**Protocol Title**: Registry and Natural History Study for Early Onset Hereditary Spastic Paraplegia

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**Protocol Synopsis:**

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| --- | --- |
| **Protocol Number** |  |
| **Protocol Title** | Registry and Natural History Study for Early Onset Hereditary Spastic Paraplegia |
| **Study Chair** | Darius Ebrahimi-Fakhari M.D., Ph.D. |
| **Institution** | Boston Children’s Hospital |
| **Study Design** | Longitudinal natural history study |
| **Primary Study Objective** | To systematically document the clinical presentation and natural history of AP-4-associated Hereditary Spastic Paraplegia (AP-4-HSP) and related early onset forms of HSP |
| **Secondary Study Objective** | To facilitate early diagnosis, enable counseling and anticipatory guidance to families and help define clinically meaningful endpoints for future interventional trials |
| **Study Population and Main Eligibility/Exclusion Criteria** | The study population consists of male or female patients of all ages with (1) onset of hereditary spastic paraplegia symptoms before the age of 18 years and/or (2) the presence of variants in HSP related genes and/or a relative of a person with such a diagnosis. |

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# **INTRODUCTION**

The hereditary spastic paraplegias (HSP) are a group of more than 80 neurodegenerative diseases that lead to progressive neurological decline. Collectively, the HSPs present the most common cause of inherited spasticity and associated disability. Bi-allelic loss-of-function variants in genes that encode subunits of the adaptor protein complex 4 (AP-4) lead to prototypical yet poorly understood forms of complex HSP in children, called AP-4-associated HSP (or AP-4-HSP). This includes four different conditions: SPG47, SPG50, SPG51, and SPG52. The molecular mechanism in all four conditions is a loss-of-function of the AP-4; hence they are thought to share a similar clinical phenotype. Most of the cases reported so far were diagnosed in the last three years, indicating that AP-4-HSP is probably an under recognized condition. Similarly, HSPs on a broader scale have been unrecognized until recently, including but not limited to SPG3, and SPG4, SPG15, which share common phenotypes with AP4-HSP but vary in disease progression and severity. The age at symptom onset is often in infancy, however significant disease progression occurs between the ages of 5-18 years. The core set of signs and symptoms of AP4-HSP are: early-onset developmental delay with delayed motor milestones and significant speech delay (~50% non-verbal); intellectual disability in the moderate to severe range; mild hypotonia in infancy followed by spastic diplegia and later tetraplegia; postnatal microcephaly (~80%); foot deformities (~70%); and epilepsy (~70%) that is intractable in a subset of patients. Episodes of stereotypic laughing, possibly consistent with a pseudobulbar affect, were found in ~60% of patients. Key features found on neuroimaging include a thin corpus callosum (90%), ventriculomegaly (65%) often with colpocephaly, and periventricular white-matter signal abnormalities (~70%). Iron deposition and polymicrogyria were also found in a subset of patients. About two-thirds of patients are born to consanguineous parents. Published reports consist of small case series only and there has been no effort to systemically delineate the spectrum of the disease or its progression. We aim to delineate the core clinical, imaging, and molecular features of AP-4-HSP and related HSP disorders across the age spectrum. This registry and natural history study will facilitate an early diagnosis, enables counseling and anticipatory guidance of affected families and will help define clinically meaningful endpoints for future interventional trials. Samples will be collected for the purpose of molecular and cellular investigation that will help identify biomarkers and novel targets for therapy. The samples and clinical information will be housed in the Translational Neuroscience Center and an AP-4-HSP REDcap database, respectively; both located in Boston Children’s Hospital (BCH), but will be available to investigators around the world after approval.

**SPECIFIC AIMS / OBJECTIVES**

The objectives of this protocol are to (1) To systematically document the clinical presentation and natural history of AP-4-associated HSP and related early-onset forms of HSP and (2) To facilitate early diagnosis, enable counseling and anticipatory guidance of affected families and help define clinically meaningful endpoints for future interventional traits.

Specifically, the aims are to:

1. Create a registry to perform an initial cross sectional analysis of clinical, imaging and molecular data to establish the disease spectrum.
2. Create a repository of biological samples and collection of longitudinal clinical data that helps establish the natural history of AP-4-HSP and related forms of HSP
3. Create a registry that allows for re- identification and re-contact of participants by appropriate investigators

**BACKGROUND AND SIGNIFICANCE**

Adaptor protein complex-4 (AP-4) belongs to a family of adaptor proteins (AP-1 through AP-5) which are evolutionarily conserved heterotetrameric protein complexes. AP-4 is composed of four subunits (β4, ε, μ4, σ4) that form an obligate complex. These subunits are encoded by the following 4 genes: *AP-4B1*, *AP-4E1*, *AP-4M1*, *and AP-4S1*. A loss of function for any one AP4 subunit makes the entire AP-4 complex nonfunctional and results in this disorder. While the molecular pathology of AP-4 deficiency remains largely unknown, AP-4-HSP has emerged as an increasingly recognized form of complex HSP which is associated with a thin corpus callosum. More broadly, early-onset HSPs display a shared set of core symptoms but vary in severity and progression of disease depending on the gene affected. Given the lack of a systematic review of the natural history of this disorder, we aim to develop a registry and natural history study in an attempt to better characterize the onset, signs/symptoms and progression of AP-4-HSP and related form of HSP.

**DESIGN AND METHODS**

**STUDY DESIGN**

The Registry and Natural History Study for Early Onset Hereditary Spastic Paraplegia (HSP) is focused on gathering longitudinal clinical data as well as biological samples (skin and/or blood and/or saliva). We are planning to recruit male or female patients of all ages with (1) onset of hereditary spastic paraplegia symptoms before the age of 18 years and/or (2) the presence of variants in HSP related genes and/or a relative of a person with such a diagnosis. After ascertainment of written and verbal consent, the following are required by all enrolled participants:

1. We may ask the designated referring physician to complete the HSP Natural History Study Questionnaire
2. We may ask the participant and/or study family to complete the CPCHILD Questionnaire
3. Completion of medical records release form to provide consent to review medical records both at BCH and outside institutions including, but not limited to clinic notes, test results, copies of MRI scans already obtained for clinical purpose, medications, pathology reports, and other relevant clinical data
4. Sample of peripheral blood by standard venipuncture methods and/or provide access to previously collected blood and/or DNA that is no longer required for clinical purposes or saliva sample collected by free spitting into a provided saliva collection kit
5. Previously collected tissue specimens that are no longer required for clinical purposes and/or provide a small additional sample specifically for research purposes, i.e. while undergoing another clinically indicated procedure that posed no more than minimal additional risk to the participant
6. Every 6-12 months, we may ask for a follow-up HSP Natural History Study Questionnaire, CPCHILD questionnaire and GMFCS questionnaire

Once these data and/or specimens are collected, the study staff will continue to have longitudinal contact with study participants (only with subjects agreement) to obtain clinical updates, repeat collection of questionnaires and/or collect additional samples.

**PARTICIPANT SELECTION AND INCLUSION/EXCLUSION CRITERIA**

**i. Participant Selection**

The samples used in this study will be gathered from individuals with a known diagnosis of spastic hereditary paraplegia and/or an AP-4-HSP related gene mutation, and/or their family members of interest (if applicable). We plan to enroll at least 100 families (approximately 200 participants) per year, and do not have a projected end-date to this project.

**ii. Inclusion Criteria**

1) Male or female patients of all ages with (1) onset of hereditary spastic paraplegia symptoms before the age of 18 years and/or (2) the presence of variants in HSP related genes and/or a relative of a person with such a diagnosis.

**iii. Exclusion Criteria**

1) Not having such a diagnosis and/or not being related to such individual.

**DEFINITION OF PRIMARY AND SECONDARY OUTCOMES/ENDPOINTS**

## The primary endpoint of this longitudinal, natural history study will be the creation and implementation of a research repository/databank of molecular, biochemical, and phenotypic information and samples that are functional and accessible for investigators with approval to access the databank.

**RECRUITMENT/DATA COLLECTION METHODS, ASSESSMENTS AND SCHEDULE**

**c. Recruitment Methods**

i. HOW, WHERE and WHEN will potential subjects be recruited?

Recruitment process will be overseen by the PI, sub-Investigator and research coordinator at BCH**.**

There are 6 ways by which participants are recruited.

1. Participants will be recruited in genetics or neurology clinic, or any other clinic/department at BCH which cares for individuals with HSP or AP-4-HSP. Clinicians at BCH may request that we recruit participants under their care after they determine the participant’s eligibility and obtain participant permission to forward their contact information to the study team. Alternatively, BCH clinicians may provide individuals under their care with the research coordinator's information for the family to contact directly. The research coordinator will then handle the enrollment process.
2. Physicians and collaborators off-site who learn of the research may request that we recruit participants under their care after they determine their participant’s eligibility and obtain participant permission to forward their contact information to the research coordinator. Alternatively, off-site clinicians may provide individuals under their care with the research coordinator's information for the family to contact directly. The BCH research coordinator will handle the enrollment and informed consent process. In certain circumstances (i.e. patient is deceased or lost to follow up), if a clinician is aware of an eligible patient however the study staff cannot obtain written consent, the study staff will request a waiver of documentation of consent to obtain only de-identified data (HSP Natural History Study Questionnaire without PHI and de-identified MRI scans).
3. Some participants may be self-referred, after becoming aware of our research through our website, other websites such as CureAP4.org, other participants, or other materials (website, flyers or advertisements). These participants can contact the BCH research coordinator or another member of the research team via telephone or email to learn more about the research.
4. Study staff may use databases (Decipher, GeneMatcher and other publicly available databases) to contact off-site physicians providing care to patients with known HSP gene mutations that were entered into those databases. The rest of the process will follow #2 on this list.
5. We will also send an IRB-approved research advertisement and letter to genetic testing laboratories, local geneticists, neurologists, genetic counselors, and behavioral/developmental pediatricians who may have patients with HSP, and invite them to refer patients to this research study.
6. We will not be actively approaching patients or contacting clinicians from EU countries regarding enrollment into the registry. However, if families or clinicians contact the study staff to participate, they can be recruited into the study by carefully following GDPR regulations.

For all six recruitment methods, eligible participants who want to participate may be recruited via the telephone, email, and/or mail. Participants are always provided with the study brochure or letter and are given opportunities to consider their options. If the eligible participant chooses to enroll, the research coordinator will arrange to meet the family/patient at their next BCH visit or at their convenience. Alternatively, enrollment can take place over the phone/via email or mail so no trip to BCH is necessary.

ii. WHAT recruitment methods and materials (e.g. posters, fliers) will be used? *- attach all materials*

**1) Recruitment of participants through the neurology, genetics clinic, and other BCH departments**

Potential participants who will be seen at BCH will be identified by their clinicians at the time of their clinic appointment. At the end of their visit, the clinician may tell the participant/family about the study. If they are interested in participating, the study coordinator can be paged to explain the study and answer any questions. The participant will be provided with HSP Registry and Natural History Study consent form and will be given time to ask questions and consider their option of participating. If the participant chooses to enroll, the research/study coordinator will consent the individual and any present family members of interest (if applicable) at this time. Blood/saliva and/or skin collection can also take place at this time, as explained below.

Patients may also be recruited in the Boston Children's Hospital neurology, genetics clinic or other clinics which have expressed interest in recruitment to our study. Patient eligibility will be determined by the research coordinator or relevant clinician. This may include a review of the patient’s chart. Eligible patients are approached by either their clinician or the genetic counselor to determine if they are interested in learning about our research project. If they are interested in participating, the research coordinator will explain the study in detail and obtain written or electronic consent/assent to participate. An electronic version of the consent form for patient enrollment may be offered by the study team at the time of enrollment. All electronic consents will be documented by obtaining an electronic signature.

If preferred, the consenting and enrollment process can also be conducted via telephone and mail. Physicians can provide the study coordinator a possible participant’s name and MRN for the study coordinator to contact and assess interest in participation. To establish a potential participant’s interest in enrolling, a Letter to Assess Interest (with opt out section) will be mailed to them. If the family wishes to opt out, the enrollment process will stop, and the individual’s choice not to participate will be documented in our recruitment database so no further contact will be made. If the family does not respond within 2 weeks, the study coordinator will contact them again to assess for interest.

After establishing the potential participant’s interest in enrolling, two copies of the consent form (one for the participant’s records, and one to be signed and returned) along with the Letter to Assess Interest (with opt out section) will be sent to the potential participant. A telephone call will be scheduled to review the consent forms with the participant and answer any questions. If the child is capable of providing assent, the research coordinator will also speak with child/adolescent directly to explain the study, and ask questions as to gauge comprehension. This must be done in addition to obtaining parental permission from child/adolescent’s parent/guardian. To document assent, ensure the parent/guardian signs and dates the correct line (specifying relationship to child) and that the child/adolescent signs and dates the subject/participant line.

Once the signed consent forms are received by the study staff, and the study participant is willing to provide samples, a sample collection kit with instructions, and mailing supplies will be mailed to the participant. If needed, the research coordinator can help potential participants coordinate the sample collection with their local clinician’s office or at a clinically certified facility.

**2) Recruitment of participants through outside clinics**

If a health care provider or other personnel contacts us through an external clinic to refer a family for the study, we require that they first contact the family about our study to obtain permission from the family to fax or mail the contact information on to us. We will then contact the family as preferred (mail, phone, email) and after interest in confirmed, recruitment/enrollment procedures will then proceed as detailed above.

**3) Self-referred patients who contact the study team about the registry and natural history study directly**

Recruitment within and outside of BCH will involve the use of our website (<http://www.childrenshospital.org/conditions-and-treatments/conditions/s/spastic-paraplegia-47>**),**  CureAP4.org and similar foundations, flyers/brochures placed in the waiting areas and clinic rooms of various BCH departments and collaborators’ sites as well as advertisements in internal/external newsletters and medical magazines targeting clinicians or patient populations of interest. Potential participants may then contact the study staff directly or through their clinician to obtain further information about the study. Study recruitment and participation would then proceed as outlined above.

**4) Databases to contact off-site physicians**

Study staff may reach out to databases (Decipher, GeneMatcher and other publicly available databases) to provide IRB-approved research advertisement and letter to off-site physicians providing care to patients in those databases with known HSP gene mutations. The off-site physician may determine their patient’s eligibility and obtain participant permission to forward their contact information to the research coordinator. The rest of the enrollment procedure follows as described in section 1 (Recruitment of participants through the neurology, genetics clinic, and other BCH departments).

**5) IRB-approved research advertisement and letter**

Study staff will send an IRB-approved research advertisement and letter to genetic testing laboratories, local geneticists, neurologists, genetic counselors, and behavioral/developmental pediatricians who may have patients with HSP, and invite them to refer eligible patients to this research study.

**Exemptions to recruitment into study**

In rare circumstances where the study staff is unable to obtain written consent from the study participants, a waiver of consent will be provided along with the HSP Natural History Questionnaire, which does not ask for protected health information. In these cases, the study staff will not ask for any biological samples from these families however, will request the referring clinician to complete the HSP Natural History Questionnaire and provide de-identified MRI images.

**Recruitment of Additional Family Members**

If during the participation process, the study coordinator realizes there is a biological family member of interest, the research coordinator will ask the participant/family if they feel this family member might be interested in enrollment. If so, the research coordinator will obtain the contact information for the family member of interest and permission to the family member. Recruitment and participation would then proceed as outlined above. Of note, at no time would an additional family member be approached by our study staff without the family’s permission.

iii. WHO will be responsible for subject recruitment?

Recruitment will be overseen by the study coordinator and investigators.

**STUDY ITEMS**:

Once consent has been obtained, we may collect the below items from our participants either in person or via the telephone/mail.

1. With the participant’s permission, study staff or the participants themselves can ask their healthcare provider to complete an HSP Natural History Study Questionnaire. Clinical features are assessed using a standardized questionnaire developed for the purpose of this registry. The questionnaire aims to gather health information to see how this disorder develops and changes over the course of the time. This includes information ascertained in clinical practice, including clinical rating scales such as the Spastic Paraplegia Rating Scale, the Modified Ashworth Scale and the SPATAX-EUROSPA Disability Scale and others.

2. Each family will complete the Caregiver Priorities and Child Health Index of Life with Disabilities (CPCHILD©) questionnaire that measures caregivers’ perspectives on the health status, comfort, well-being, and ease of caregiving of children with severe developmental disabilities.

3. Provide consent via a medical record release form to review medical records both at BCH and outside institutions including, but not limited to, clinic notes, test results, copies of MRI scans already obtained for clinical purpose, medications, pathology reports, and other relevant clinical data. These records will continue to be collected on at least a yearly basis to allow for longitudinal data collection.

4. Provide blood or saliva for DNA, RNA, or other biochemical molecules extraction from the participant and any participating biological family members of interest at initial enrollment. Additional samples may be requested on a 6-12 month basis to allow for longitudinal sample collection only if the participants have scheduled blood draws for a clinically-relevant reason. Longitudinal sample collection is necessary to explore and define biomarkers, such as markers that indicate disease progression over time. Every effort will be made to limit the number of blood draws and to combine the sample collection with blood draws for routine clinical care whenever possible.

5. Provide access to previously collected tissue specimens that are no longer required for clinical purposes and/or provide a small amount of additional skin sample specifically for research purposes (by research-only punch biopsy or if undergoing a future clinically-indicated procedure).

6. Every 6-12 months, the study staff will reach out to the family’s healthcare provider to complete a follow-up HSP Natural History Study Questionnaire. At the same time, the study staff will request the family to complete a CPCHILD questionnaire and a Gross Motor Function Classification System (GMFCS) Family Report Questionnaire.

**SAMPLE COLLECTION**

**Questionnaires**: Via the CPCHILD questionnaire, the participant and/or family members will be asked a series of questions that measure caregivers’ perspectives on the health status, comfort, well-being, and ease of caregiving of children with severe developmental disabilities. If the family is non-English speaking, we will provide them with a CPCHILD questionnaire in the family’s native language (if available). Every 6-12 months, in order to obtain longitudinal caregivers’ perspective, we will request the participant and/or family members to fill out the CPCHILD questionnaire. In addition to the CPCHILD questionnaire, a GMFCS Family Report Questionnaire will also need to be completed at those times.

In addition, the HSP Natural History Study questionnaire will be provided to the participant’s healthcare provider to complete. Healthcare providers will be able to use information ascertained in routine clinical practice, including clinical rating scales such as the Spastic Paraplegia Rating Scale, the Modified Ashworth Scale and the SPATAX-EUROSPA Disability Scale and others. To obtain longitudinal clinical data, we will request the clinicians to complete a follow-up questionnaire every 6-12 months. The follow-up questionnaire will include clinical rating scales such as the Spastic Paraplegia Rating Scale, Modified Ashworth Scale, Longitudinal Disability Score and the Clinical Global Impression Scale and others.

Study staff will request follow-up questionnaires and clinical data on at least a yearly basis to participants that are appropriately consented and expressed interest in re-contact.

**Medical/family history and medical record collection:** Copies of original medical records, including (but not limited to) clinic notes, biopsy reports, pathology slides, genetic test results and other diagnostic testing results (i.e. EMG, EKG, MRI scans, etc.) will be solicited from referring clinicians, pathologists, and other identified medical care providers. Data related to genetic testing (i.e. raw genomic sequencing data) may be requested from other labs which may have performed it on a clinical basis to the registry for additional analysis. Families will always be contacted first before such data is requested, and permission from the family will be documented in the research record. The study staff will request the data transfer from the lab, however if required by the transferring institution, the family may request the data transfer as well. In some cases, study subjects may provide copies of their own medical records. In case of data sharing with international collaborators through their local IRB-approved protocols, study staff will ensure that the local protocols/consents allow for sharing of data by either translating the consent forms or obtaining a letter from the local IRB that precisely states the data sharing policy. To obtain longitudinal clinical data, we will request medical records on at least a yearly basis.

**Phlebotomy:** For our study, the proband and any biological family members of interest (if applicable) will each provide a blood sample for the study. If a blood sample is obtained, four tubes of blood totaling up to 30 ccs will be drawn. Of note, given that we will be enrolling younger-aged children in this study, institutional guidelines will be followed with regard to the total amount of blood ultimately drawn as outlined by CCI Policy Document 081.022 which states the appropriate amount of blood to be obtained in regards to a participant’s size and age. For Children’s Hospital patients, this will be performed by the outpatient phlebotomy laboratory staff, ward staff (if inpatient) or other qualified medical personnel, and any additional costs will be charged to our research budget. Patients referred by outside collaborators will have blood drawn by those clinicians, their designees or at a certified clinical facility. For these participants, a blood draw kit and instructions will be sent to the participant’s home to be brought to the designated clinician’s office or certified clinical facility. If not covered by the collaborating clinician’s budget, the cost of phlebotomy and specimen mailing will be covered by our research funds. For either in house or outside blood collection, a form will be completed at the time of collection by the individual performing the phlebotomy. After the DNA is extracted, it will be stored in a secure -80°C freezer. Double-labeling protocols will be followed with all samples. To obtain longitudinal clinical data, we may ask for additional samples.

**Saliva collection:** If a saliva sample is collected, this can be performed before/after a clinic visit at BCH or at the participant’s home according to participant preference. A saliva collection kit will consist of two Oragene collection vials and/or swab sponge collection and deposited into the Oragene collection vial. A saliva collection kit, collection instructions, and sample collection form will be sent to the participant and returned to our laboratory with the cost of mailing paid for by our research funds. In the rare situation where a second saliva sample is required, participants may be re-contacted. To obtain longitudinal clinical data, we may ask for additional samples.

**Tissue collection:** A tissue sample will be obtained for DNA/RNA extraction and various analytical techniques/biochemical assays. Pre-existing or pending tissue biopsies will be requested directly from the relevant pathology departments, and copies of the signed Informed Consent forms will be provided to document the request. In many cases, insufficient amounts of tissue remain from such diagnostic biopsies. Therefore, participants may provide a new specimen specifically for research purposes if the participant is already undergoing a clinically indicated surgical procedure. These procedures will always have been recommended by the participant’s clinician and not a member of the research staff. Furthermore, removal of any tissue will only be performed when the requesting physician and the surgeon agree that it would represent no additional risk to the participant.

Additional tissue samples may be collected from:

1) Any tissue that is removed or discarded as part of the standard clinically indicated procedure

2) Cells/tissues collected prenatally for clinical reasons which would normally be discarded.

3) Skin Biopsy Collection

Skin biopsies up to 2 mm will be collected into tissue culture media. The procedure will be performed by a clinician with training to do so. In general, it is anticipated that most of the skin biopsy will be performed solely for research purposes. However, in most instances the study subjects will likely undergo sedation or anesthesia for any other procedures (such as a bone marrow aspirate and biopsy) efforts will be made to perform the skin biopsy at the same time to minimize pain and avoid additional anesthetics. In those cases we will perform the skin biopsy at the same insertion site as the bone marrow biopsy or at the site of a surgical incision to avoid any extra minimal scar from the skin biopsy. In rare instances where this is not possible the punch biopsy will be obtained from a site on the upper extremity to be determined by the individual performing the punch biopsy.

A 2mm punch biopsy will be obtained, which requires no sutures. The family will be asked if there is a history of excessive scar formation, and if so, the subject will be excluded. EMLA will be applied prior to the procedure, followed by lidocaine/epinephrine local anesthesia. Subjects will be informed that collection of skin biopsy samples poses the risks of pain and discomfort (similar to a blood draw), visible scarring at the biopsy site, and bleeding and infection at the biopsy site which will require keeping the area clean and covered with a Band-Aid for a longer period of time than a blood draw.

The skin biopsy may be performed as a bedside / office procedure under standard lidocaine/epinephrine local anesthesia or during the time of a clinically indicated procedure like surgery or a bone marrow biopsy. All skin biopsy sites will be dressed with sterile white petroleum jelly and a Band-Aid. Wound care consisting of daily dressing changes will be explained to the subject. If there are any concerns or if the subject prefers for any reason, he/she will return to Children’s Hospital to see the physician performing the biopsy at any time, and at 2 weeks for suture removal.

The consent has check boxes for the patient to indicate a willingness to provide a research specimen of skin while undergoing a clinically indicated surgical procedure that requires cutting of skin or as a separate out-patient procedure. These procedures will always have been recommended and will be performed at the request of the subject’s personal physician (i.e., not someone on the research study staff) and removal of additional skin tissue for research will only be done when both the requesting physician and the surgeon agree that it would represent no more that minimal additional risk to the subject. The study staff will have a conversation with the clinician performing the biopsy beforehand to discuss collection instructions and to remind the clinician to mention the sample’s procurement in any relevant procedure notes. Written instructions for this will also be provided. Collection of the biopsy samples poses potentially greater than minimal risks of pain and discomfort and of scarring at the biopsy site, and the minimal physical risks of bleeding and infection at the biopsy site, however we believe these risks are minimal in comparison to the risks associated with the clinically indicated procedures listed above. Study staff will contact the family approximately 2 days after the tissue sample collection to assess for any problems or concerns they may have with the collection site.

Wound healing, dressing, and care will not change for any of these tissue procurements.

For discarded prenatal samples, the cells/tissues will also be an invaluable resource to help us understand early fetal development without any additional risk to the fetus or mother. If the fetus is born, and the family is interested in enrolling the child into the study, we will obtain new consent for that child, using our previously approved consent.

All tissues will be procured by standard tissue handling protocol, assigned the appropriate study ID (“HSP ID”) and double labeled using a “Sample ID.” The HSP ID will remain a constant for each participant and the Sample ID will be generated by the lab for each of the participant’s samples. Both of these are not easily linked the participant’s personal information.

**STUDY TIMELINE**

We expect our studies to go on for an indefinite period of time. Analysis of collected specimens will continue for an indefinite period of time and participants will be considered “actively enrolled” unless they contact the study coordinator and ask to be withdrawn from the study.

**ADVERSE EVENT CRITERIA AND REPORTING PROCEDURES**

Adverse events are defined as any undesirable experience occurring to a subject during the study**.**

This protocol does not involve any medical therapies/interventions so this study is considered to be minimum risk. However, if there is any study related adverse events reported by the subject or observed by the investigator or his staff it will be recorded and reported to the IRB as per the current IRB reporting guidelines, and at the annual renewal for the protocol.

There is a very small risk of infection or continuous bleeding at the site of the venipuncture. There are also no known risks to saliva collection by spitting into the collection container or using a swab sponge. If an adverse event occurs, it will be reported to the IRB.

**DATA MANAGEMENT METHODS**

A paper hard copy or electronic copy of the questionnaires will be used. The collected hard copy data will be entered into a HSP REDCap database when applicable. In this way, mistakes while entering the data will be minimized. Each hard copy data form will have a field for the data and initial of the person who entered the data into the database in order to minimize the double entry of data.

Medical records will be evaluated by the investigators and research coordinator and entered into the database manually, through a database utilizing standard nomenclature. This allows for flexibility about what can be entered from the medical records, while also minimizing the occurrence of input variations for the same feature/phenotype. The database will then be searchable by these terms. Medical records may be requested from outside institutions as frequently as once a year or when a new feature or result is available.

Samples and correlating clinical and family history information collected under this protocol will be available for study staff with appropriate human research training directly and will be identifiable. They may also be available to other investigators in a de-identified fashion. Collaborations will either be established by the study staff or external requests for samples will be received via an application form and reviewed by the HSP Steering Committee. The study staff will confirm that the goals of the research are in line with the goals of this protocol and that the intended use is covered in the scope of this study’s consent form. When appropriate, a Material Transfer Agreement (MTA) will be completed by the receiving party. Investigators requesting de-identified information will not require an IRB protocol.

Investigators seeking select information that falls within a “limited data set” will sign the MTA as required under the HIPAA Privacy Rule. Limited datasets can include 1). Dates (e.g. admission date, birth date), geographic information excluding street address (e.g. city, zip code). Investigators requesting limited datasets will not need IRB approval.

Identifiable samples and clinical and family history collected under this protocol will be available to investigators upon request of the participant and investigators with appropriate IRB approval to receive identifiable specimens and information from this study. The PI/study coordinator will also confirm that the goals of the research are in line with the goals of this protocol and that the intended use is covered in the scope of the study’s consent form.

**BIOLOGICAL SAMPLE MANAGEMENT**

Blood or saliva will be collected for DNA extraction and will be labeled with an HSP ID and Sample ID. DNA will be extracted from the samples and stored at a Translational Neuroscience Center at BCH until such time that analysis is requested. Samples and data will be distributed de-identified. 2) Additional blood/saliva, and pre-existing tissue samples may be collected for further DNA analysis, RNA analysis, and biochemical assays. When these samples arrive, they will be assigned an “HSP ID” and double-labeled as with an “HSP ID” as described above. These samples will then be stored in the Translational Neuroscience Center -80°C freezer or liquid nitrogen tanks on the 14th floor of the Center for Life Sciences. The data for this study will be collected by a trained member of the study staff.

Once the individual’s enrollment information is entered, an “HSP ID” will be given to each study participant. None of identifying personal information of the subject will be used to make up the “AP-4-HSP ID” code for a specific participant. The link between the HSP ID and the participant's name will be kept in a separate HSP Study folder locked in the study coordinator’s office and on a password-protected section for the registry which is only accessible by the research coordinator, and appropriate members of the study staff. The key/passwords are only known to the principal investigator and an appropriate designee/research coordinator.

This information allows better understanding of a participant’s clinical symptoms and progression, helping us to establish appropriate genotype/phenotype correlations. Molecular and biochemical data will be collected by study staff and recorded in registry by the research coordinator or another study staff member. The database contains multiple sections of information, including general participant information, medical and family history, record of blood, saliva and tissue specimens received, results of diagnostic tests that are in progress or completed, and referring clinicians’ information. The database is double-password protected and only accessible by the PI, study coordinator, laboratory supervisor or other appropriate study staff members.

Searchable aspects of the database will not contain identifiable information and will only be accessible to physicians who create an “Investigator’s Profile” and are granted access by the research coordinator.The computer servers and database hardware will all be stored at BCH and data will be stored in a secure manner compliant with Institutional standards. All participant information is de-identified prior to being shared with non-staff members, except for referring clinicians, other individuals designated by the participant.

To prevent disclosure, information about participation, including research data collected and the results of the research, will not be placed in the medical record. Any documented link between an AP-4-HSP ID and a participant’s information will be kept locked in the study coordinator’s office and on a double password protected database. Only the study administrator and appropriate study members will have access to the link between patient ID and names. Data and samples will be coded without the use of identifiers for anyone browsing the databank. Only appropriate members of the registry and physicians allowed by the participant will have access to identifiable information. Participants undergoing genetic testing by a licensed clinical genetics laboratory may have blood or buccal samples sent with an accession number, name, MRN, and/or DOB. Remaining samples will return to BCH at the conclusion of the study.

3) De-identified data (including information about variants and associated phenotypes in aggregate, or individually) may be deposited into other repositories and shared databases such as dbGap, dbSNP, ClinVar and others to contribute to the national effort of improvement of interpretation of genomic information and understanding of human disease.

4) The study is designed to collect biological materials (blood/saliva and tissue) for the purposes of (1) analyzing DNA, RNA and protein expression to identify disease markers and disease mechanisms (2) establishing pluripotent stem cell lines by in vitro reprogramming methods that involve biological, chemical or genetic manipulation and selection in a variety of cell culture conditions that favor pluripotent stem cell derivation, and (3) further manipulation of all patient-derived cell lines to study the underlying pathophysiology and to identify new therapeutic targets . Collected cell lines will be used to further study the biological mechanisms using standard cell biological, biochemical and molecular biology techniques. Furthermore, cell lines are a resource for testing possible treatments, and can be used as a model for the participant's disorder. The induced pluripotent stem cell creation may occur in the Translational Neuroscience Core (TNC) at BCH, or other facilities which has extensive experience in the derivation and manipulation of human pluripotent stem cells.

**IDENTIFYING AND RE-CONTACTING PATIENTS**

Once an individual is entered into HSP registry, no identifiable information (name, birth date, MR#, etc.) will be seen while searching the database. Only de-identifiable information will be accessible to researchers. Only the study coordinator, study administrators under his/her supervision, or other appropriate study personnel listed on this protocol will have access to identifiable information.

Researchers interested in working on AP4 or other forms of HSPs and requesting data or samples may apply to the HSP Steering Committee for approval and access.

Study staff will strictly be re-contacting those patients that indicated on the consent forms that they would like to be re-contacted for future studies.

**REPORTING FINDINGS**

Individual results will not be provided back to participants and/or family members. Aggregate results will be provided in the form of a website posting, newsletter or publication. Results or study progress report will be provided upon request.

**QUALITY CONTROL METHOD**

The investigators and research coordinators will collect the clinical data and have experience in collecting medical and family history data. In addition, once a year, the phenotypic data collected will be reviewed for completeness by the investigators/research coordinators. If there is any missing data, it will be noted and then collected through either extraction from the hospital databases or via querying the participants directly.

Additionally, all clinical data and samples collected will be carefully labeled with the HSP ID so that the PI and his appropriate designees can establish to which participant the information relates as needed for this study.

**DATA ANALYSIS PLAN**

The goal of this project is to develop a repository of data/samples that will contain the genotype and phenotype information on enough individuals to allow for additional studies.

**STATISTICAL POWER AND SAMPLE CONSIDERATIONS**

The goal of this project is to develop a repository of data that will, over time, contain phenotype and genotype data on enough individuals to allow large scale studies. Since this is a registry, repository, and natural history study we do not have formal power calculations. Furthermore, since this is a study looking at early onsetHSP with small participant populations, we do not have formal power calculations.

# **PROTOCOL DEVIATIONS**

Any deviations from the protocol will be reported to the local IRB at time of continuing review. This information will also be captured on the protocol deviation CRF in the REDCap database. Any significant deviations will be reported within 24 hours to the local IRB.

**STUDY ORGANIZATION**

The majority of data gathering and analysis will take place at BCH. In some situations, de-identified data, samples, or results will be shared with our scientific collaborators. Since we are studying rare conditions, pooling resources is often essential to identifying the genes involved or gaining understanding of the underlying pathophysiology. External collaborators include investigators at outside institutions, including internationally, looking specifically at the participant’s phenotype and centralized repositories studying broader health histories.

**RISKS AND DISCOMFORTS**

The specific physical risks and discomforts, including those associated with obtaining blood, skin or saliva from family members and patients, are minimal. These risks will be minimized by having blood draws and skin biopsies collected by experiences health care professionals. There is no known physical risk for collecting saliva samples.

Information about participation in a study on a genetic condition may influence insurance and/or employers regarding health status. To help prevent disclosure, information about participation including research data collected and the results of the research will not be placed in the BCH medical record. In addition, data and samples will be coded without the use of identifiers in the REDcap database. Only the appropriate study members will be able to view identifiable information. Since the link between a study participant and their HSP ID is only stored in the HSP databank system and a separate research record, the chance that loss of confidentiality will occur is highly unlikely.

There is a chance that participation in this study could cause psychological distress. If these feelings arise at any time during the study, the participant/family may contact us and we will arrange for them to speak with a medical professional and notify the PI, so that the PI can follow up on such referrals. The PI will keep track of these referrals so that this can be reported to the IRB in the annual review. At that time, the IRB can assess whether the number of referrals warrants a change in the consent form or other practices in the protocol.

All data will be stored in REDCap and transfer will be secure and HIPAA compliant. The data stored for the project will contain a minimum of participant identifiable data, further minimizing any risk. All identifiable data will be stored in a locked filing cabinet in the study coordinator’s office or in a double password protected database. Only coded data and samples will be shared among future collaborators without the expressed consent of the participant. Not sharing information about participation in this study with others will also minimize the risks associated with this study.

**POTENTIAL BENEFITS**

There is a possibility that information learned through this project would provide a participant’s family with more information about the disorder in their family. However, there is not expected to be any direct benefit from participation in this study. However, the identification of genes that regulate the onset of various rare disorders may provide insight into the causes of AP-4-HSP and HSPs in general. We believe that the potential advancement of scientific knowledge from this study will allow progression from understanding to the prevention and cure of AP-4-HSP and HSPs in general and therefore, outweighs the minimal risk of study participation. Thus, the results of this study will likely benefit many children with this condition and help us understand the basis for it.

**PRIVACY PROVISIONS**

In order to protect participants’ privacy, participants will only be approached about enrollment after they express interest in the research to their clinician, contacts us directly or does not opt out from contact. If possible, initial contact with potential participants will be made in the same method they contact us, i.e. by phone, email, or mail.

All searchable information will remain de-identified to those querying the repository. Only after explicit consent from the participant/participant’s family will identifiable information be made accessible to specifically named investigators, or physician(s) involved in the participant’s care. This process will allow investigators with specific areas of interest to contact participants for other studies, while also ensuring privacy and autonomy to the participant (see below in section 5.b. for more information about this process).

**CONFIDENTIALITY PROVISIONS**

In order to maintain participants’ confidentiality, the participant database with identifiable information is double-password protected and only directly accessible by the PI, research coordinator/s, lab supervisor or other study staff members under their direct supervision who have appropriate human subjects training. Participants’ research files are maintained in a locked room except when in use and are only accessible by study personnel. Participants’ information is de-identified whenever shared with non-study personnel other than referring clinicians, collaborators, other individuals requested by the participant, or when samples are sent to a licensed clinical genetics diagnostic laboratory that has agreements with BCH. Data shared by clinical labs or amongst collaborators will be de-identified to the best of our ability.

**REFERENCES**

Ebrahimi-Fakhari, D., et al., Clinical and genetic characterization of AP4B1-associated SPG47. Am J Med Genet A, 2018. 176(2): p. 311-318.

**APPENDIX MATERIALS** –

Attached are the following appendices:

**A. Letters to eligible families**

Appendix 1. Consent

Appendix 2. Medical Record Release Form

Appendix 3. HSP Natural History Study Questionnaire

Appendix 4. Follow-up HSP Natural History Study Questionnaire

Appendix 5. CPCHILD Questionnaire

Appendix 6. GMFCS Family Report Questionnaire

Appendix 7 Blood Collection Instructions

Appendix 8. Skin Biopsy Collection Instructions (outside of BCH)

Appendix 9. Outside referral form

Appendix 10. Initial contact letter to Families

Appendix 11. Initial contact letter to Clinicians

Appendix 12. Initial contact letter to genetic testing laboratories, local geneticists, neurologists, genetic counselors, and behavioral/developmental pediatricians