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Protocol for an exploratory, randomised, single-blind clinical trial of aerobic exercise to promote remyelination in multiple sclerosis

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ABSTRACT

Introduction There is an urgent need for remyelinating therapies that restore function in people with multiple sclerosis (pwMS). Aerobic exercise is a promising remyelinating strategy because it promotes remyelination in animal models both independently and synergistically with medications. Here, in this study, we present an innovative, randomised, single-blind, clinical trial designed to explore: the relationship between demyelination and mobility (part 1), and if 24 weeks of aerobic exercise promotes remyelination in pwMS (part 2).

Methods and analysis Sedentary participants (n=60; aged 18–64 years) with stable MS will undergo a baseline visit with the following outcomes to assess associations between demyelination and mobility (part 1): spinal cord demyelination (somatosensory-evoked potentials, SSEPs), mobility (6-Minute Timed Walk, Timed 25-Foot Walk, Timed Up and Go, 9-Hole Peg Test) and patient-reported outcomes (PROs). After baseline testing, participants with significantly prolonged SSEP latency will advance to the clinical exercise trial (part 2) and will be randomised 1:1 to active or control conditions for 24 weeks. The active condition will be aerobic stationary cycling three times per week with graded virtual supervision. The control condition will be monthly virtual MS symptom education groups (six sessions). SSEP latency (remyelination endpoint), mobility outcomes and PROs will be measured at 12 and 24 weeks in all clinical trial participants. A subset of 11 active and 11 control participants will undergo a brain MRI with quantitative T_1 myelin water fraction at baseline and 24 weeks (exploratory remyelination endpoint).

Ethics and dissemination Ethical approval was obtained from the Oregon Health & Science University Institutional Review Board (#21045). Dissemination of findings will include peer-reviewed publications, conference presentations and media releases. The proposed study will inform the feasibility, study design and sample size for a fully powered clinical trial of aerobic exercise to promote remyelination in pwMS.

Trial registration number NCT04539002.

STRENGTHS AND LIMITATIONS OF THIS STUDY

⇒ Strengths of this study include an aerobic intervention that is based on an individual's maximum heart rate and an active control condition (the multiple sclerosis education control group) and blinded conduct of outcome measures.
⇒ Another strength of this study is the blinded analysis of structural (brain myelin water fraction) and functional (somatosensory-evoked potentials) remyelination outcomes.
⇒ Limitations of this study include a small participant number that may not detect a small effect size on clinical or remyelination outcomes, limited geographical catchment area, and not examining the durability of the effects of the aerobic exercise intervention.

INTRODUCTION

Multiple sclerosis (MS) is the most common non-traumatic cause of neurological disability in young adults, affecting around 2.8 million people worldwide.[1] In MS, there is intermittent, focal inflammation that leads to demyelination and axonal injury, and ultimately results in disability.[2] Remyelination, however, improves conduction velocity and also provides neuroprotection across the axon; therefore, remyelination has the potential to be an effective long-term strategy to both improve and protect against future disability in people with MS (pwMS).[3 4]

Numerous clinical trials demonstrate that aerobic exercise can improve physical fitness, fatigue, depression and walking mobility,[5 6] and may also reduce the rate of relapse and disease progression in MS.[7–9] Yet, the mechanisms underlying the effects of exercise/physical activity remains a high priority question for wellness research in MS.[10]

In preclinical models of MS, aerobic exercise promotes remyelination, both alone and synergistically with pharmacotherapy. In a lysolecithin-induced demyelination study in mice, aerobic exercise was associated

with remyelination of 84% of the demyelinated axons compared with 51% of the axons that remyelinated spontaneously without exercise. The antihistamine clemastine also promoted remyelination, resulting in 87% axonal remyelination, but adding aerobic exercise to clemastine was even more effective, resulting in 98% axonal remyelination. Additionally, the myelin formed after exercise alone or after exercise with clemastine was thicker than the myelin formed in the mice that did not exercise, supporting that exercise promoted not only more complete, but also more robust remyelination.[11] Aerobic exercise may promote remyelination through various mechanisms including inhibition of auto-immune T-cell and blood brain barrier disruption, optimisation of the microenvironment by increased removal of myelin debris, increased oligodendrocyte precursor cell proliferation, increased local neurotrophic factors and activation of pathways that increase myelin thickness.[11–17] To our knowledge, no clinical trials of aerobic exercise have explored remyelination as the primary outcome in pwMS.

Remyelination clinical trials also require real-world treatment biomarkers that are reliable, valid, reproducible, affordable and correlate to clinically meaningful outcomes. Somatosensory-evoked potential (SSEP) latencies from the upper and lower extremities meet many of these requirements as they are reliable, estimate demyelination[18] and are suitable for multicentre trials.[19] SSEPs are also available for commercial use, so this outcome could be used clinically by MS providers to follow treatment response to remyelinating medications. Motor-evoked potentials (MEPs) are also promising potential remyelination biomarkers, but MEPs are dependent on participant arousal and voluntary contraction can lead to intra-individual variability and confound this measure.[20–22] While studies have correlated SSEP latencies to imbalance and fall frequency in pwMS, this will be the first study to examine the associations between SSEP latencies and a panel of mobility metrics in pwMS to inform clinical outcomes for future remyelination clinical trials in pwMS.[23 24]

We will conduct a randomised, single-blind, parallel group, exploratory clinical trial of aerobic stationary cycling compared with an MS education control group in sedentary pwMS. The purpose of this study is to explore the relationship between demyelination, as measured by SSEP, and clinical measures of mobility in pwMS to explore potential clinical outcomes in future remyelination trials (part 1) and, to explore if aerobic exercise improves mobility and promotes remyelination as measured by functional (SSEPs) and structural (myelin water fraction, MWF) measures (part 2). The central hypotheses of this study are that (1) greater spinal cord demyelination (indicated by longer SSEP latency) will be associated with increased motor disability in pwMS, and (2) the aerobic exercise group will demonstrate decreased SSEP latency, increased MWF and clinical improvement compared with an educational control group. This trial will inform the feasibility, clinical outcome measures, design and sample size for a fully powered clinical trial of aerobic exercise for remyelination in pwMS.

METHODS AND ANALYSIS

Study design

This study includes two parts. Part 1 is a baseline assessment to explore the relationship between demyelination, as measured by SSEP, and mobility in pwMS. Part 2 is a randomised controlled trial of an aerobic exercise intervention in those who have sensory demyelination at baseline, based on SSEP. Sixty participants will enter part 1 and undergo baseline testing, including SSEPs, mobility outcomes and patient-reported outcomes (PROs). The clinical outcome with the strongest correlation with SSEP latency in part 1 of the study will become the primary clinical outcome to measure remyelination in part 2 of the clinical trial. The first 44 participants with a qualifying lower extremity SSEP z-score will enter part 2 and be randomly allocated to an aerobic cycling intervention with graded supervision (MSCYCLE) or an education control group (MS Take Control, MSTC)[15 15] (figure 1). Randomisation to MSCYCLE and MSTC will be performed by an unblinded research assistant in a 1:1 ratio using a random number generator in blocks of four. All participants in part 2 will undergo repeat testing (SSEPs, mobility outcomes and PROs) at 12 and 24 weeks by study staff blinded to group assignment. A subset of clinical trial participants (11 active and 11 controls) will also be invited to undergo a brain MRI with MWF at baseline and 24 weeks. The first participant was enrolled in April 2021. We anticipate that recruitment will be complete by January 2024 and the last participant will exit the trial by July 2024.

Participants

Inclusion criteria

Participants are included in the study if they meet the following criteria: (1) diagnosis of MS based on the 2017 McDonald criteria, (2) aged between 18 and 64 years (in response to Institutional Review Board (IRB) feedback as those aged 65 years and above were considered to be at high risk of severe COVID-19) and (3) have access to the internet and a device that can access virtual visits.[25] The first 44 participants with a height-adjusted lower extremity SSEP latency z-score ≥2 or inter-side difference in z-score ≥2 for P40 will advance to part 2.

Exclusion criteria

Participants will be excluded from the study if they: (1) have medical or biophysical conditions that prohibit exercise, (2) already engage in >30 min/week of moderate or vigorous aerobic exercise, (3) have a clinically confirmed MS relapse in the past 3 months or change in their MS disease modifying therapy in the last 6 months, (4) are pregnant or are planning to become pregnant, (5) received steroids for MS relapse in the last 30 days, (6) have a known history of severe spinal canal stenosis (which would impact SSEPs) and (7) use 4-aminopyridine or

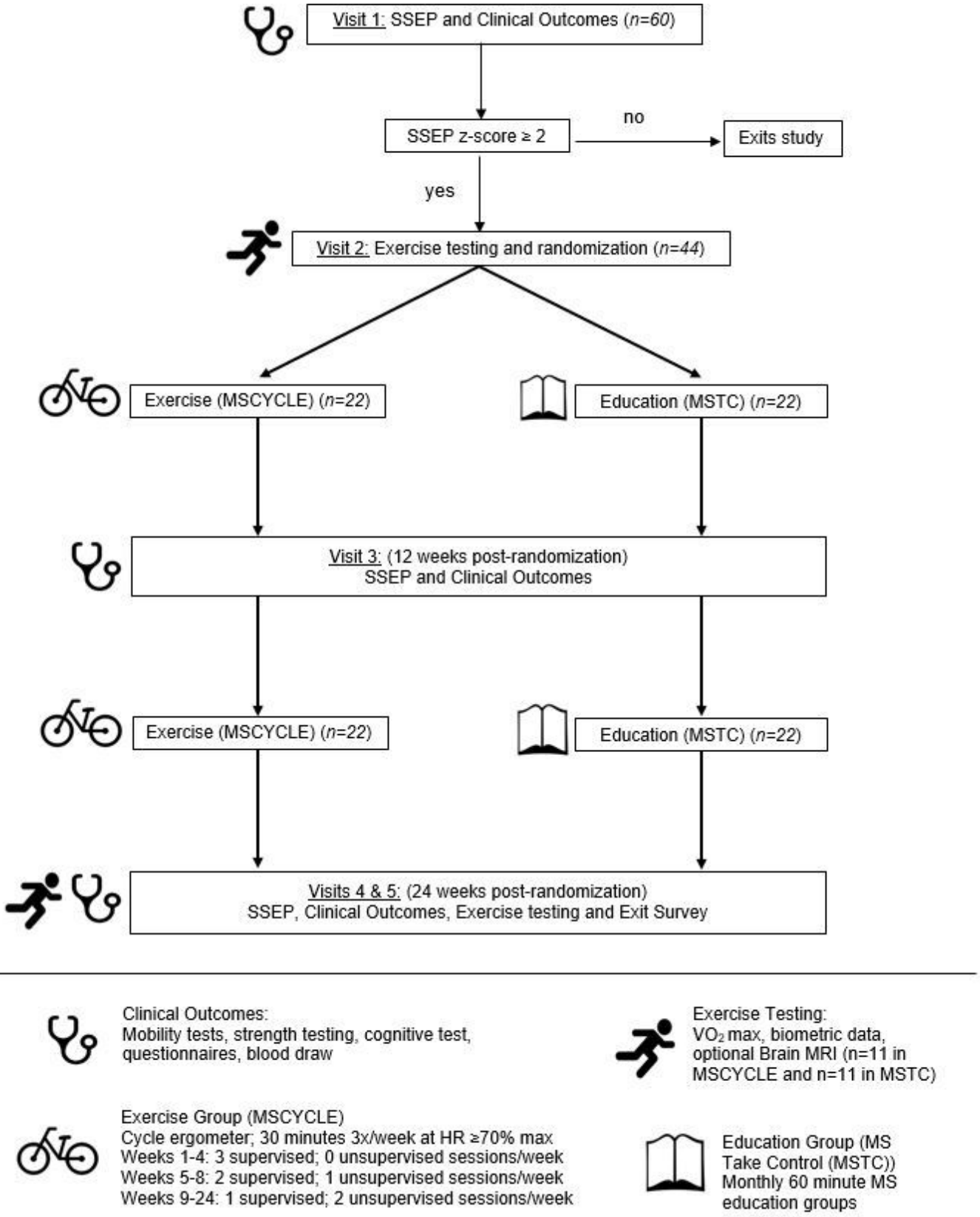

Figure 1 Participant flow through the study. MS, multiple sclerosis; MSTC, MS Take Control; SSEP, somatosensory-evoked potential; VO2max, maximal oxygen consumption

dalfampridine (medications which can temporarily alter SSEP) and unwilling to discontinue it for 2 days prior to each SSEP testing session.

Recruitment

Patients with MS will be recruited from National MS Society and the Oregon Health & Science University (OHSU) MS

Clinic, through OHSU MyChart recruitment invitations, and the OHSU MS Research Recruitment Repository with IRB-approved flyers. PwMS identified as trial candidates will be screened for meeting the inclusion and exclusion criteria including a Physical Activity Readiness Questionnaire (PAR-Q) to determine if the participant can safely commence an exercise routine.[26] If the patient answers 'YES' to any of the seven questions in this survey, we will consult with their doctor to determine if they are safe to engage in physical activity. If physician permission is not obtained, the patient will be excluded. Those interested in and determined safe for participation will be provided with the Informed Consent Form by study staff to review prior to signing the form at their baseline visit with study staff (see online supplemental material 1).

Sample size calculation

Part 1 of the study will explore the relationship of demyelination with mobility in pwMS. Based on prior studies associating transformed SSEP z-scores and the expanded disability status scale (rho=0.81, p<0.001) in pwMS, n=60 participants would have 88% power to detect a correlation rho=0.40 (α=0.05). All power analyses were computed using Power And Sample Size (PASS) software (V.15). For part 2 of the study (to explore if aerobic exercise improves mobility and promotes remyelination), we assume the effect size of z-transformed evoked potential values between baseline and 24 weeks is 0.54 in pwMS based on prior studies.[27] Assuming a 20% drop out rate, enrolling 44 participants in the clinical trial would achieve the necessary final sample size of 36 participants, or 18 per group. This will give us >80% power to detect a 50% effect size (α=0.05). Our sample size is similar to prior remyelination clinical trials of the pharmacotherapies clemastine and bexarotene in MS using visual evoked potentials, a similar functional myelination biomarker of the visual system.[28 29] As this is the first clinical trial of exercise to use SSEP to assess for remyelination, it is unknown if the study intervention, different evoked potential modality or other factors may impact the required sample size.

To our knowledge, this study is the first clinical trial in MS to use quantitative T_1 MWF as a biomarker of structural remyelination and therefore a well-informed power analysis cannot be performed. However, 11 per group is similar to other studies that have demonstrated changes in white matter integrity in pwMS after rehabilitative interventions.[30 31] If additional funding is obtained, the sample size of the MRI subset may be increased.

Interventions

Aerobic exercise group (MSCYCLE)

The active intervention, MSCYCLE, is a three times per week, home-based, stationary aerobic cycling intervention conducted over 24 weeks with virtual graded supervision by trained study personnel (hereto called the 'trainer') who will be blinded to the study outcomes. The trainer will provide supervision and collect study data (eg, minutes in the target HR range, report injuries/adverse events) via a video link.

Before starting the intervention, participants will undergo a VO_2max test to determine their HR_{peak} from which to calculate their target HR during the aerobic exercise intervention; the continuous exercise intervention target HR will be 70–80% of HR_{peak}.

Participants will be provided with a recumbent cycle ergometer and a chest strap to monitor their HR. The HR monitor will be linked to a smartphone application which will display their HR during the session and will also record minutes spent in their target HR zone. Participants will also be given an *Aerobic Training Guide* which will review study exercise procedures, injury prevention and includes a training log.

The MSCYCLE sessions will be conducted with graded virtual supervision by a trainer, progressing from three supervised sessions per week during Weeks 1–4, two supervised and one unsupervised session during Weeks 5–8, to one supervised and two unsupervised sessions per week during Week 9–24. The trainer will work with the participant to increase the heart rate range, as tolerated, on the following schedule: Week 1: 50–60% HR_{peak}; Week 2: 55–65% HR_{peak}; Week 3: 60–70% HR_{peak}; Week 4: 65+% HR_{peak}; Weeks 5–24 70–80% HR_{peak}.

During the supervised sessions, the trainer and participant will have a scheduled video visit to conduct the exercise intervention. The sessions will include a 5-min warm-up period, a 30-min continuous exercise period at their target HR range, followed by a 5-min cool down period. At the end of the exercise session, the trainer (or the participant if the session is unsupervised) will record the number of minutes during the 30-min active period that the participant was able to maintain their target HR. The exercise sessions will be modified as needed to accommodate injury or other changes in medical status, or discontinued if the participant is no longer able to exercise safely.

Control group (MSTC)

The control intervention, MSTC, is a monthly, virtual group of 60-min class with 1–9 participants led by a trained facilitator, conducted on a secure virtual platform. We have used this intervention as a control in other studies.[32]

For MSTC, participants will be given informational materials at the time of the randomisation and be instructed to read one of the following National MS Society pamphlets before each of the 6 monthly sessions: *Driving with MS*, *Managing Cognitive Problems in MS*, *Taming Stress*, *Clear Thinking About Alternative Therapies*, *Urinary Dysfunction and MS* and *Vitamins, Minerals and Herbs in MS: An Introduction*. These topics were chosen as they are of interest to pwMS and the pamphlets provide high quality information produced specifically for pwMS. One pamphlet will be reviewed by the group during each 60-min session.

Participants in both MSCYCLE and MSTC will be instructed to maintain their current activity levels outside

Table 1 Study activities

	#1	#2	Intervention		#3	Intervention		#4	#5
Study visit #	**Baseline**				**Week 12**			**Week 24**	
Consent	X		MSCYCLE Stationary aerobic cycling ×3/ week with graded supervision	MS: Take Control (MSTC) Monthly MS-education control group		MSCYCLE Stationary aerobic cycling ×3/ week with graded supervision	MS: Take Control (MSTC) Monthly MS-education control group		
Medical and MS history	X	X			X			X	X
Patient-derived disease steps	X								
Inclusion/exclusion and review adverse events	X				X			X	
Biometric data		X							X
7-Site Skinfold Testing		X							X
VO_2max		X							X
Strength testing	X				X			X	
Somatosensory-evoked potential	X				X			X	
6-Minute Timed Walk*	X				X			X	
Timed Up and Go*	X				X			X	
Timed 25 Foot Walk	X				X			X	
9-Hole Peg Test	X				X			X	
Postural sway*	X				X			X	
Retrospective 30-day fall frequency	X				X			X	
Symbol Digit Modalities Test	X				X			X	
International Physical Activity Questionnaire-Short Form	X				X			X	
Modified Fatigue Impact Scale	X				X			X	
Pain Effects Scale	X				X			X	
Neuro-QOL Lower Extremity Function	X				X			X	
Neuro-QOL Upper Extremity Function	X				X			X	
Neuro-QOL Depression	X				X			X	
Neuro-QOL Sleep Disturbance	X				X			X	
PROMIS Self-Efficacy questionnaire	X				X			X	
Blood draw	X	X			X			X	
MRI for Myelin Water Fraction (MWF)†		X							X
Exit survey									X

*Performed with body-worn sensors.
†Performed on a subset of trial participants (n=22).
MS, multiple sclerosis; QOL, quality of life.

of the sessions during the trial, but are permitted to participate in physical or occupational therapy.

Outcomes

All outcomes will be conducted and interpreted by study staff blinded to group allocation (table 1).

Primary outcomes

SSEP

SSEP will be performed at baseline, and at 12 and 24 weeks to assess remyelination (Cadwell Elite, Kennewick, Washington, USA). SSEP will be recorded after median nerve and posterior nerve electrical stimulation starting with a square pulse of 0.2 ms duration at a rate of 3.11 Hz at 2 mA above motor threshold. The averages of two or more subsequent runs (at least 2×500 pulses) will be inspected for reproducibility and the mean will be used for analysis. For the upper extremities, active surface electrodes will be placed at C3'/C4' contralateral to stimulation (reference: Fz) to obtain N20. For the lower extremities, active surface electrodes will be placed at Cz (reference: Fz) to obtain P40. Recordings will follow the recommendations of the American Clinical Neurophysiology Society.[33] Prolonged latencies of N20 or P40 reflect demyelination within the upper and lower extremity sensory pathways,

respectively.[20] SSEP latency z-scores for each limb will be calculated using previously established normative values.[34]

Mobility and disability outcomes

Mobility tests will be conducted at baseline, 12 and 24 weeks. Exercise endurance will be evaluated with the 6-Minute Timed Walk (6MTW).[35] Participants will be asked to walk for 6 min and the total distance will be recorded. Walking mobility will be measured with the Timed Up and Go (TUG).[36] Participants will be asked to stand up from a chair, walk 7 metres, turn, return to the chair, turn and sit down. The average of two timed tests will be recorded. Walking speed will be measured by the Timed 25-Foot Walk (T25FW).[37] To estimate fall frequency, participants will be asked how many falls they have sustained in the prior 30 days. Upper extremity mobility will be measured with the 9 Hole Peg Test (9HPT).[37] During this test the time for participants to place nine pegs into a platform with holes and then take them out again will be measured. MS disability will be measured with the MS Functional Composite (MSFC), a three-part, standardised, quantitative, assessment instrument in MS clinical trials measuring cognition, leg function and arm/hand function and will be calculated using the T25FW, 9HPT and Symbol Digit Modalities Test (SDMT, described in *Cognitive Function* below).[37 38]

Participants will perform the 6MTW and TUG wearing wireless, synchronised inertial sensors on both wrists and ankles as well as on the torso at lumbar vertebra level 5 and chest (superior sternum level).[39] These inertial sensors record three-dimensional linear acceleration and angular velocities. Participants will also be asked to stand quietly with their hands on hips and their eyes open and closed for 30 s each with the sensors on to measure postural sway. These body worn sensors provide robust and quantitative measurements of multiple gait parameters that are reliable in pwMS[39] and are more sensitive than the T25FW for gait dysfunction in early MS.[40] These metrics also correlate strongly with fall frequency[41] and were successfully employed in our group's previous clinical trial of lipoic acid.[42]

Of the aforementioned mobility outcomes (ie, 6MTW, TUG, T25FW, MSFC), the one with the strongest correlation with SSEP latency in part 1 of the study will become the primary clinical outcome to measure remyelination in part 2 of this clinical trial.

Secondary outcomes

Aerobic fitness and biometrics

Aerobic fitness will be measured by VO_2max at baseline and after the 24-week active or control intervention using an electrocardiogram-monitored, maximal, graded exercise test on a cycle ergometer (Ergoline Via Sprint 150P) and an open-circuit spirometry system (Medgraphics Ultima CardiO2). The initial work rate will be 25 W, increasing by 25 W every 3 min until volitional fatigue and will be followed by a cool-down period, consistent with American College of Sports Medicine guidelines for VO_2max testing for pwMS.[43] HR and Rated Perceived Exertion (RPE) will be obtained every 3 min. VO_2max HR will be determined when two of four criteria are met: (1) VO_2 plateaus with increasing work rate; (2) respiratory exchange ratio ≥1.10; (3) peak HR is within 10 beats/min of age-predicted maximum (ie, ~1 SD); or (4) peak RPE>7 (0–10 scale).[44] VO_2max will be used to both to determine the target HR for MSCYCLE participants and to assess the change in aerobic fitness from before to after the interventions.

Body composition will be measured by 7-site skinfold testing using a validated algorithm (Harpenden Skinfold Caliper, FG1056; BodyTracker Software) at baseline and 24 weeks. Strength will be measured at baseline, 12 and 24 weeks; strength of the quadriceps and hamstrings will be measured by a hand-held dynamometer (microFET, Hogan Health, Ogden, Utah, USA). Hand grip strength will also be measured (Jamar Hydraulic Hand Dynamometer).

Cognitive function

Cognitive function will be measured at baseline, 12 and 24 weeks using the SDMT, a valid and reliable measure of processing speed and attention in pwMS.[45] During this test, participants are given 90 s to match symbols with numbers and the examiner records the total number correct.

PROs

We will measure several PROs at baseline, 12 and 24 weeks to determine if relevant MS symptoms are modified by the interventions. To measure fatigue, we will use the Modified Fatigue Impact Scale,[46] a multidimensional questionnaire of fatigue developed for pwMS. To assess pain, we will use the Pain Effects Scale,[47] a validated assessment of the ways in which pain impacts pwMS. To measure physical activity levels, we will use the International Physical Activity Questionnaire Short Form, a validated measurement of physical activity and intensity in adults.[48] To measure health-related quality of life (QOL), we will use computerised adaptive test versions of Neuro-QOL questionnaires of Upper Extremity Function, Lower Extremity Function, Depression, Sleep Disturbance and the Patient-Reported Outcomes Measurement (PROMIS) Self-Efficacy questionnaire. The PROMIS and Neuro-QOL tools were developed by the National Institutes of Health to assess QOL within and across various general and neurologic conditions.[49]

Brain imaging

A subset of 22 participants (11 active and 11 control group) will undergo an optional MRI at baseline and 24 weeks as an exploratory outcome measure of structural remyelination of the brain. Whole brain MPRAGE images will be acquired on a 7T MAGNETOM system (Siemens, Erlangen, Germany) using a 24-channel array RF coil (Nova Medical, Wilmington, Massachusetts, USA) for

co-registration and planning purposes (TI/TE/TR/α/data matrix/res=1.05 s/3.1 ms/2.2 s/7°/320×320×240/0.7 mm isotropic). A highly sampled inversion recovery (IR) technique—progressively unsaturated relaxation during perturbed recovery from inversion sequence with 32 inversion time points will be implemented to acquire 5 mm thick slices (TI/TE/TR/α/data matrix/res=0.017 8.0 s/7 ms/14.0 s/5°/128×128; (2 mm in-plane)). We will segment white matter structures (the corpus callosum and bilateral internal capsules) and MS lesions superior to the tentorium cerebelli. A multiexponential fitting procedure will be used on a voxel-wise basis to decompose IR recovery curves into short and long T components. The short T_1 component will be assigned to myelin associated water, and its amplitude will be used to calculate MWF. Tissue water content will be quantified and used to correct MWF. No contrast agent will be administered.

Blood sample biobanking

We will obtain 30 cc blood samples at four time points: the initial visit, the VO_2max visit before starting the clinical trial, and 12 and 24 weeks. We are not aware of any accepted remyelination biomarkers in the blood, at present. Serum and platelet-free plasma samples will be stored for analysis of any future remyelination biomarkers.

Other outcomes

Patient-Determined Disease Steps (PDDS) will be assessed at baseline to describe the sample. The PDDS is a PRO of disability in MS and has nine ordinal levels ranging between 0 (normal) and 8 (bedridden).[50] Medical and MS history will be obtained at each study visit and the PAR-Q will be performed at baseline and 12 weeks to evaluate for any interval relapses and assure that the participant is still safe to engage in aerobic exercise.[26]

Data management and analysis plan

The original hard copies of study data will be stored in a locked office and de-identified data will be saved on a secure electronic database. De-identified data will be added to an institutional repository after study completion. Data analysis will be overseen by a biostatistician. To determine the relationship between SSEP and measures of clinical disability and mobility, we will perform partial correlations between SSEP P40 and 6MTW, T25FW, MSFC, TUG, postural sway and other body-worn sensor measurements, fall frequency and lower extremity Neuro-QOL at baseline. We will also perform partial correlations to examine associations between SSEP N20 and upper extremity dexterity (9HPT) and upper extremity Neuro-QOL. In performing partial correlations, we will control for muscle strength, relevant demographic variables and disease factors, as needed. As an exploratory outcome, we will also perform partial correlations to examine associations between body structures (VO_2max, quadricep and hamstring muscle strength, hand grip) and the mobility measures listed above. These analyses will include all participants who provide baseline data.

To determine the safety and feasibility of the cycling intervention, we will track retention and adherence. Adherence will be measured as the number of sessions completed, with successful completion defined as participation in ≥80% of MSCYCLE or MSTC sessions. For MSCYCLE participants, we will perform a subanalysis to examine correlations between average time spent in the target HR zone and improvement in SSEP outcomes and improvement in aerobic fitness as measured by VO_2max.

To determine whether the cycling intervention is associated with clinical and objective evidence of remyelination, we will perform mixed effect linear regression analyses to model change in clinical measures and SSEP N20 and P40 from baseline to 12 and 24 weeks in active (n=22) and control (n=22) participants. Regression models will include random effects for participants and for limbs within participants. We will also perform a subanalysis looking at only the worst leg. Determination of the primary clinical outcome for part 2 of the study will be made after analysis of baseline data (see *Mobility and Disability Outcomes*, above), and will be determined before model building begins. Associations between change in SSEP latency and changes in walking-related outcomes will be analysed by linear regression or analyses of correlation (eg, Pearson's correlation) on the change scores. To model change in MWF from baseline to 24 weeks, we will perform mixed effects linear regression analyses in active (n=11) and control (n=11) participants of the corpus callosum, bilateral internal capsules and MS lesions. For all analyses involving multiple testing, an appropriate method of controlling the family-wise error or false detection rate will be chosen prior to beginning the analyses.

Data monitoring

Safety monitoring will involve yearly review of adverse events, dropouts, issues or breaches of confidentiality. Participants will be encouraged to report any potential problems at any time to the research coordinator or other study staff. We will inquire about adverse events during the assessment visits and MSCYCLE sessions. Adverse events will be judged by the monitor as related, possibly related or unrelated to the study procedures and categorised as mild, moderate or severe. Severe adverse events, unanticipated problems and protocol deviations that are greater than minor will be reported to the OHSU IRB. The OHSU IRB determined that a data monitoring committee was not needed because the study is low risk.

Patient and public involvement

Patients were not involved in the development of the experimental design. Participants in the trial will be invited to provide feedback about their satisfaction with the study and this feedback will be considered in the development of future studies. Results of the trial will be summarised using lay language in an OHSU MS Center newsletter which will be sent to the study participants.

ETHICS AND DISSEMINATION

The proposed study will take place at OHSU in Portland, Oregon, USA, has been approved by the OHSU IRB (reference: 21045), and has been registered on ClinicalTrials.gov. All participants will provide informed consent prior to study commencement. The results of the study will be presented at scientific conferences and published in peer-reviewed journals, regardless of the findings.

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Patient and public involvement Patients and/or the public were not involved in the design, or conduct, or reporting, or dissemination plans of this research.

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