PROTOCOL

Background

1. Provide the scientific background, rationale and relevance of this project.

INSTRUCTIONS

- This should include a referenced systematic evidenced-based review when possible.
- If this study involves qualitative research explain the major constructs of your study.
- Do not state in this section what you plan to do in this study. This information should be entered later under "What will be done in this protocol?"
- Do not include the bibliography in this section.
- For studies submitted under the Expedited review criteria, this section need not be more than a few paragraphs.
- For those studies where data will be analyzed collaboratively by multiple sites doing a similar study for which there is no common protocol (Collaborative Site Analysis Study) include a description of the common scientific goals/ procedures/data points.
- If this is an update to current templates from Protocol Builder make sure the information throughout the protocol includes the most current information.

Answer/Response: Multiple sclerosis (MS) is a neuroinflammatory disease characterized by immune-mediated demyelination and axonal degeneration in the central nervous system (CNS). The combination of demyelination and the resultant slowing of reversible deterioration in gait and an increased degree of pyramidal weakness of the lower limbs in some MS patients. The phenomenon was attributed to Dr. M. J. McArdle who taught but had not published on the subject (O'Neill et al.). This phenomenon had not been studied further until recently (Savoldi et al.; Schilaty, Savoldi, Nasr, and Weinshenker; Schilaty, Savoldi, Nasr, Delgado, et al.). The PI has developed a clinical method to evaluate this phenomenon as a clinical test (McArdle sign) and has developed a device to quantitate this phenomenon, as described below. By correlating McArdle sign with measures of central motor conduction determined by motor evoked potential studies, the PI recently suggested that McArdle sign results from slowed conduction resulting from the mechanical stretch of intact demyelinated axons (Hoffman et al.).

A preliminary blinded study conducted at Mayo Clinic, led by the P.I., showed that the difference in finger extensor strength with neck flexion versus neck extension was greater in patients with MS compared to both normal controls and patients with alternative causes of cervical myelopathy. This study utilized a first generation device, similar to the one that will be used in the current study, that measured the difference between the peak torque strength with maximum contraction of finger extensor against both a fixed resistance and against a constant resistance forcing the patients to subtend an arc of approximately 60 degrees.

This study will be conducted with a similar 2nd generation device designed to improve reliability and accuracy and potentially to increase sensitivity compared to the 1st generation device used in the published study. The device is a torque cell measurement device built at Mayo Clinic by an engineering group there that conducted a safety analysis, which has been submitted to Clinical Engineering at UVA and is approved (see the UVA clinical engineering website for listing of the device as approved). We have submitted this safety assessment for IRB review. There is no plan to commercially develop this device. It is noninvasive, carries minimal risk and discomfort. Briefly the device consists of a restraint for the patients' forearm consisting of a plastic molded base and Velcro straps that encircle the patients forearm and hand. The fingers are extended against a bar covered with foam, that can displace the fingers of the patient downward through an arc of approximately 60 degrees. The key measurement devices are a torque cell that records the strength and displays it on a computer interface and records the data, and a gyroscope worn on the patient's head as a "crown" that is kept in place by a band that is tightened by a screw to fit comfortable on the patient's head that records head position in two planes.. The innovation in the new device compared to the previous first-generation device that we used in previous studies is that the new device incorporates a motor to generate a constant resistance against which patients forcibly extend their fingers. This study will be preparatory for future research by defining the optimal settings to detect McArdle sign in patients with MS stratified by degree of background weakness and clinically detected degree of McArdle sign (stratified as 0-3+).

Objectives/Hypothesis

INSTRUCTIONS:
If this study involves biomedical research clearly state the objectives and hypotheses and clearly define the primary and any secondary outcome measures. If this study involves qualitative research clearly state your research hypothesis or question.

This section should not include information already included in other sections such as background information or information from the procedures section.

Answer/Response: The specific hypothesis for this preparatory study is that McArdle sign varies based on baseline weakness and strength. The objective is to enhance the sensitivity to detect McArdle sign's and to individualize the settings of the detection device stratified to baseline strength and clinically detected degree of McArdle sign (stratified as 0-3+) in patients with MS. This protocol is preparatory for future research that will study patients with cervical myelopathy associated with different causes and different types of inflammatory demyelination with different pathogenesis (e.g. neuromyelitis optica, which is an astrocytopathy rather than an a myelin-targeted disorder). In the future, we may perform additional studies evaluating the pathophysiology of McArdle sign and its utility in predicting response to drug treatment, such as dalfampridine. The current study will enroll only patients with MS and will not include any

correlative studies with treatment response. We do not plan to publish the data from this study directly, but hope to retain the data from this study and combine with additional data to be collected in future studies to be submitted to the IRB to further the general goals of the program: to understand the phenomenon of McArdle sign and its application to diagnosis, predicting response to treatment, serving as outcome measure in patients with MS.

Study Design: Biomedical

1. Will controls be used?

Answer/Response: No

►IF YES, explain the kind of controls to be used.

Answer/Response:

2. What is the study design?

Example: case series, case control study, cohort study, randomized control study, single-blind, double-blind, met-analysis, systematic reviews, other. You may also view the IRB-HSR Learning Shot on this topic to help you answer this question.
Writing a Clinical Research Protocol

Answer/Response: Case series, single-blind

3. Does the study involve a placebo?

Answer/Response: No

►IF YES, provide a justification for the use of a placebo

Answer/Response:

Human Participants

Ages: _18-80___
Sex: _M and F___
Race: __No restriction__

Subjects- see below

INSTRUCTIONS: For question 1-4 below insert an exact #. Ranges or OPEN is not allowed. This # should be the maximum # you expect to need to enroll (i.e. sign consent) If you are only collecting specimens the number of participants should equate to the # of specimens you need. If you are collecting only data from a chart review the number should designate the number of subjects whose medical records you plan to review. Age/ Sex/Race criteria should designate the demographics of participants from whom you will obtain the specimen/data.

1. Provide target # of subjects (at all sites) needed to complete protocol.

INSTRUCTIONS: If this is NOT a database protocol, this number should be the same as the number of subjects needed to obtain statistically significant results.

Answer/Response: We estimated that the study will require 35 enrolled patients to have greater than an 85% power of detecting a difference of at least a 5-percentage point difference for both the "medium" and "high" speed device settings for the lever that displaces the fingers of patients against resistance as predictors in a mixed-effects model with patient as a random effect to account for repeated measures at an alpha of 0.025 to account for two comparisons (0.05/2). We assume that 5 percent difference in force between neck flexed and extended conditions, a conservative estimate based on our preliminary studies.

2. Describe expected rate of screen failure/ dropouts/withdrawals from all sites.

Answer/Response: Single testing session. Minimal risk, inconvenience. Although it is a single testing session, and withdrawal is not anticipated, we allow for up to 10% of patients to voluntarily withdraw prior to completion of the full testing session.

3. How many subjects will be enrolled at all sites?

INSTRUCTIONS: This number must be the same or higher than the # from question # 1 in order to account for the # of screen failures, dropouts, withdrawals described in question # 2.

Answer/Response: 35

4. How many subjects will sign a consent form under this UVA protocol?

INSTRUCTIONS: If the protocol does not have a consent form- the number listed here should reflect such things as the number of subjects from whom specimens will be obtained, the number of charts to be reviewed etc.

Answer/Response: 35

Inclusion/Exclusion Criteria

INSTRUCTIONS:

- The inclusion and exclusion criteria should be written in bullet format.
- *This item applicable if the study will require consent (verbal or written).* Unless there is a scientific reason for not recruiting a certain type of vulnerable population(e.g. not enrolling fetuses, neonates or children in a study regarding Alzheimer's) list the following vulnerable populations under either Inclusion or Exclusion criteria below: pregnant women, fetuses, neonates, children, prisoners, cognitively impaired, educational or economically disadvantage, non- English speaking subjects .
- If you will not enroll subjects who do not speak English because certain procedures cannot be carried out if the subject does not speak English (e.g. a survey is not validated in other languages) insert the following as an Inclusion Criteria: Willingness and ability to comply with scheduled visits and study procedures.
- If this is a collection of only retrospective* specimens or data, the inclusion criteria must include a start and stop date for when specimens/ data will be collected.

- The stop date must be prior to the version date of this protocol.
- *Retrospective: all specimens are in a lab at the time this protocol is approved by the IRB. All data exists in medical records or records from previous studies at the time this protocol is approved by the IRB.

1. List the criteria for inclusion

Answer/Response:

1. Diagnosis of inflammatory demyelinating disease of the central nervous system satisfying clinical criteria for multiple sclerosis (McDonald, 2017)
2. Willing and able to comply with study procedures and scheduled visits

2. List the criteria for exclusion

Answer/Response:

1. Hand pain or physical impairment that precludes effective resistance to forced finger flexion.
2. Inability to comprehend or follow simple directions that precludes reliable assessment in the study.

3. List any restrictions on use of other drugs or treatments.

INSTRUCTIONS: List only those drugs or treatments that are prohibited while on study, not those listed as an exclusion criteria.

Answer/Response: None

Statistical Considerations

1. Is stratification/randomization involved?

Answer/Response: Yes

►IF YES, describe the stratification/ randomization scheme.

INSTRUCTIONS:
The stratification factors and/or the randomization plan should be identified. If there is no randomization component or important patient characteristics that will be used in treatment allocation or data analysis, a statement to this effect should be included.

Stratification factors: These are pretreatment patient characteristics which could be balanced across treatment arms by design or may be used to determine starting dose or treatment allocation.

If randomization is going to be used, the details of the randomization plan should be described.

The description should include:
--the method and timing of randomization
--the type of randomization scheme that will be used in the study

--whether or not the randomization masked/blinded/if so, then to whom is it masked/blinded
--who has access to the randomization scheme

Answer/Response: We will randomize the order in which patient go through the device speed settings (low, medium, and high). Additionally, we will balance the number of possible order combinations (order matters).

►**IF YES, who will generate the randomization scheme?**

_____ Sponsor
__X__ UVA Statistician. Insert name Answer/Response: Chad M. Aldridge will conduct the randomization scheme for the order in which the device speed settings are applied to each enrolled participant.
_____ UVA Investigational Drug Service (IDS)
_____ Other*:* Specify Answer/Response:

2. What are the statistical considerations for the protocol?

The objectives section and the statistical section should correspond, and any objective for which analysis is unfeasible should be deleted. Also, the estimates and non-statistical assumptions of the statistical section should be supported by discussion in the background section.

The answer to this question should include:
--Study Design/Endpoints
--Recap of study objectives and endpoint definitions. An assessment of how study objectives will be assessed by identifying & defining which endpoints will be used to assess each component of the study objectives.
--The study design should include contingencies for early stopping, interim analyses, stratification factors (If applicable), and any characteristics to be incorporated in analyses.
--The power/precision of the study to address the major study endpoint(s), the assumptions involved in the determination of power/precision.
--If statistical hypothesis testing is included then specify the null and alternative hypotheses, the test statistic, and the type I and II error rates
--If precision of an estimate, then provide a definition for precision
--If other, then specify

Answer/Response: The goal is to determine whether clinical McArdle sign can be detected by the torque cell measurement device and the optimal settings on the device to detect McArdle sign in each stratified category.

3. Provide a justification for the sample size used in this protocol.

Include sample size calculations or statistical power estimation. If not applicable, please provide explanation.

Also include the anticipated accrual rate, the accrual goal for the study, including accrual goals by strata if appropriate, adjustments for drop-outs etc. and study duration.

Answer/Response: We estimated that the study will require 30 enrolled patients to have greater than an 85% power of detecting a difference of at least a 5-percentage point difference for both the "medium" and "high" speed device settings as predictors in a mixed-effects model with patient as a random effect to account for repeated measures at an alpha of 0.025 to account for two comparisons (0.05/2). We assume that the percent difference in force between neck flexed and extended conditions is 5 percentage points. This also factors in the different prevalence of McArdle sign severity grades from 0 to 3. We utilized prior published data regarding the mean and standard deviation of the percent difference in force from Savoldi et al. (2019)

4. What is your plan for primary variable analysis?

Include primary outcome(s)/predictor variable(s), statistical methods/models/tests to be employed, or descriptive summaries as appropriate. If not applicable, please provide explanation.

Answer/Response: The primary outcome is the percent difference in the average peak force recorded by the device between neck flexed and neck extended conditions. We will utilize a mixed-effects model to predict the percent difference (outcome variable) using indicator variables for medium and high isoinertial speed settings of the device with participant ID as a random effect to account for repeated measures within subjects. This approach allows us to determine if knowing the isoinertial speed setting of the device is better than the null model. The null model is the outcome variable predicted solely be the intercept and participant ID as a random effect, this reflects the null hypothesis that Low = Medium = High isoinertial speed settings are equivalent in their influence on percent difference values. We will utilize additional model statistics including the AIC, BIC, LRT, and Baye's factor when comparing the null model versus our full model to validate that the full model is preferred over the null model.

Descriptive statistics will be done for the enrolled cohort of participants utilizing standard parametric and non-parametric approaches as appropriate to the underlying distributions of the descriptive variables and data types.

5. What is your plan for secondary variable analysis?

Include the following:
--Secondary outcome(s)/predictor variables, statistical methods/models/tests to be employed, or descriptive summaries as appropriate. If not applicable, please provide explanation.
--For phase III studies, the power/precision of the study to address the secondary objective(s).

Answer/Response: To replicate the association between percent difference values and clinical McArdle sign grades (0=absent; 1=present, but minor difference in strength with neck flexion; 2= present, moderate difference; 3= present, prominent difference), we plan to perform ROC curve analyses using the percent difference values as predictors of McArdle sign: (1) 0 versus 1-3, (2) 0-1 versus 2-3, (3) 0-2 versus 3.

6. Have you been working with a statistician in designing this protocol?

Consultation with a professional statistician is highly recommended to ensure good science of the study and facilitate the review process.

Answer/Response: Yes

IF YES, what is their name?
Answer/Response: Chad M. Aldridge

7. Will data from multiple sites be combined during analysis?
Answer/Response: No

INSTRUCTIONS: IF YES, answer the following questions

7(a). Does the study involve randomization?
Answer/Response:

IF YES, will randomization be done at each site or among sites?
Answer/Response:

7(b). Has the sample size calculation considered the variation among sites?
Answer/Response:

7(c). When combining the data from multiple sites to assess the study results, is the effect of the treatment to be tested (or the association to be tested) assumed to be the same across sites or vary among sites? What is the modelling strategy?
Answer/Response:

7(d). Is there a common protocol used in all sites?
Answer/Response:

IF NO, how will differences among sites, such as those related to the implementation, inclusion criteria, patient characteristics, or other sites characteristics, be considered to assess the study results?
Answer/Response:

Study Procedures-Biomedical Research

1. What will be done in this protocol?

INSTRUCTIONS:
This should include everything that will be done as part of this protocol. Do not repeat information that is included in other sections such as Background or Hypothesis sections.
This section should include an indication of which research interventions if any offer a prospect for direct benefit and which interventions (invasive measurements, collection of blood, tissue, data, surveys, etc.) are being done solely to answer a research question and generate generalizable knowledge. If the interventions done solely for research purposes are associated with greater than minimal risk they need to be justified. Describe and justify any control and experimental arm and include method, dose, and duration of drug administration. Reference any claim of clinical equipoise if applicable.

If you are obtaining specimens or data, provide information regarding the type of specimen/data, amount of specimen needed and how the specimen/data will be obtained and what analysis will be done with the specimen/data.

Special note for studies with waiver of consent/waiver of documentation of consent:
Include a statement regarding how subjects will be recruited. For other studies this information is captured in Recruitment does not need to be duplicated in this section.

Answer/Response:

1. Informed consent
2. Demographic information including age, sex, race, and ethnicity
3. Review of the medical record and/or questioning the patient to obtain the following information:
    a. Multiple sclerosis characteristics (onset, duration, key manifestations, disability scales, such as Expanded Disability Status Scale of Kurtzke and functional scores, based on neurological examination; findings on MRI of the head and cervical spine from information available on existing medical records)
    b. Multiple sclerosis treatment (particularly whether receiving dalfampridine and an MS disease modifying treatment)
    c. Co-morbidities
4. Clinical examination of finger extensor strength with neck voluntarily extended and flexed)
5. Testing of bilateral finger extension strength for approximately 5 seconds with neck extended and flexed alternately with approximately 40 seconds between trials using a torque measurement device; head position will be monitored with a gyroscope that fits as a band around the patient's head

2. If this protocol involves study treatment, explain how a subject will be transitioned from study treatment when they have completed their participation in the study.

Example: If the subject will be taking an investigational drug, will they need to be put back on an approved drug when they have completed the study? If yes, explain how this will be accomplished and who will cover the cost. If the subject has a device implanted will it be removed? Again- who will cover the cost of the removal?
Instructions: Answer NA if this study does not involve a study treatment.

Answer/Response: NA

Bibliography

INSTRUCTIONS: Provide a current bibliography supporting the hypothesis, background and methodology including references to papers and abstracts that have resulted from previous work by the investigator and references to the work of others.

Hoffman, E. Matthew, et al. "McArdle Sign and Neck Flexion-Induced Change in Central Motor Conduction in Multiple Sclerosis." *Multiple Sclerosis Journal*, Nov. 2024, p. 13524585241295868. *SAGE Journals*, https://doi.org/10.1177/13524585241295868.

O'Neill, J. H., et al. "McArdle's Sign in Multiple Sclerosis." *Journal of Neurology, Neurosurgery, and Psychiatry*, vol. 50, no. 12, Dec. 1987, pp. 1691–93.

Savoldi, Filippo, et al. "McArdle Sign: A Specific Sign of Multiple Sclerosis." *Mayo Clinic Proceedings*, vol. 94, no. 8, Aug. 2019, pp. 1427–35. *www.mayoclinicproceedings.org*, https://doi.org/10.1016/j.mayocp.2019.01.047.

Schilaty, Nathan D., Filippo Savoldi, Zahra Nasr, Adriana M. Delgado, et al. "Biomechanical Muscle Stiffness Measures of Extensor Digitorum Explain Potential Mechanism of McArdle Sign." *Clinical Biomechanics (Bristol, Avon)*, vol. 82, Feb. 2021, p. 105277. *PubMed*, https://doi.org/10.1016/j.clinbiomech.2021.105277.

Schilaty, Nathan D., Filippo Savoldi, Zahra Nasr, and Brian G. Weinshenker. “Neuromotor Control Associates with Muscle Weakness Observed with McArdle Sign of Multiple Sclerosis.” *Annals of Clinical and Translational Neurology*, vol. 9, no. 4, Apr. 2022, pp. 515–28. *PubMed*, https://doi.org/10.1002/acn3.51526.