possible analyses of samples after the end of study are specified in the ICF.

8.6. Pharmacodynamics

To assess the effect of cladribine tablets on ALC, white blood cell counts, hemoglobin, platelets, and hematocrit will be part of the safety laboratory assessments in this study (Section 8.2.4).

8.7. Pharmacogenetics

No DNA sampling is planned for this study.

8.8. Biomarkers

Exploratory research on cellular and soluble biomarkers in CSF and whole blood is a required part of this study, to better understand the MoA of cladribine tablets and to generate hypotheses regarding potential predictive markers. Cellular biomarkers will be assessed locally at each site, soluble biomarkers will be assessed centrally.

In addition, participants may consent to provide optional CSF and blood samples for single cell RNA sequencing and for RNA sample banking.

After all tests have been completed, investigators will ship any remaining sample volume to the sponsor site (Washington University in St. Louis). Investigators and sponsor will store the samples in a secure storage space with adequate measures to protect confidentiality. Retention time and possible analyses of samples after the end of study are specified in the respective ICF.

Investigators will be responsible for ensureing all staff are appropriately trained on applicable regulations and standards in shipping biological materials.

8.8.1. Cellular Biomarkers

For all participants, the following cellular markers will be assessed locally in all CSF- and LP-aligned blood samples obtained:

- Lymphocytes and subtypes:
  - CD3+ T lymphocytes and CD19+ B lymphocytes
  - CD4+ T cells and Tregs
  - CD8+ T cells
  - Plasmablasts
  - Natural killer cells

- CD14+ cells
- Other cell types of interest identified during the course of the study.

The CSF and whole blood samples for these assessments will be obtained at the time points scheduled in the SoA (Section 1.3). The required sample volumes are described in Table 5 and Table 6.

Assessments will be performed locally at each site, using a standardized, quality-controlled flow cytometry protocol.

Investigators will be provided with detailed instructions regarding the collection, handling, processing, and storage of the CSF and blood samples, and the validated cytometry protocol in a separate Laboratory Manual.

8.8.2. Soluble Biomarkers

For all participants, the following soluble biomarkers will be assessed centrally:

- NfL levels, planned to be assessed in both CSF (primary) and serum (secondary)
- Biomarkers related to inflammation, planned to be assessed in both CSF and serum:
  - IgG, IgM, IgA
  - Oligoclonal bands
  - CXCL12, CXCL13, CCL19, CCL21
- Biomarkers related to inflammation, planned to be assessed in serum only:
  - IFNγ
  - Interleukin 10, interleukin 4
  - Osteopontin
  - Complement levels
- Biomarkers related to neuronal injury or degeneration, planned to be assessed in serum only:
  - NfH
  - Tau
  - Isoprostanes

Other biomarkers considered important to explore the MoA of cladribine tablets or to generate hypotheses regarding potential predictive biomarkers may be added to the panel of biomarkers assessed as the study proceeds.

The CSF and whole blood samples for these assessments will be obtained at the time points scheduled in the SoA (Section 1.3). The required sample volumes are described in Table 5 and Table 6.

All biomarker analyses will be performed centrally; OLINK technology will be used whenever applicable.

Precise details for the preparation and shipment of the CSF and blood (serum) samples for biomarker assessments will be provided by the central vendor as a separate user's manual.

8.8.3. Optional Single Cell RNA Sequencing

Single cell RNA sequencing of cells obtained from CSF and blood samples may help to illuminate the genetic properties and degree of heterogeneity of these cells, which might impact a participant's response to cladribine tablets treatment, and the severity and progression of disease.

The individual sites of this study will have the option to perform single cell RNA sequencing using samples from participants who were enrolled at their sites and who consented to this component of the study.

Participants who do not wish to participate may still participate in the study, including any other optional procedures.

From all participants who were enrolled at sites performing single cell RNA sequencing and who consented to this component of the study, a portion of each CSF- and LP-aligned blood sample obtained (for timing see SoA in Section 1.3) will be used for single cell RNA extraction, if sufficient CSF and blood sample material is available.

These assays will be performed locally at each site, using local site procedures as appropriate. The results of these optional analyses may be reported in a separate study summary by each site. After testing is complete, investigators will ship any remaining sample volume to the sponsor site (Washington University in St. Louis).

Investigators and sponsor will store all samples in a secure storage space with adequate measures to protect confidentiality. Retention time and possible analyses of samples after the end of study are specified in the respective ICF.

8.8.4. Optional RNA Sample Banking

Participation in the RNA sample banking component of this study is optional. Participants who do not wish to participate may still participate in the study, including any other optional procedures.

From all participants who consented to the optional RNA sample banking, small samples of each CSF- and LP-aligned blood sample obtained (for timing see SoA in Section 1.3) will be used for RNA extraction. These RNA samples will then be stored for future analysis.

All RNA samples for banking will be shipped to and stored at the sponsor site (Washington University in St. Louis). Investigators and sponsor will store all RNA samples in a secure storage space with adequate measures to protect confidentiality. Details on the processes for collection, handling, processing, shipment, storage, and destruction of these samples will be provided in a separate Manual. Retention time will be specified in the respective ICF.

8.9. Optional Vaccine Samples

Evaluating blood samples of participants receiving cladribine tablets treatment, who receive a vaccine, may provide understanding of the impact of cladribine on development of antibody titers. Cladribine acts by decreasing lymphocyte counts and can impair immune responses. A vaccine helps the body generate adequate antibody titers to prevent future infection. There is no data available to indicate if cladribine impacts the development of antibody titers which could make a vaccine response inadequate to prevent infection.

Participation in the vaccine sample collection component of the study is optional. Participants who do not wish to participate may still participate in the study, including any other optional studies or procedures.

Consenting participants planning to obtain a standard of care vaccine, who are receiving cladribine tablets treatment, will provide a blood sample within 3 weeks prior to receiving the vaccine, 1 month after receiving the vaccine, and 6 months after receving the vaccine.

Samples will be shipped and analyzed centrally at the sponsor site (Washington University in St. Louis).

8.10. Immunogenicity Assessments

Not applicable.

8.11. Medical Resource Utilization and Health Economics

Not applicable.

9. Statistical Considerations

9.1. Statistical Hypotheses

This is an exploratory, hypotheses-generating study. No formal hypotheses are planned for this study.

9.2. Sample Size Determination

The sample size for this hypothesis-generating study is based on the feasibility of enrollment. A sample size of 42 study participants is considered sufficient to describe the changes in CSF and serum biomarkers during treatment with cladribine tablets and to perform the exploratory modeling analyses to identify potentially predictive CSF or serum biomarkers. Assuming 15% of study participants will drop out during the 2-year study duration, a total of 50 participants who meet all inclusion and exclusion criteria will need to be randomized and dosed to have at least 42 participants who complete the study with all assessments performed.

9.3. Populations for Analyses

The following populations are defined:

Population	Description
Enrolled	All participants who sign the Informed Consent Form
Randomized (to lumbar puncture)	All enrolled participants who meet all inclusion and exclusion criteria
Randomized and treated	All randomized participants who receive at least one cladribine 10 mg tablet
Safety	All participants who receive at least one cladribine 10 mg tablet

All randomized and treated study participants will be included in the analysis, regardless of adherence to the study protocol. This includes participants who do not receive the study intervention (LP) as intended, have discontinued use of cladribine tablets, or those who are found to have been included in the study erroneously.

Data from participants who have withdrawn consent for the study will be included in the analysis, unless they have specified that their data not be used.

Analysis of safety outcomes will include all participants who have received at least one dose of cladribine tablets with follow-up safety data. Should there be any participants who received a baseline LP but no dose of cladribine tablets, AEs and other safety outcomes for these participants would be listed separately.

9.4. Statistical Analyses

The Statistical Analysis Plan (SAP) will be finalized at least 2 months prior to the first interim analysis and it will include a more technical and detailed description of the statistical analyses described in this section. This Section is a summary of the planned statistical analyses of the most important endpoints including primary and key secondary endpoints.

9.4.1. General Considerations

In general, data will be summarized descriptively and presented in tables and graphically. Continuous variables will be summarized with means, standard deviations, and two-sided 95% confidence intervals (CIs), if normally-distributed, or medians and range, if not normally-distributed. Categorical/discrete variables will be displayed as frequency tables. Correlations will be analyzed using Spearman's or Pearson's correlation. The parametric assumptions underlining the models specified will be examined. P-values will be considered statistically significant at the two-sided α=0.05 level and will be corrected for multiple comparisons using the Holm-Bonferroni step-down correction within each type of data (6 data types as described in Sections 8.1.1, 8.1.2, 8.1.3, 8.8.1, 8.8.2, and 8.8.3), as appropriate.

Although this is a multicenter study, it is not expected to enroll enough participants at each site to allow for within-site analyses. Therefore, participants' data will be pooled across sites for all analyses.

For a small number of participants, the second (Year 2) cladribine tablets treatment course may need to be delayed, e.g. to allow for lymphocyte recovery to at least 800 cells/µL. Up to 6 months of delay are allowed for Year 2 treatment. For participants delaying Year 2 treatment by ≤6 months, data will be treated as if the treatment had occurred at the original time point specified, and inferences will be conducted accordingly.

Due to the length of the study, it is likely that there will be some missing outcome data due to loss to follow-up. Patterns and degrees of missingness will be summarized and will inform the approach taken to dealing with missing data. More detail about missing data handling will be provided in the SAP.

9.4.2. Primary Endpoints

The primary endpoints will be analyzed at Week 10 (Interim Analysis 1), Month 12 (Interim Analysis 2, and Month 24 (final analysis). The absolute and percent change from baseline will be presented for each time point for CD3+ T lymphocytes, CD19+ B lymphocytes, and NfL levels in CSF, with a two-sided 95% CI and p-value. As these data are not expected to be normally distributed, the nonparametric Wilcoxon signed rank test will be used to compare each biomarker at baseline and during treatment. Spearman rank correlations will be used to examine the relationship between biomarkers, and between biomarkers and clinical/safety endpoints.

9.4.3. Secondary Endpoints

All secondary endpoints will be analyzed at Week 10 (Interim Analysis 1), Month 12 (Interim Analysis 2), and Month 24 (final analysis). Data will be summarized descriptively.

Other CSF and serum biomarkers: Same as for primary endpoint analysis.

ARR: The ARR over the 12- and 24-month periods will be presented, accompanied by the respective 95% CIs. The proportion (and 95% CI) of participants experiencing a relapse over the 12-month and 24-month periods will also be presented.

EDSS: The change in EDSS from baseline will be reported at Month 12 and Month 24. In addition, the 6-month confirmed EDSS progression will be assessed.

MRI data: The mean number of Gd+ and new or newly enlarging T2 lesions over the 12- and 24-month periods will be summarized descriptively. The proportion of participants with no Gd+ lesions will be presented together with the corresponding 95% CI. The change from baseline over the 12- and 24-month periods in R2t* will also be reported.

The analytical approach will be further detailed in the SAP.

9.4.4. Tertiary/Exploratory Endpoints

Exploratory endpoints will be analyzed at Week 10 (Interim Analysis 1), Month 12 (Interim Analysis 2), and Month 24 (final analysis). Data will be summarized descriptively.

The (optional) exploratory endpoint for vaccine responses will be analyzed when a minimum of 15 participants have provided pre-vaccine through Week 4 samples (Interim Analysis) and when final samples have been collected from all participants (Final Analysis).

The analytical approach will be further detailed in the SAP.

9.4.5. Other Safety Analyses

All safety analyses will be made on the Safety Population. All safety data (AEs and routine hematology data) will be evaluated descriptively. For each DMC meeting, tables and listings of AEs, UPs, and SAEs will be prepared. Full details will be provided in a separate DMC Charter.

9.5. Interim Analyses

There will be 2 interim analyses; the primary, secondary, and exploratory endpoints will be assessed at each interim analysis. All available data for participants starting treatment with cladribine tablets will be included in each of the analyses. Data cleaning will be undertaken on all data involved in interim reporting.

The first interim analysis will be performed after Group 1 and Group 2 participants have completed their second LP (after Week 10). The analysis of the Week 5 LP data (Group 1 participants) is expected to provide information on the concentration of cladribine in the CSF. The data from the Week 10 LP (Group 2 participants) will provide information on the early impact of cladribine on neuronal injury and immune markers in the CSF at the time of the expected nadir of immune cell counts.

The second interim analysis will be performed after Group 3 participants have completed their second LP (after Month 12), to evaluate the persistence of effects of cladribine tablets in the CSF.

The final analysis will be on all participants' data after Month 24.

The SAP will describe the planned interim analyses in greater detail.

9.6. Data Monitoring Committee

A DMC will be established as an independent external committee appointed by the sponsor. It is anticipated that the committee will be composed of at least 2 members who will have complementary skills in the area of neurology, biostatistics, and/or clinical trial methodology. DMC objectives include the following:

1. To protect participants' safety by anticipating, identifying, and responding to potential participant safety issues
2. To review study outcomes and SAEs at a prearranged schedule
3. To evaluate new clinical research evidence that may become available during the course of the trial with regard to impact of this evidence on the ethics and feasibility of continuing the study; and
4. Ultimately, to periodically recommend if the study should be continued without modification, continued with modification, or stopped based on an overall assessment of safety.

Tables and listings of AEs, UPs, and SAEs will be prepared for each DMC meeting to facilitate meeting these objectives. Full details will be provided in a separate DMC Charter.

10. Supporting Documentation and Operational Considerations

10.1. Appendix 1: Regulatory, Ethical, and Study Oversight Considerations

10.1.1. Regulatory and Ethical Considerations

The study will be conducted in accordance with the protocol and with the following:

- Consensus ethical principles derived from international guidelines including the Declaration of Helsinki and Council for International Organizations of Medical Sciences International Ethical Guidelines
- Applicable ICH guidelines on GCP
- Applicable laws and regulations.

The protocol, protocol amendments, ICF, Investigator's Brochure, and other relevant documents (e.g. advertisements) must be submitted to an IEC/IRB by the investigator and reviewed and approved by the IEC/IRB before the study is initiated.

Any amendments to the protocol will require IEC/IRB approval before implementation of changes made to the study design, except for changes necessary to eliminate an immediate hazard to study participants.

The investigator will be responsible for the following:

- Providing written summaries of the status of the study to the IEC/IRB annually or more frequently in accordance with the requirements, policies, and procedures established by the IEC/IRB
- Notifying the IEC/IRB of UPs, SAEs, or other significant safety findings as required by IEC/IRB procedures
- Providing oversight of the conduct of the study at the site and adherence to requirements of 21 CFR, ICH guidelines, the IEC/IRB, European regulation 536/2014 for clinical studies (if applicable), and all other applicable local regulations.

10.1.2. Financial Disclosure

Investigators will provide the sponsor with sufficient, accurate financial information as requested to allow the sponsor to submit complete and accurate financial certification or disclosure statements to the appropriate regulatory authorities.

Details on investigators' responsibilities will be provided in the site contracts.

10.1.3. Informed Consent Process

The investigator or his/her representative will explain the nature of the study, including the potential risks and benefits of the required LPs, to the participant or his/her legally authorized representative and answer all questions regarding the study.

Participants must be informed that their participation is voluntary. Participants or their legally authorized representative will be required to sign a statement of informed consent that meets the

requirements of 21 CFR 50, local regulations, ICH guidelines, Health Insurance Portability and Accountability Act (HIPAA) requirements, where applicable, and the IEC/IRB or study center.

It must be documented that written informed consent was obtained before the participant was enrolled in the study and the date the written consent was obtained. The authorized person obtaining the informed consent must also sign the ICF.

Participants must be consented to the most current version of the ICF during their participation in the study. In the event of an ICF amendment, participants may need to be re-consented if mandated by the IRB. A copy of the ICF(s) must be provided to the participant or the participant's legally authorized representative.

The ICF will contain separate sections that address

- The use of remaining mandatory CSF and blood samples for optional exploratory research. The investigator or authorized designee will explain to each participant the objectives of the exploratory research.
- The performance of an optional third LP at the end of Year 2 for participants randomly assigned to LP Groups 1-3.
- The optional use of CSF and blood samples for single cell RNA sequencing.
- The optional storage of RNA samples obtained from additional CSF and blood samples for future research.
- The optional collection of vaccine samples

For each of these optional procedures, participants will be told that they are free to refuse to participate in any of them separately, and that they may withdraw their consent at any time and for any reason during the storage period.

Separate signatures will be required to document a participant's agreement to allow any remaining specimens to be used for exploratory research, or any of the other optional procedures outlined above. Participants who decline to participate in one or more of the optional research procedures will not provide this separate signature for the respective part.

10.1.4. Data Protection

Participants will be assigned a unique identifier. Any participant records or datasets that are transferred to the sponsor or to any vendor will contain the identifier only; participant names or any information that would make the participant identifiable will not be transferred.

The participant must be informed that his/her personal study-related data will be used by the sponsor in accordance with local data protection law. The level of disclosure must also be explained to the participant who will be required to give consent for their data to be used as described in the ICF.

The participant must be informed that his/her medical records may be examined by Clinical Quality Assurance auditors or other authorized personnel appointed by the sponsor, by appropriate IEC/IRB members, and by inspectors from regulatory authorities.

10.1.5. Dissemination of Clinical Study Data

This study will be conducted in accordance with the FDA Amendments Act of 2007 (FDAAA), Clinical Trials Registration and Results Information Submission rule. As such, this trial will be registered at ClinicalTrials.gov, and results information from this trial will be submitted to ClinicalTrials.gov.

Preparation and disclosure of the Clinical Study Report will be performed following all legal and regulatory requirements.

In addition, every attempt will be made to publish results in peer-reviewed journals.

Data from this study may be requested from other researchers after the completion of the study. Access to analyzable datasets will be granted through a secure system, following an independent assessment of the scientific merit of a rigorously defined research question from a third party.

Details on the Clinical Study Report preparation, dissemination of data and their use for subsequent research, and the publication policy (Section 10.1.9) will be provided in the site contracts.

10.1.6. Data Quality Assurance

All participant data relating to the study will be recorded on printed or electronic CRF unless transmitted to the sponsor or designee electronically (e.g. laboratory data). The investigator is responsible for verifying that data entries are accurate and correct by physically or electronically signing the CRF.

The investigator must maintain accurate documentation (source data) that supports the information entered in the CRF.

The investigator must permit study-related monitoring, audits, IEC/IRB review, and regulatory agency inspections, and has to provide direct access to source data documents.

As the sponsor, Washington University will be responsible for ensuring that participating sites are adequately monitored, utilizing a risk-based approach that includes a combination of remote central monitoring of data submitted to the sponsor and on-site monitoring.

The investigator will permit authorized representatives of Washington University, local health authorities, and industry partners to inspect relevant facilities and records.

The sponsor or designee is responsible for the data management of this study including quality checking of the data.

The sponsor assumes accountability for actions delegated to other individuals (for example, contract research organizations).

Records and documents, including signed ICFs, pertaining to the conduct of this study must be retained by the investigator for 7 years after study completion unless local regulations or institutional policies require a longer retention period. No records may be destroyed during the retention period without the written approval of the sponsor. No records may be transferred to another location or party without written notification to the sponsor.

10.1.7. Source Documents

Source documents provide evidence for the existence of the participant and substantiate the integrity of the data collected. Source documents are filed at the investigator's site.

Data entered in the eCRF that are transcribed from source documents must be consistent with the source documents or the discrepancies must be explained. Some source documents, with patient identifiers removed, will be required to be included with the eCRF. The investigator may need to request previous medical records or transfer records, depending on the study. Also, current medical records must be available.

10.1.8. Study and Site Start and Closure

The study start date is the date on which the clinical study will be open for recruitment of participants.

The first act of recruitment is the first site open and will be the study start date.

The sponsor or designee reserves the right to close the study site or terminate the study at any time for any reason at the sole discretion of the sponsor. Study sites will be closed upon study completion. A study site is considered closed when all required documents and study supplies have been collected and a study-site closure visit has been performed.

The investigator may initiate study-site closure at any time, provided there is reasonable cause and sufficient notice is given in advance of the intended termination.

Reasons for the early closure of a study site by the sponsor or investigator may include but are not limited to:

- Failure of the investigator to comply with the protocol, the requirements of the IEC/IRB or local health authorities, the sponsor's procedures, or GCP guidelines
- Inadequate recruitment of participants by the investigator
- Discontinuation of further study intervention development.

If the study is prematurely terminated or suspended, the sponsor shall promptly inform the investigators, the IECs/IRBs, the regulatory authorities, and any contract research organization(s) used in the study of the reason for termination or suspension, as specified by the applicable regulatory requirements. The investigator shall promptly inform each participant and should assure appropriate therapy and/or follow-up for each participant.

10.1.9. Publication Policy

The results of this study may be published or presented at scientific meetings. If this is foreseen, the investigator agrees to submit all manuscripts or abstracts to the sponsor before submission. Authorship will be determined by mutual agreement and in line with International Committee of Medical Journal Editors authorship requirements.

Details regarding publication generation and submission will be described in the site contracts.

10.2. Appendix 2: Clinical Laboratory Tests

Protocol-required routine laboratory safety assessments, as summarized in Table 7, include all assessments that are required per the approved USPI for cladribine tablets. Unless notified otherwise, these assessments will be performed by the local laboratories at each site.

Assessments should be scheduled as described in the SoA (Section 1.3). Additional tests may be performed at any time during the study as determined necessary by the investigator or required by local regulations.

Investigators must document their review of each laboratory safety report.

Protocol-specific requirements for inclusion or exclusion of participants are detailed in Section 5.

Table 7. Protocol-Required, Safety Local Laboratory Assessments

Laboratory assessments	Parameters	
Hematology (local laboratory)	Absolute lymphocyte count	Additionally from complete blood count (will be recorded on eCRF) • White blood cells • Platelets • Hemoglobin • Hematocrit
Biochemistry (local laboratory)[a]	• Aminotransferase • Child-Pugh score assessment	
Other screening tests (local laboratory)[a]	• Pregnancy test (per routine practice in local laboratory) • Serology: HIV test, hepatitis B and C, tuberculin test; varicella zoster antibody	

[a] These USPI-required assessments are used to assess eligibility for cladribine tablets treatment only

eCRF=electronic Case Report Form, HIV=human immunodeficiency virus, USPI=United States Prescribing Information.

10.3. Appendix 3: Adverse Events and Unanticipated Problems: Definitions and Procedures for Recording, Evaluating, Follow-up, and Reporting

10.3.1. Definition of Adverse Events

An AE is any untoward medical occurrence in a participant administered a pharmaceutical product, regardless of causal relationship with this treatment. An AE can therefore be any unfavorable and unintended sign (including an abnormal laboratory finding), symptom, or disease temporally associated with the use of a medicinal product, regardless if it is considered related to the medicinal product.

For surgical or diagnostic procedures, the condition/illness leading to such a procedure is considered as the AE rather than the procedure itself.

The investigator is required to grade the severity or toxicity of each AE. Investigators must assess the severity of AEs according to the Qualitative Toxicity Scale, as follows:

- Mild: The participant is aware of the event or symptom, but the event or symptom is easily tolerated.
- Moderate: The participant experiences sufficient discomfort to interfere with or reduce his or her usual level of activity.
- Severe: Significant impairment of functioning: the participant is unable to carry out his or her usual activities.

If death occurs, the primary cause of death or event leading to death should be recorded and reported as an SAE. “Fatal” will be recorded as the outcome of this specific event and death will not be recorded as separate event. Only, if no cause of death can be reported (e.g. sudden death, unexplained death), the death per se might then be reported as an SAE.

Investigators must also systematically assess the causal relationship of AEs to study intervention using the following definitions. Decisive factors for the assessment of causal relationship of an AE to cladribine tablets or to the LP include, but may not be limited to, temporal relationship between the AE and the study intervention, known side effects of cladribine tablets, medical history, concomitant medication, course of the underlying disease, and study procedures.

Unrelated: Not reasonably related to the study intervention. AE could not medically (pharmacologically/clinically) be attributed to the study intervention under study in this clinical study protocol. A reasonable alternative explanation must be available.

Related: Reasonably related to the study intervention. AE could medically (pharmacologically/clinically) be attributed to the study intervention under study in this clinical study protocol.

Abnormal Laboratory Findings and Other Abnormal Investigational Findings

Abnormal laboratory findings and other abnormal investigational findings (for example, on an ECG trace) should not be reported as AEs unless they are associated with clinical signs and symptoms, lead to study intervention discontinuation or are considered otherwise medically

important by the investigator. If a laboratory abnormality fulfills these criteria, the identified medical condition (e.g. anemia or increased alanine aminotransferase [ALT]) must be reported as the AE rather than the abnormal value itself.

10.3.2. Definition of Serious Adverse Events

An SAE is any untoward medical occurrence that at any dose:

- Results in death
- Is life-threatening. Life-threatening refers to an event in which the participant is at risk of death at the time of the event, not an event that hypothetically might have caused death if it was more severe.
- Requires inpatient hospitalization or prolongs an existing hospitalization
- Results in persistent or significant disability or incapacity
- Is a congenital anomaly or birth defect
- Is otherwise considered to be medically important. Important medical events that may not result in death, be life-threatening, or require hospitalization may be considered as SAEs when, based upon appropriate medical judgment, they may jeopardize the participant or may require medical or surgical intervention to prevent one of the outcomes listed above. Examples of such events include allergic bronchospasm requiring intensive treatment in an emergency room or at home, blood dyscrasias, or convulsions that do not result in inpatient hospitalization, or the development of drug dependency or drug abuse.

For the purposes of reporting, any suspected transmission of an infectious agent via a study intervention is also considered an SAE, as specified below for reporting SAEs, adverse events of special interest, and dose-limiting toxicities.

Events That Do Not Meet the Definition of an SAE

Elective hospitalizations to administer, or to simplify study intervention or procedures (e.g. an overnight stay to facilitate IV therapy), are not considered SAEs. However, all events leading to unplanned hospitalizations or unplanned prolongation of an elective hospitalization (i.e. undesirable effects of any administered treatment) must be documented and reported as SAEs.

10.3.3. Events Not to Be Considered as AEs/SAEs

Medical conditions present at the initial study visit that do not worsen in severity or frequency during the study are defined as Baseline Medical Conditions, and are not to be considered AEs.

In this protocol, symptoms and signs of relapse or worsening of MS will usually be captured in the context of the efficacy assessment, and recorded on the relapse module of the eCRF. Symptoms, relapses, or worsening of MS (including laboratory abnormalities) will therefore not be considered as AEs nor captured on the AE module of the eCRF, unless the participant's general condition is more severe than expected for the participant's condition (i.e. worsening is not consistent with the anticipated natural progression of the disease), and/or unless the outcome is fatal within the AE reporting period (see Section 8.3.1).

10.3.4. Definition of an Unanticipated Problem

An Unanticipated Problem is considered to include any incident, experience, or outcome that meets all of the following criteria:

- Unexpected (in terms of nature, severity, or frequency) given the research procedures that are described in the protocol-related documents, such as the IRB-approved research protocol and informed consent document, and the characteristics of the subject population being studied.
- Related to participation in the research; and
- Suggests that the research places subjects or others at a greater risk of harm than was previously known or recognized.

10.3.5. Recording and Follow-Up of AE, UP and/or SAE

It is important that each AE/UP/SAE report include a description of the event, its duration (onset and resolution dates and also onset and resolution times, when it is important to assess the time of AE/UP onset relative to the recorded study intervention administration time), its severity, its causal relationship with the study intervention, any other potential causal factors, any treatment given or other action taken, including dose modification or discontinuation of the study intervention, and its outcome. In addition, serious cases should be identified and the appropriate seriousness criteria documented.

10.3.6. Adverse events of Special Interest

Not applicable.

10.3.7. Reporting Unanticipated Problems and Serious Adverse Events

In the event of any new UP/SAE occurring during the reporting period, the investigator must immediately (within a maximum of 1 working day after becoming aware of the event) inform the sponsor or its designee, using the electronic UP/SAE Report Form in the eCRF, which must be completed by the investigator following specific completion instructions.

In case the eCRF is not available, the UP/SAEs must be reported via email using the paper SAE Report Form following specific completion instructions. Also in exceptional circumstances, a UP/SAE (or follow-up information) may be reported by telephone; in these cases the eCRF must be completed as soon as it becomes available.

Relevant pages from the CRF may be provided in parallel (for example, medical history, concomitant drugs). Additional documents may be provided by the investigator, if available (for example, laboratory results, hospital report, or autopsy report). In all cases, the information provided on the UP/SAE Report Form must be consistent with the data about the event recorded in the eCRF.

The investigator must respond to any request for follow-up information (for example, additional information, outcome, final evaluation, other records where needed) or to any question the sponsor/designee may have on the UP/AE within the same timelines as those noted above for initial reports. This is necessary to ensure prompt assessment of the event by the sponsor or designee and (as applicable) to allow the sponsor to meet strict regulatory timelines associated

with expedited safety reporting obligations. The sponsor is responsible for safety reporting, has to identify and follow up on ADRs/SAEs experienced by participants in the study, and has to forward the information to EMD Serono as specified in the collaboration contract between EMD Serono and the sponsor.

Requests for follow-up will usually be made via the responsible monitor, although in exceptional circumstances the EMD Serono Drug Safety department may contact the sponsor directly to obtain further information or to discuss the event.

10.4. Appendix 5: Abbreviations

ADR(s)	Adverse drug reaction(s)
AE	Adverse event
ALC	Absolute lymphocyte count
ARR	Annualized relapse rate
CBC	Complete blood count
CD	Cluster of differentiation
CI	Confidence interval
CNS	Central nervous system
CRF	Case Report Form
CSF	Cerebrospinal fluid
DMC	Data Monitoring Committee
DMD	Disease-modifying drug
ECG	Electrocardiogram
eCRF	Electronic Case Report Form
EDSS	Expanded Disability Status Scale
FDA	Food and Drug Administration
GCP	Good Clinical Practice
Gd+	Gadolinium-enhancing
HIV	Human immunodeficiency virus
ICF	Informed Consent Form
ICH	International Council for Harmonisation of Technical Requirements for Pharmaceuticals for Human Use
IEC	Independent Ethics Committee
IFN	Interferon

Ig	Immunoglobulin
IRB	Institutional Review Board
IWRS	Interactive Web Response System
IV	Intravenous
LP	Lumbar puncture
MoA	Mechanism of action
MRI	Magnetic resonance imaging
MS	Multiple sclerosis
NfH	Neurofilament heavy chain
NfL	Neurofilament light chain
Tregs	Regulatory T cells
PML	Progressive multifocal leukoencephalopathy
RMS	Relapsing forms of MS
RRMS	Relapsing-remitting MS
SAE	Serious adverse event
SAP	Statistical Analysis Plan
SoA	Schedule of Activities
TEAE	Treatment-emergent adverse event
Th cell	T helper cell
UP	Unanticipated Problem
US	United States
USPI	United States Prescribing Information

11. References

Alcolea D, Martínez-Lage P, Izagirre A, et al (2014). Feasibility of lumbar puncture in the study of cerebrospinal fluid biomarkers for Alzheimer's disease: a multicenter study in Spain. J Alzheimers Dis. 39:719-26.

Alvermann S, Hennig C, Stüve O, et al (2014). Immunophenotyping of cerebrospinal fluid cells in multiple sclerosis: in search of biomarkers. JAMA Neurol. 71:905-12.

Avsar T, Korkmaz D, Tütüncü M, et al (2012). Protein biomarkers for multiple sclerosis: semi-quantitative analysis of cerebrospinal fluid candidate protein biomarkers in different forms of multiple sclerosis. Mult Scler. 18(8):1081-91.

Cai L, Huang J (2018). Neurofilament light chain as a biological marker for multiple sclerosis: a meta-analysis study. Neuropsychiatr Dis Treat. 14:2241-54.

Comi G, Cook S, Giovannoni G, et al (2019). Effect of cladribine tablets on lymphocyte reduction and repopulation dynamics in patients with relapsing multiple sclerosis. Mult Scler Relat Disord. 29:168-74.

Doherty CM, Forbes RB (2014). Diagnostic lumbar puncture. Ulster Med J. 83:93-102.

Duits FH, Martinez-Lage P, Paquet C, et al (2016). Performance and complications of lumbar puncture in memory clinics: results of the multicenter lumbar puncture feasibility study. Alzheimers Dement. 12:154-63.

EMD Serono, Inc: Cladribine Tablets. United States Prescribing Information, 2019. Available at: https://www.drugs.com/pro/cladribine.html. Accessed on: April 15, 2019.

Engelborghs S, Niemantsverdriet E, Struyfs H, et al (2017). Consensus guidelines for lumbar puncture in patients with neurological diseases. Alzheimers Dement (Amst). 8:111-26.

Giovannoni G, Soelberg Sorensen S, Cook S, et al (2018). Safety and efficacy of cladribine tablets in patients with relapsing-remitting multiple sclerosis: Results from the randomized extension trial of the CLARITY study. Mult Scler. 24:1594-1604.

Giovannoni G, Comi G, Cook S, et al (2010). A placebo-controlled trial of oral cladribine for relapsing multiple sclerosis. N Engl J Med. 362:416-26.

Han S, Lin YC, Wu T, et al (2014). Comprehensive immunophenotyping of cerebrospinal fluid cells in patients with neuroimmunological diseases. J Immunol. 192(6):2551-63.

Hyun JW, Kim Y, Kim G, et al (2019). Longitudinal analysis of serum neurofilament light chain: A potential therapeutic monitoring biomarker for multiple sclerosis. Mult Scler. Mar 26, 2019 [epub ahead of print].

Howell JC, Parker MW, Watts KD, et al (2016). Research lumbar punctures among African Americans and Caucasians: Perception predicts experience. Front Aging Neurosci. 8:296.

Kingwell E, Marriott JJ, Jetté N, et al (2013). Incidence and prevalence of multiple sclerosis in Europe: a systematic review. BMC Neurol 13:128.

Kuhle J, Kropshofer H, Haering DA, et al (2019). Blood neurofilament light chain as a biomarker of MS disease activity and treatment response. Neurology. 92:e1007-15.

Kurtzke JF (1983). Rating neurologic impairment in multiple sclerosis: an expanded disability status scale (EDSS). Neurology. 33:1444-52.

Leist TP, Comi G, Cree BA, et al (2014). Effect of oral cladribine on time to conversion to clinically definite multiple sclerosis in patients with a first demyelinating event (ORACLE MS): a phase 3 randomised trial. Lancet Neurol. 13:257-67.

Lublin FD, Reingold SC, Cohen JA, et al (2014). Defining the clinical course of multiple sclerosis: the 2013 revisions. Neurology. 83:278-86.

Meyer-Moock S, Feng YS, Maeurer M, Dippel FW, Kohmann T (2014). Systematic literature review and validity evaluation of the Expanded Disability Status Scale (EDSS) and the Multiple Sclerosis Functional Composite (MSFC) in patients with multiple sclerosis. BMC Neurol: 14:58.

Menéndez-González M (2014). Routine lumbar puncture for the early diagnosis of Alzheimer's disease. Is it safe? Front Aging Neurosci. 6:65.

Multiple Sclerosis International Federation (2013). Atlas of MS. Available at: https://www.msif.org/about-us/who-we-are-and-what-we-do/advocacy/atlas/. Accessed on: April 15, 2019.

Noseworthy JH, Lucchinetti C, Rodriguez M, et al (2000). Multiple sclerosis. N Engl J Med. 343:938-52.

Novakova L, Axelsson M, Khademi M, et al (2017). Cerebrospinal fluid biomarkers as a measure of disease activity and treatment efficacy in relapsing-remitting multiple sclerosis. J Neurochem. 141:296-304.

Pardo G, Jones DE (2017). The sequence of disease-modifying therapies in relapsing multiple sclerosis: safety and immunologic considerations. J Neurol. 264:2351-74.

Przybek J, Gniatkowska I, Mirowska-Guzel D, et al (2015). Evolution of diagnostic criteria for multiple sclerosis. Neurol Neurochir Pol. 49:313-21.

Rejdak K, Stelmasiak Z, Grieb P (2019). Cladribine induces long lasting oligoclonal bands disappearance in relapsing multiple sclerosis patients: 10-year observational study. Mult Scler Relat Disord. 27:117-20.

Scalfari A, Knappertz V, Cutter G, et al (2013). Mortality in patients with multiple sclerosis. Neurology. 81:184-92.

Tsvetkova DZ, Bergquist SH, Parker MW, et al (2017). Fear and uncertainty do not influence reported willingness to undergo lumbar punctures in a U.S. multi-cultural cohort. Front Aging Neurosci. 9:22.

Tumani H, Hartung HP, Hemmer B, et al (2009). Cerebrospinal fluid biomarkers in multiple sclerosis. Neurobiol Dis. 35(2):117-27.

Wallin MT, Culpepper WJ, Campbell J, et al (2017). The prevalence of multiple sclerosis in the United States: a population-based healthcare database approach. Abstract. Multiple Sclerosis Journal. 23(S3):125.