Childhood Liver Disease Research Network (ChiLDReN)

BILIARY ATRESIA STUDY IN INFANTS AND CHILDREN (BASIC)

Manual of Operations (MOO)
Version 3.2
April 3, 2017

Table of Contents

List of Appendices

1 Overview

1.1 Purpose of the Manual of Operations (MOO)

The MOO is designed to facilitate consistency in protocol implementation and data collection across studies, study participants, and study sites. Further, the MOO provides reassurance to all participants that scientific integrity and study participant safety are closely monitored and increases the likelihood that the results of the study will be scientifically credible. The MOO is analogous to a toolkit, in that it contains information needed for the conduct and operations of the study, and it can be used as a training document. See Appendix A for Summary of Changes to MOO version April 3, 2017.

The MOO is a dynamic document that is updated throughout the course of the study to record changes and refinement of procedures. The version number and date should appear on each page of the MOO (header) to track all changes and additions to the document. The MOO may be downloaded from the Childhood Liver Disease Research Network (ChiLDReN) website. The Data Coordinating Center (DCC) will inform the study site via email of any changes in the MOO or any other study-related documents. It is the study sites' responsibility to ensure that they are using the most current version. The study site does not need to archive old MOO documents. The DCC will have the archived documents available on the website.

1.2 Study Center Numbers

Each study site has been assigned a ChiLDReN Center ID number and respective National Institute of Diabetes and Digestive and Kidney Diseases (NIDDK) number. These site-specific IDs are used for electronic Case Report Forms (eCRFs) and sample shipping manifests. Table 1 shows each study site in the network with respective ID numbers.

Table 1. ChiLDReN Centers and Their Associated IDs.

Center Name/City	ChiLDReN Center ID[1]	ChiLDReN Center NIDDK ID[2] (BASIC)
Chicago	02 (B)	D02
Cincinnati	03 (C)	D03
Denver	04 (D)	D04
Philadelphia	06 (F)	D06
Pittsburgh	07 (G)	D07
San Francisco	08 (H)	D08
Houston	10 (J)	D10
Indianapolis	12 (L)	D12
Seattle	13 (M)	D13
Toronto	14 (R)	D14
Salt Lake City	15 (S)	D15
Los Angeles	17 (N)	D17
Atlanta	18 (P)	D18

[1]The two-digit numeric ID is the ID as of 1/22/2009; the letter ID is the current ChiLDReN center ID.
[2]Assigned by the NIDDK data repository (Information Management System [IMS]).

1.3 Summary of Study

The purpose of this database is to collect the pertinent clinical information, genetic material, and body fluid samples to enable investigators to address the following hypotheses.

1.4 Specific Aims

1. To identify the gene or genes implicated in the etiology of biliary atresia (BA)
2. To identify polymorphisms that may be important in disease progression such as human leukocyte antigen (HLA) polymorphisms
    i. To perform high resolution HLA-A, B, C, DRB1, DRB3 DRB4, DRB5, DQA1, DQB1, DPA1, and DPB1 typing on patients with BA.
    ii. To utilize a novel computer algorithm that permits screening large numbers of HLA alleles to detect shared epitopes in patients with BA.
    iii. To assess the role of HLA polymorphism in incidence and severity of BA using traditional analysis of allele frequency and a novel shared epitope algorithm.
3. To characterize the natural history of the older, non-transplanted child with BA.

1.5 Hypotheses Related to Specific Aims

1. A genetic defect is a likely causative factor for BA among children with BA and multiple congenital anomalies.
2. Autoimmune factors are likely to contribute to disease progression or acquisition and can be identified by correlating HLA among children with BA to healthy controls and by comparison of those who develop early complications including variceal bleed, ascites, and growth failure compared with those who do not.
3. Sentinel events such as variceal bleeding, ascites, and growth failure are earlier predictors of death or need for liver transplantation than the pediatric end-stage liver disease score (PELD)
4. Health-related quality of life (QOL) will be impaired compared with healthy age-matched children and will relate to severity of illness.
5. Growth failure as measured by anthropometrics and nutritional supplementation will be predictive of onset of sentinel events (ascites, variceal bleed, death, and transplant) in the following 24 months.

2 Study Organization

2.1 Sponsor

The study is funded by the NIDDK, which is part of the National Institutes of Health (NIH). ChiLDReN is governed by a Steering Committee comprised of the Principal Investigators (PIs) from each participating clinical site, the DCC PI, and the NIDDK project scientist.

At the end of the grant period, specimens will be kept in repositories under contract to NIDDK for future use by investigators using a peer review process.

2.2 DCC

Arbor Research Collaborative for Health is the DCC for ChiLDReN studies. The DCC provides project management, logistical coordination, and statistical leadership for the development, implementation, and analysis of ChiLDReN studies. In addition, the DCC will conduct training in protocol implementation, data management, monitoring, and quality control. The DCC supports

regulatory and technical functions (i.e., ChiLDReNLink). For a list of DCC personnel and their roles and contact information, please refer to the Study Directory located on the website (ChildrenNetwork.org/Secured/StudyDirectory.aspx).

2.3 Clinical Sites and PIs

Participating centers and current site PIs are kept updated on the study directory, located on the ChiLDReN website ChildrenNetwork.org. Please refer to this website for up-to-date information.

2.4 NIDDK Data and Safety Monitoring Board

The Data and Safety Monitoring Board (DSMB) is appointed by the NIDDK and serves in a consultative capacity to the NIDDK.

The Board meets twice a year to provide independent review of data safety and monitoring procedures for ChiLDReN Studies. The Board may also convene to review the study protocols, if significant safety concerns arise. All protocols are reviewed and approved by the DSMB prior to implementation. The Board meets to examine endpoints, participant enrollment, protocol compliance, completion of samples and data, toxicity, and safety data from NIDDK-supported protocols. Since the ChiLDReN Studies are observational studies with no drug or other medical interventions, few adverse events (AEs) related to study-mandated procedures are expected. Reference the DSMB Charter and DSMB Membership List (see study website) for additional information regarding the DSMB.

2.5 ChiLDReN Website

Publicly-accessible information about ChilDReN is available on the ChiLDReN website home page. Some portions of the website are password-controlled to limit access to study group members (Clinical Centers, DCC, NIDDK, and the DSMB); protect the integrity, security, and confidentiality of sensitive project information and the information system; and allow auditing of appropriate use.

The website contains workgroup/subcommittee member lists, meeting agendas, materials, minutes, slides and presentations, master documents (including final protocols and consent templates), a calendar of events, and the study directory. The secure ChiLDReNLink data entry system is linked via the password-protected portion of the website, affording a double login/password for access to subject data.

2.6 Website Uniform Resource Locator (URL) and Access Instructions

The URL for the ChiLDReN website is ChildrenNetwork.org. Website management resides with the DCC. The DCC is responsible for login accounts, study directory updates, postings, and maintenance. Upon assigning a username and password, an automatic welcome email will be generated, informing the user that access has been granted to the restricted areas of the website. Users must change their system-assigned password within 72 hours of the welcome email receipt or website access will be denied.

Usernames and passwords should not be shared. New personnel requiring access to the ChiLDReN website should request a unique username and password. For new account requests or trouble with usernames and passwords, please contact ChiLDReN-Monitors@arborresearch.org.

3 IRB Submission and Regulatory Documents

3.1 Protocol Version Control, Finalization, and Approval Process

Protocol version control is extremely important to ensure that all participating sites and their respective Institutional Review Boards (IRBs) receive identical documents. Before a protocol is considered final and assigned a version (e.g., version 1.0), it must go through a formal review by the Steering Committee. Once finalized, the protocol document, consent templates, and any supplemental materials will be distributed to the sites by the DCC. Sites should submit only materials distributed by the DCC to their IRBs. Finalized protocols must NOT be edited, changed, or altered.

All amendments (a written description of a change or changes to or formal clarification of a protocol) must undergo a similar approval process. Sites should only submit protocols and amendments to IRBs as instructed by the DCC or NIDDK.

The current Protocol is Version 5.0, Amendment 4: November 3, 2016; (see Appendix B). The protocol can also be located on the study website, along with previous versions.

3.2 Consent Form Finalization and Approval Process

Protocol-specific consent document templates will be provided to all sites. Site-specific language should be inserted into the template. Please refer to Appendix C to view the Consent Templates.

Each site-specific Informed Consent form will be reviewed by the DCC for inclusion of all essential elements and compliance with federal regulations. After DCC review, the sites' Informed Consent document drafts will be reviewed by the NIDDK Biosample Repository staff. After that review, the NIDDK will return the draft consent to the DCC. The DCC will then return the reviewed/edited draft consents to the sites for correction and submission to the IRBs. Please note this is the process for initial approval prior to site initiation. For any amendments that include revisions to the consent form (additional procedures, increased risk, etc.), the DCC will send sites' IRB-approved consents to the NIDDK for final review.

File the IRB-approved consent document(s) (memo, consent, and other documents) in the site regulatory binder. Scan all approved documents and send electronically to the DCC. Throughout the course of the study, the DCC will request these documents when there is an amendment to the protocol and at the time of each site's IRB annual renewal.

3.3 Certificates of Confidentiality

Certificates of Confidentiality constitute an important tool to protect the privacy of research study participants. Certificates of Confidentiality are issued by the NIH and/or the Food and Drug Administration (FDA) to protect identifiable research information from forced disclosure. They allow the investigator and others who have access to research records to refuse to disclose identifying information on research participants in any civil, criminal, administrative, legislative, or other proceeding, whether at the federal, state, or local level. Certificates of Confidentiality may be granted for studies collecting information that, if disclosed, could have adverse consequences for participants or damage their financial standing, employability, insurability, or reputation. By protecting researchers and institutions from being compelled to disclose information that would identify research participants, Certificates of Confidentiality help achieve the research objectives and promote participation in studies by assuring confidentiality and privacy to participants. For more information, please see the NIH's Certificate of Confidentiality Kiosk: grants.nih.gov/grants/policy/coc/.

The DCC will obtain and maintain Certificates of Confidentiality for the study. These Certificates provide coverage to all clinical sites. Please refer to Appendix D to view the study's Certificates of Confidentiality.

3.4 Essential Documents for the Conduct of a Clinical Trial

Essential Documents are those documents that individually and collectively permit evaluation of the conduct of a trial and the quality of the data produced. These documents serve to demonstrate the compliance of the investigator, sponsor, and the monitor with the standards of Good Clinical Practice (GCP) and with all applicable regulatory standards. Sites must send all IRB approval notifications to the DCC via email to ChiLDReN-Monitors@arborresearch.org.

Required regulatory documents are to be kept on-file at the site. The regulatory binder must be kept current and available for review during site monitoring visits.

If the site maintains master files for CVs, lab normals, etc., a note to file should be placed in the study-specific regulatory binder to reflect the location of the documents.

REMEMBER, WHEN THE STUDY IS FINISHED AND READY FOR ARCHIVING, ALL DOCUMENTS IN THE MASTER FILES MUST BE COPIED TO BE STUDY-SPECIFIC DURING THE CONDUCT OF THE TRIAL. DOCUMENTS WILL BE STORED FOR THE LENGTH OF TIME DESIGNATED BY THE SPONSOR.

4 Informed Consent

4.1 Informed Consent Document

For each study, the DCC will provide protocol-specific Informed Consent templates for all study sites. Each study site will customize the template and the resulting site-specific draft consent will be reviewed by the DCC for inclusion of all essential elements and compliance with federal regulations. After DCC review, the sites' draft Informed Consent documents will be returned to the sites for correction and submission to the IRBs. Once approved the site will submit their Informed Consent documents to the DCC, the documents will be sent to the NIDDK for review and approval. If approved by the NIDDK a letter stating approval and version number will be distributed to each site through the DCC.

The written Informed Consent should be brief and written in plain language so that a participant who has not graduated from high school can understand the contents. An investigator or investigator delegate, participant (in the case of assent) or parent/guardian (in the case of a minor, as defined by the local IRB) and witness (if required by the local IRB) should each sign and date the Informed Consent documents. The participant should receive a copy of the signed and dated Informed Consent form. The study site must maintain a signed copy of the Informed Consent document for each participant in the study. GCP guidelines require that source documents should indicate that the Informed Consent form was signed, along with the date of signing.

4.2 Obtaining Informed Consent and Assent

All potential participants identified by the local PI and/or designee who meet the inclusion/exclusion criteria will be given the opportunity to participate.

Parents/guardians/participants will be given the consent/assent forms to review and opportunity to ask questions about the study.

Any additional questions they have will be answered prior to signing the consent/assent. Once the consent/assent form is signed, a copy will be provided to the parent/guardian/participant. All participants will be consented/assented by the PI and/or designee, who have received appropriate training regarding human subject protection and Health Insurance Portability & Accountability Act (HIPAA) compliance, as established by the local institutional governing body requirements. Local IRB enrollment regulations will be followed in all situations, including for example, if the child refuses.

Assent will be sought from participants, if applicable, based on age and local IRB requirements. Consent will be obtained before the participant is given a study ID number. Each study site is responsible for having an appropriate consenting procedure in place.

Telephone Consents may be used if approved by the DCC and local IRB/REB. Sites will need to follow guidelines set forth by local IRB/REB for use.

Failure to give Informed Consent renders the participant ineligible for the study. No research procedures will occur before Informed Consent has been obtained.

4.3 Re-Consent

If there is a change in any of the study procedures that may affect the participant, the Informed Consent document must be revised and again approved by the IRB. Any participants enrolled in the study prior to such changes may be required to sign the amended consent form, dependent on your local IRB requirements.

4.4 HIPAA Compliance

The HIPAA provides guidelines for investigators pertaining to protection of participant confidentiality. Investigators should review information provided in *Impact of the HIPAA Privacy Rule on NIH Processes Involving the Review, Funding, and Progress Monitoring of Grants, Cooperative Agreements, and Research Contracts* (http://grants.nih.gov/grants/guide/notice-files/NOT-OD-03-025.html). Investigators should contact their appropriate institutional officials to learn how the Privacy Rule applies to them, their organizations, and their specific research projects. Another helpful source is *Protecting Personal Health Information in Research: Understanding the HIPAA Privacy Rule*, NIH Publication 03-5388, available online at privacyruleandresearch.nih.gov.

At most study sites, a HIPAA form is presented to a potential participant for signature, in addition to the Informed Consent form, unless the necessary assurances are incorporated into the Informed Consent form. The HIPAA form describes participant and data confidentiality associated with the study.

4.5 Non-English-Speaking Participants

Many IRBs mandate whether a translated consent document is needed to obtain consent from non-English speaking participants or whether a translator can be used to obtain consent. Each study site must conform to their local requirements. With respect to completing CRFs, each study site should attempt to do their best to avoid errors as a result of translation.

4.6 Other Issues Related to Translators

A Human Protection certificate is not needed for the translator because the translator is only translating what the health care professional is stating; translators do not provide participant care or collect data.

Translation of any instructions is the responsibility of the study site and should be handled in the

same manner as for non-research participants.

All expenses and budget issues related to using translators fall to the study site and should be discussed with the PI prior to any expenses being incurred.

NOTE: Translator issues are study site-specific; they are the responsibility of the study site/PI.

5 Training

Site staff will receive study training from the DCC prior to implementation of the study. Training will include, but not be limited to, review of:

- Main protocol
- Informed Consent process
- MOO
- Data collection eCRFs
- Schedule of events
- Study-specific procedures
- Collecting, processing, labeling, shipping, and tracking of biosamples
- Use of ChiLDReNLink
- Site initiations and monitoring plan

Please notify the DCC (ChiLDReN-Monitors@arborresearch.org) of new study team personnel so they can receive the appropriate training and web access.

5.1 New Study Site Personnel

- When a study site has new personnel who will be working on the ChiLDReN study, please contact the DCC as soon as possible at ChiLDReN-Monitors@arborresearch.org**.** Please see study website for current Onboarding/Offboarding/Change of Information form.
- New study site personnel need to sign the site signature log and have their delegated study responsibilities listed.

6 Screening and Recruitment

6.1 Population

The study population to be enrolled will consist of male and female children and young adults with confirmed diagnosis of BA who are greater than or equal to 6 months of age. All racial and ethnic groups will be included.

6.2 Screening/Recruitment Plan

Recruitment will be open to any eligible participant. The method of contacting the study participant will conform to local IRB guidelines. In general, the parents or guardians of all eligible participants at each ChiLDReN center, or the participants themselves if 18 years of age or older, will be contacted to introduce the study. Methods to contact participants may include sending an IRB approved letter of introduction, followed by a telephone call and, if willing, arrangement of an appointment, at which time Informed Consent will be obtained. New patients who are at least 6 months of age and are not participating in the ChiLDReN Prospective Study of Biliary Atresia Epidemiology (PROBE) will also be approached. The study will be listed on clinicaltrials.gov and the ChiLDReN and related websites. Clinical sites may advertise to attract self-referrals, subject to local IRB approval.

6.3 Eligibility/Exclusion Criteria

Inclusion Criteria

- Participants need to have a confirmed diagnosis of BA determined by chart review, including review of pertinent diagnostic biopsy reports, radiologic reports, and / or surgical reports (if surgery was performed). Acceptable confirmation would include: any medical record document that confirms a Kasai procedure was completed (i.e., surgical report), or any diagnostic report, pathologic or radiologic report that has a definitive diagnosis of BA documented. This will be acceptable because the sites do not need to have all three to confirm a diagnosis of BA. PI confirmation of eligibility is required for study entry. If the diagnosis is indeterminate, this would be exclusion criteria for study entry.
- Participants need to be greater than or equal to 6 months of age up to and equal to the age of 20 (participants enrolled at 20 years of age will have one visit).
- Participants can either have their native liver or have a confirmed liver transplantation.
- Parent, guardian, or participant (if 18 years of age or older) is willing to provide Informed Consent. When appropriate, the subject is willing to assent.

Exclusion Criteria

- Active enrollment in the CHiLDReN study PROBE (NOTE: non-active PROBE participants may be enrolled.)
- Inability to confirm original diagnostic evaluation of BA
- Inability or unwillingness of family or participant to participate in all scheduled visits.

6.4 Exceptions to the Inclusion/Exclusion Criteria

Whenever the answer to an inclusion criterion is no, or the answer to an exclusion criterion is yes (even if the condition for the exclusion is fulfilled), an exemption will be required. The site study coordinator will notify the clinical monitor via email: ChiLDReN-Monitors@arborresearch.org and provide as much information as possible. Once the clinical monitor has sufficient information an email will be sent to the Exemption Committee for a vote. Additional emails may be required to gather information or answer questions from the committee. This committee will be composed of three of the site PIs, an NIH representative, and a DCC member. The committee will review the request and will decide by majority vote to either allow the participant to be enrolled or deny enrollment.

The Exemption request Form (Form 15) should be completed at the time of consent by the requesting site. Upon completion of review by the Exemption Committee the DCC will notify the site of the vote. Instructions for completion of the Exemption Form 15 are found in Section 7.2.2 **Miscellaneous Form 15.**

If the participant is allowed to enroll in the study through an exemption, answer the exemption question appropriately in Section G of the eligibility section of ChiLDReNLink.

Following the completion of the Exemption Form 15 the study coordinator must also complete a Protocol Deviation Form 40. The investigator will sign and date the Form 40, scan the form and submit to the DCC via: ChiLDReN-Monitors@arborresearch.org for review and DCC staff signatures. The Protocol Deviation is entered into ChiLDRenLink. The granted exemption and protocol deviation are reported to the sites IRB according to the site's IRB regulations for reporting of these types of events. The Exemption Form is filed in the participant's binder, and the Protocol Deviation is filed in the regulatory binder. Response from the IRB in reference to the exemption and protocol deviation are filed in the regulatory binder under the appropriate section.

6.5 Screening/Enrollment Logs

Screening and Enrollment Forms: Two essential documents that record all individuals who are screened (Screening Log) and enrolled (Enrollment Log) into a ChiLDReN study. The forms can be located in Appendix E and printed off or developed independently by study sites to capture this essential information. In either circumstance, they should be kept up-to-date throughout the study and made available for review for monitoring purposes.

7 Study Visit Details

7.1 Visit Descriptions

Native Liver: Participants with a native liver will be seen at baseline and will continue to be followed at annual visits until 20 years of age (+/- 6 months of age) or until transplanted. . If the participant undergoes a liver transplant, an annual visit collection of data and specimens should be obtained at the time of transplant. Once all consented DNA samples are completed, participation will end. In the manual and in subsequent analysis, these participants will be referred to as being in the pre-transplant cohort.

Transplant: Participants with a liver transplant before enrollment will be seen at **baseline only.** The baseline visit has an 18 month window to allow sites time to coordinate the collection of DNA samples and completion of study data. These participants are referred to in the manual and subsequent analysis as being in the post- transplant cohort. The only biosample collected for this population and their parents is blood for the DNA repository. This is optional, based on consent by parent and/or child. Participants will have a baseline visit with limited data collection (Table 3).

7.2 Data Collection

After Informed Consent is obtained, the participant will be followed at a ChiLDReN clinical site, where a detailed interview and physical examination will be conducted. Laboratory results and QOL assessments will be completed, and data will be recorded in the study database (ChiLDReNLink). Historical instructions and CRFs are located on the study website for reference.

If biosamples are not collected at the visit, but are able to be collected at a future visit that is within the visit window (example annual follow-up visit windows are +/- 6 months). For these visits, the biosample collection adhoc visit label (processed blood) should be used.

7.2.1 Baseline

Eligibility – Form 1

- Eligibility: Fulfillment of the inclusion/exclusion criteria is to be reviewed by the PI.The coordinator is to enter “yes” and the date of PI review on the eligibility form to signify PI confirms eligibility for the study. This data must be collected at recruitment. The data is entered in the ChiLDReN data entry system (eCRF). The Eligibility eCRF (Form 1) should be reviewed by the PI, confirming eligibility.
- The parents or guardians may selectively participate in this study. The participant can selectively participate in the DNA testing. Document this information on the Eligibility eCRF for each parent and child.
- For pre-transplant patients only – parents who refuse DNA sample collectionfor their child: The participant is still eligible if the parents or guardians elect not to have a DNA sample collected and stored at the repository.
- Section D – Use Pending when a parent is not present, but is expected to be present in the future. Use NA when there is no biological parent, or when the parent is not present and is not likely to be present. Update answer to Yes or No when the parent is available; enter date of consent on the Eligiblity eCRF.

Demographics – Form 3A

Demographics: Information on gender, ethnicity, and race will be collected at baseline. Data is collected by interview with the parents or guardians. All demographic data is required for any NIH study. Participants should be given the opportunity to self-report the ethnic and racial category(s) that they identify with. There should always be an option for a participant to deny reporting these data, in which case they will report this as "unknown/not reported". There may be situations where self-report of race and ethnicity is not feasible because the participant is incapable of providing the information. In these situations, investigators should determine what is the most reasonable approach, such as obtaining the information from other sources (e.g., medical records, family members, etc.) or whether it is more appropriate to indicate "unknown/not reported." Participants should be offered the choice to select as many racial categories that they deem appropriate. When reporting to the NIH, these individuals will be aggregated under the "more than one race" category.

If the interviewee needs ethnicity or race defined, definitions according to NIH guidelines are outlined below.

Definitions

- **ETHNICITY**
  Hispanic or Latino: A person of Cuban, Mexican, Puerto Rican, South or Central American, or other Spanish culture or origin, regardless of race. The term "Spanish origin" can be used in addition to "Hispanic or Latino".

  Not Hispanic or Latino: Does not fall into the Hispanic or Latino category as described above.

  Unknown/Unreported: This category should be used when a participant declines reporting these or is unable to answer this, and the investigator deems it appropriate to use this category instead of other means of data collection (e.g., medical records, family members, etc.).

- **RACIAL CATEGORIES**
  American Indian or Alaska Native: A person having origins in any of the original peoples of Northand South America(including Central America) and who maintains tribal affiliations or community attachment.

  Asian: A person having origins in any of the original peoples of the Far East, Southeast Asia, or the Indian subcontinent, including, for example, Cambodia, China, India, Japan, Korea, Malaysia, Pakistan, the Philippine Islands, Thailand, and Vietnam.

  Black or African American: A person having origins in any of the black racial groups of Africa. Terms such as "Haitian" or "Negro" can be used in addition to "Black or African American".

  Native Hawaiian or Other Pacific Islander: A person having origins in any of the original peoples of Hawaii, Guam, Samoa, or other Pacific Islands.

  White : A person having origins in any of the original peoples of Europe, the Middle East, or North Africa.

  Unknown/Unreported: This category should be used when a participant declines reporting these or is unable to answer this, and the investigator deems it appropriate to use this category instead of other means of data collection (e.g., medical records, family members, etc.)

Medical History – Forms 3B, 3C, & Form 24

Past Medical History 3B: These questions will vary based on whether the participant is in the pre-transplant or post-transplant cohort. The liver and targeted medical history information should be collected by verbal history and review of available medical records (from birth to present) upon enrollment of participants. The information should be reviewed confirming accuracy of the medical history listed and meeting ChiLDReN Network definitions. Participant targeted medical conditions that are currently active, but controlled with medication, should be reported as persistent on this form.

Past sentinel events are collected for participants enrolled in the pre-transplant cohort only. Medical history from birth to present will be collected by verbal history and review of medical records upon enrollment. The pertinent information is obtained by query of occurrences of: ascites, hepatopulmonary syndrome (HPS), nutritional supplementation, cholangitis, esophageal variceal bleed, other gastrointestinal (GI) bleed, sepsis, peritonitis, bone fracture,

Targeted liver events must meet the definition of sentinel events (for recording of these events on this eCRF.

At each follow-up visit, the clinical research coordinator (CRC) will ask the parents if the child has had any medical visits or sentinel events since the last study visit. This includes hospitalizations. These definitions are included in Appendix F and should be referred to and reviewed frequently when reviewing a participant's medical history at baseline and at all future study visit intervals.

Congenital Anomalies – Form 3C
Details of associated congenital anomalies will be obtained by chart review and verbal recollection from the primary caretaker. As part of the manual of operations, we have compiled a complete list of which anomalies to record.
This data is to be reviewed by the PI prior to data entry.

Follow-Up Medical History – Form 24
NOTE: Pre-transplant participants only: Follow-up medical history (Form 24) will be obtained at each follow-up visit for the pre-transplant participants. The sentinel events should be reviewed, and all new events occurring since the last recorded study visit should be recorded. If the participant is listed for transplant at a follow-up visit, the clinical site will need to complete a Form 25L to document the transplant listing data. Record all surgical/endoscopic procedures since the last study visit. If a subject has had an esophageal or other GI bleed, an endoscopy, a ligation procedure, and/or sclerotherapy, complete a separate Endoscopy eCRF Form 41. See additional instructions located in the Appendix G Endoscopy Form 41.

Pregnancy History – Form 4A & 4B

Both pre- cohort participants will complete this form at baseline, if aged 18 years and greater. Female participants age 18 and older of child-bearing potential will complete the Female Adolescent Questionnaire (Form 4A). All male participants age 18 and older will complete the Male Adolescent Questionnaire (Form 4B). Pre-transplant participants will complete this form at annual follow-up visits if 18 years or older.

Maternal and Paternal History – Forms 5 & 6
Maternal/Paternal (biological mother and father) family history with an emphasis on liver and autoimmune diseases and congenital abnormalities: Data is collected by interview with the parent(s). Detailed disease history should be obtained for all first-order biological relatives, including the infant's mother, mother's siblings, infant's maternal grandparents, and biological siblings of the infant. Information on siblings who are related by blood, including half-siblings, should be included on this form. If biological mother and/or father are not available, this

information may be collected from current guardian, if known.

Physical Exam – Form 7 & 20

Pre-Transplant Subjects Only

- Weight, height, head circumference (only recorded on participants less than 3 years of age), right triceps skinfold, right subscapular skinfold, and right mid arm circumference. It is acceptable to use the left arm for these measurements. ChiLDReNLink will have an additional drop-down to include left arm, if right arm was not used. Directed abdominal and skin exam pertinent to liver disease. At each follow-up visit, a physical exam will be performed for the pre-transplant participants.
- The baseline physical examination can occur at the time that eligibility is determined at the ChiLDReN clinical site. Often, this will occur prior to Informed Consent, and data from the medical record can be used. The checkbox for source data from the Medical Record should be checked.
- Following are guidelines that should be followed for the Physical Exam Assessments:

Head Circumference

- Head Circumference is only measured on subjects <3 years of age.
- Measured using a cloth or paper-measuring tape.
- The participant's head should be held straight. The measurer's eyes should be level with the measuring tape when placed around the participant's head.
- The tape should be halfway over the eyebrows in the front and on the most posterior aspect of the head (the occiput) in the back.
- The measurement is read to the nearest 0.1 cm and recorded.

Arm Circumference

- Measured with a cloth or paper-measuring tape.
- The measurer should locate the midpoint of the upper arm by measuring the length of the upper arm.
- The tape is wrapped around the arm, making sure not to indent the skin. The tape should be kept level around the arm in a plane perpendicular to the long axis of the arm, to ensure an accurate measurement.
- The measurement is read to the nearest 0.1 cm and recorded.

Skinfold Thickness

Triceps Measurement

- Measured along the midline on the back of the triceps of the right arm.
- Determine the midpoint located between the top of the acromial process (top of the shoulder) to the bottom of the olecranon process of the ulna (elbow). Pinch the skin so that the fold is running vertically.
- Grab the skin with the thumb and forefinger about 0.5 inches from the measurement site, following the natural fold of the skin.
- Lift the skin up from the muscle, apply the calipers, and wait for 4 seconds before reading the calipers. Fat is compressible, so reading the scale before or after the 4-second delay may affect the results.

Subscapular Skinfold Measurement

- Measured along the lower angle of the scapula.
- The pinch is made following the natural fold of the skin, approximately on a line running laterally (away from the body) and downwards (at about 45 degrees).
- Lift the skin up from the muscle, apply the calipers, and wait for 4 seconds before reading the calipers. Fat is compressible, so reading the scale before or after the 4-second delay may affect the results.

Liver Assessments

- Enter "not done", if applicable.
- Determine the location of the liver by palpation.
- Measure liver span:
  - If on the right side, measure the liver span in the mid-clavicular line on the right side.
  - If on the left side, measure the liver span in the mid-clavicular line on the left side.
  - If in the midline, use the larger of the two spans.
- Measure liver edge:
  - Either below the right costal margin or the left costal margin, based on the side that the liver is located.
  - If the liver is in the midline, use the larger of the two measures.
  - If not palpable, enter "Liver edge not palpable".
- Assess the liver texture, defined as:
  - Soft – normal, easily pliable liver edge.
  - Firm – rubbery feel to liver edge, but still pliable.
  - Hard – liver edge not pliable, feels like wood or stone.
  - Nodular and hard – hard liver with palpable nodules or bumps.
  - Not palpable

Spleen Assessments

- Enter "not done", if applicable.
- Determine the location of the spleen by palpation.
- When the spleen is not palpable, check "not palpable", and leave the other boxes empty.
- If on the left side, measure its size below the left costal margin.
- If on the right side, measure its size below the right costal margin.
- If in the midline, use the larger of the two measurements.

Ascites Assessments

- The Physical Exam Definition for Ascites is the presence of excess fluid in the abdominal cavity. Ascites is diagnosed by the presence of shifting dullness, ballottable fluid, bulging flanks, or a fluid wave.

TANNER Score

If child is 8 years or older, or if precocious puberty is suspected:

- The study coordinator will print either the GIRLS or BOYS version of the form and distribute it to the child for completion. This is a self-assessed form, and should be completed by the child.

- The study coordinator will enter results from the form into the physical exam section for the TANNER Results at a later date. The completed TANNER form will be stored in the participant's study binder as source documentation.

Labs – Forms 8 & 23

- If participant previously had labs done prior to enrollment visit you may enter labs from the prior 7 days if clinical labs were not done on day of enrollment visit.
- Lab entry window of +/- 3 months from date of visit for Annual Follow-up visits.

Pre-Transplant Participants Only

- Routine tests for clinical care will be obtained at baseline and yearly follow-up visits. These labs will be included on the eCRF if ordered as part of standard of care. Do not order these labs specifically for this study.

- These include: total bilirubin, indirect bilirubin, direct bilirubin, unconjugated bilirubin, conjugated bilirubin, AST, ALT, alkaline phosphatase, albumin, total protein, GGTP, PT/INR, , triglycerides, sodium, potassium, chloride, bicarbonate, creatinine, bun, glucose, CBC, vitamin A, 25-OH vitamin D and vitamin E levels. , The tests will be performed at specified intervals during outpatient follow-up. These tests will be analyzed in a Clinical Laboratory Improvement Amendments (CLIA) and/or College of American Pathologists (CAP)-approved laboratory since clinical decisions will be made based on the results.

- Lab Dates – If all the dates are the same as the visit date there is no need to enter any date. Otherwise, enter a date for the first lab (highest on the page) performed. Only enter a new date when the date of the lab data changes from the one listed above.

- Some labs will provide results in different units other than what is documented on the eCRF. If this is the case, please send a message thru the "contact us" link in the database and request to have the additional unit added. Sites are not expected or encouraged to convert units of measurement.

- When a complete panel is not obtained, check the ND box opposite of the name of the lab panel. When individual labs are not obtained check the individual box opposite the specific lab.

For pre-transplant participants who receive a transplant during the course of the study, the following labs will be recorded: total bilirubin, indirect bilirubin, direct bilirubin, unconjugated bilirubin, conjugated bilirubin, AST, ALT, alkaline phosphatase, albumin, total protein, GGTP, PT/INR, triglycerides, sodium, potassium, chloride, bicarbonate, creatinine, BUN, glucose, and CBC.

- Approximately, 6.5 ml of blood may be removed from the child at each visit to evaluate hepatic function, and electrolytes. In addition, 3 ml of blood may be removed to test vitamin levels. More blood may be withdrawn to perform additional clinically indicated lab tests.

- When the participant is a patient at the ChiLDReN site, the blood for clinical

tests will be drawn as part of the annual clinical evaluation of the subject. When the subject is a research subject and not a patient, this blood volume will be included in the total computation of blood volume for research use. However, this blood will not be drawn from research subjects who are post-transplant. These tests will be performed by the clinical research sites local lab.

Priority List for Blood Samples
When there is insufficient blood for all samples, the priority of blood samples is as follows:

1. Hepatic function and electrolytes and complete blood count*
2. Other clinically indicated tests (non-research tests only)*
3. Blood for DNA
4. Plasma*
5. Serum*

** These tests are not performed for research purposes when the participant is post- transplant.*

Note: When insufficient blood is collected for both ***plasma and serum***, first collect the blood for plasma and collect any remaining blood for serum.

QOL – PedsQL
Pre-Transplant Subjects Only
The PedsQL 4.0 (Pediatric QOL Inventory Version 4.0): Age-appropriate QOL information will be collected for Pre-transplant subjects. This evaluation will be performed at enrollment and at annual follow up visits for participants with their native liver.

Autopsy Report – Form 35A
In the event of death, if an autopsy report is completed record all relevant data on the autopsy CRF.

Endoscopy Form – Form 41
This form is completed for participants in the pre-transplant cohort only, when Follow-up Form 24 endoscopy related events are checked yes. This eCRF should be printed and the investigator should review and sign for source documentation. The form should be data entered into ChiLDReNLink.

7.2.2

Protocol Exemption Request – Form 15

- This form is completed by the site after the email request has been submitted to ChiLDReN-Monitors@arborresearch.org requesting a protocol exemption.
- B1. Describe the inclusion/exclusion criteria that are violated or reason for exemption being requested.
- B4-B5. Enter the name email address of the investigator requesting the exemption.
- C1. Check appropriate box once notified by DCC regarding decision of the Exemption Committee.
- C2-C3. Complete with date of decision and name of the person reporting decision to site. This could be name of DCC designee or name of chair of Exemption Committee.
- Z1. Should be checked yes.
- If samples from ineligible participants have been collected and sent to the repository, they cannot be used and will need to be destroyed. If samples need to be destroyed please notify the DCC by using the ChiLDRen-Monitors@arborresearch.org email address. Please specify samples to be destroyed (subject identifier, barcode, and visit). The DCC will notify the Repository and/or other appropriate lab. Once the DCC is notified by the Repository and/or other appropriate lab that samples have been destroyed we will send you an email confirmation to file in your study documents. Therefore, when an exemption is requested, please do not send samples to the repository until after the eligibility decision is made.

Final Status – Form 35

Pre-Transplant Participants Only Definitions:

- **Lost to follow-up**: Participant has missed 3 consecutive (annual) visits without known transfer of care
- **Other early termination**: Participant has moved out of treatment area, transferred to adult care, or transferred to non-ChiLDReN center
- **Completed Study**: Participant has aged out (reached 20.5 years of age)
- **Transplant**: Participant has received a liver transplant and completed DNA trio collection.

NOTE: Post-transplant (those transplanted during the study) participants who reach 20.5 years of age should be exited regardless of DNA collection status. The reason provided should be Completed Study.

FORM IS NOT COMPLETED FOR THE FOLLOWING PARTICIPANTS!

- Post-transplant participants have only one visit (baseline), which has an 18 month window to complete all study procedures, and their study participation ends.

Missing Visits

If a Final Status happens as a visit date and appears in your “widgets” on the homepage in ChiLDReNLink as an overdue visit you will need to mark the visit as a “missed visit”.

Protocol Deviation – Form 40

A protocol deviation is a departure from the expected conduct of an approved study that is not consistent with the current research protocol, consent document, or study addenda that had not been anticipated. All protocol deviations must be reported in writing to the DCC immediately upon discovery, using a protocol deviation report eCRF form, and must be reported to local IRBs/ERCs, if required by your local IRB/ERC.

A protocol deviation may be a divergence in a procedure from that indicated in the protocol (such as drawing more blood than indicated in the protocol). Specific categories are provided on the eCRF.

Enrolling a participant who does not meet eligibility criteria, but for whom enrollment is authorized by the Exemptions Committee, also constitutes as a protocol deviation. The NIDDK and the DSMB will track the enrollment of participants who require exemptions to the inclusion/exclusion criteria.

Protocol Deviations

A protocol deviation includes a deviation that impacts one of the following:

- The inclusion and/or exclusion criteria
- The ability of the sponsor to evaluate the endpoints of the study
- A consent violation

Protocol deviations also include situations that result in noncompliance with the study protocol, or GCP.

Below is a list of some protocol deviations(major and minor) the DCC will be tracking:

- Participant enrolled, but does not meet eligibility criteria
- Non-adherence to study design
- Loss of samples or data as per protocol schedule of events
- Failure to obtain Informed Consent prior to initiation of study-related procedures
- Falsifying research or medical records
- Performing tests beyond professional scope
- Working under an expired professional license/certificate
- Breach of confidentiality
- Improper or inadequate Informed Consent procedure

Study Coordinators will print out the Protocol Deviation Form 40, and complete the form with all available information. Once completed, the deviation form is scanned and sent to the DCC via ChiLDReN-Monitors@arborresearch.org. The DCC clinical monitors and clinical study process manager will review the deviation and determine whether it's a major or minor deviation, sign the deviation and return to the site for IRB submission.

NOTE: Further follow-up in reference to the action plan outlined in the protocol deviation may be requested from the DCC clinical monitor.

Protocol deviation reports are to be submitted to your IRB per their reporting procedures. The response from your IRB to the deviation reports are to be filed in the regulatory binder under major correspondence.

7.3 Schedule of Evaluations

The following two tables describe the evaluations to be performed at each visit: Table 2 for a subject with a native liver (pre-transplant cohort) and Table 3 for a subject who had a liver transplant prior to study enrollment (post-transplant cohort). A visit may extend over more than 1 day. When possible, research visits will be scheduled to coincide with clinical visits.

Table 2: Evaluations for Participants Enrolled with Native Liver				
Evaluations for participants enrolled with native liver	**Baseline**	**Annual follow up**	**At transplant**	**Once (any visit)**
Window for visit		± 6 months		
Informed consent & eligibility	X			
Lab Testing				
CBC	X	X	X	
Hepatic function	X	X	X	
PT/INR	X	X	X	
Vitamin D: 25-OH vitD	X	X		
Vitamin A with RBP	X	X		
Vitamin E	X	X		
Directed Physical Exam				
Weight	X	X	X	
Height	X	X	X	
Head circumference	X	X	X	
Vital signs/anthropometry	X	X	X	
Liver size & texture	X	X		
Spleen size & texture	X	X		
Skin	X	X		
Information				
Demographics (gender, race, DOB, ethnicity, #)	X			
Targeted Liver and medical history (SPLIT) (lifetime), PELD/MELD	X			
Detail of associated anomalies	X			
Medications/herbal remedies	X	X	X	
Targeted liver events		X	X	
Quality of life (PedsQL4.0)	X	X		
Pregnancy questionnaires	X	X		
Specimens from Child				
Serum for repository	X	X	X	
Plasma for repository	X	X	X	
DNA for repository				X
Liver specimens for repository			X	
Specimens from Parents				
DNA for repository				X

TABLE 3: Evaluations for Participants Recruited with Liver Transplant	
Visit Window: Complete data collection within 18 months	**Baseline**
Informed consent & eligibility	X
Information	
Demographics (gender, race, DOB, ethnicity)	X
Detail of associated anomalies	X
Target liver and medical /surgical history	X
Family medical history	X
Specimens from Participant	
DNA for repository	X
Specimens from Parents	
DNA for repository	X

7.4 Participant Follow-up/Status

7.4.1 Time of Transplant for Participants Enrolled as Pre-Transplant in BASIC

If Child Has Transplant: When a transplant is performed, samples should be collected for the ChiLDReN repository and for the NIDDK repository (Table 2). Labs and medical history are collected at time of transplant, using the last data available before transplant.

7.4.2 Termination or Withdrawal of Participant

The patient's parents or guardians may request that the patient be removed from the study at any time. In addition, the PI investigator may withdraw a participant from the study if the PI determines that it is in the participant's best interests.

Note: Upon request of the parents or guardians, samples and data that have been submitted to the NIDDK repository or to the DCC may be destroyed unless the samples have already been used or the data have been included in reported analyses, or unless the linkage between the research identifier and the participant has been destroyed.

When the study ends at a clinical site or the participant completes the study, the linkage between the samples and the participant will be destroyed. Once this linkage has been destroyed, it will no longer be possible to withdraw samples and data from the repository and the database in response to a patient request.

7.4.3 Visit Windows

Clinical Visits
Visit window for follow-up visits for pre-transplant participants is ± 6 months.

Transplant Visit
When a transplant is performed, samples should be collected for the NIDDK repository and lab data, and if more than 3 months since the last annual follow-up visit, a medical history should be repeated at the time of transplant, using the last data available before transplant.

Out of Window Visits
Annual visit windows are ± 6 months from the baseline visit to the next annual follow-up

visit. Visit compliance will be monitored by the DCC and reported to the site on a regular basis. If a study visit is unable to be scheduled within the timeline for that visit year, the study coordinator will mark the visit as missed and complete the next visit within the visit window (ChiLDReNLink User Guide, Appendix H).

In Hospital at Time of BASIC Visit

The clinical site should make all attempts to get as much data as possible from the hospital records for that BASIC visit. If this is not possible, the CRC should document the visit as a missed visit. At the next scheduled visit, it is the CRC's responsibility to collect all data related at that hospitalization (if the participant has been discharged) and report it in the eCRFs.

8 Specimen Collection

8.1 Schedule for Specimen Collection from the Child

NOTE: Blood samples should be drawn at the same time that blood is taken for clinical testing or when there is IV access for a clinical procedure. Blood samples must be drawn in accordance with local IRB regulations with respect to timing and amounts.

- Baseline
- Annual Follow-Up visits through age 20 or until transplanted
- ***Transplantation

***When blood for DNA has NOT been drawn prior to transplantation, also draw:

4 ml of whole blood in one ethylenediamine tetraacetic acid (EDTA) vacutainers provided by the Rutgers University Cell & DNA Repository that will be used for DNA extraction. This draw should only occur if consent for extraction and storing DNA has been obtained. This draw should occur either before or at transplantation or at a clinical visit after transplantation at the time of a routine blood draw. If it is drawn after transplantation, it should not be done until at least 2 weeks after transplantation.

***NOTE: When possible, blood samples will be obtained prior to liver transplant, when the transplant is 3 months or more after an annual visit.

8.1.1 Timetable for Collection of Specimens from the Participant (Child)

Serum	**Plasma**		**DNA Whole Blood/ or Saliva**	**Tissue Specimens**
PRE-TRANSPLANT PARTICIPANTS AT BASELINE:				
X	X		X	NA
PRE-TRANSPLANT PARTICIPANTS AT FOLLOW-UPS:				
X	X		Z	NA
PRE-TRANSPLANT PARTICIPANTS AT TRANSPLANT***:				
X	X		Z	• 5 frozen liver specimens: and • 20 unstained paraffin-embedded slides from wedge biopsy
POST-TRANSPLANT PARTICIPANTS:				
NA	NA	NA	X	NA

X: Specimen collected at this visit.
Z: Specimen is collected only once; at baseline or at a time that is convenient for the participant.
4 - If whole blood collection for DNA extraction is not possible or contraindicated, saliva may be obtained for DNA extraction purposes. 2 ml of saliva will be obtained in a saliva collection kit.
NA: Specimen not collected at this visit.
Plasma: 2 ml in ethylenediamine tetraacetic acid (EDTA) vacutainer to be processed into plasma and placed in 6 cryovials
Serum: 2 ml in Serum Separator Tube (SST) vacutainer to be processed into serum and placed in 6 cryovials

When a BA participant has a transplant but has never had a Kasai, instructions for samples taken at transplant (i.e., liver tissue, slides, serum, plasma and) should be followed in the same manner, regardless of whether or not the participant has had a Kasai.

Post-Transplant Collection

1. If whole blood is not obtained at the Baseline visit (only visit), try to obtain within the next year.
2. If unable to obtain within the next year, send a saliva kit to the participant, prior to the end of the 18 month window.
3. This entire process can be completed within the 18 month window.

8.1.2 From Each Parent at Baseline (or When Convenient)

	Process
10 ml whole blood/saliva	In 1 EDTA vacutainer DNA only – send to Rutgers within 24 hours

8.2 Collecting DNA for Rutgers

8.2.1 Rutgers: Specimen Collection and Processing

Collection: Collect the blood specimen into the vacutainers provided by Rutgers DNA may be collected via whole blood or, in the event that whole blood collection is not possible or contraindicated by saliva.

- Child: 4 ml in one 4 ml EDTA vacutainer or 2 ml of saliva collected in a saliva collection kit.when the participant weighs less than 50 ky (110lbs), OR
- 10 ml of whole blood in one 10 ml EDTA vacutainer or 2 ml of saliva collected in a saliva collection kit when the participant weighs over 50 kg (110 lbs). OR
- Parents: 10 ml in one 10 ml EDTA vacutainer or 2 ml of saliva collected in a saliva collection kit.
- The saliva samples will be sent to a facility (currently Rutgers University) under contract with NIDDK.

Processing: After collecting whole blood into the tubes, gently invert the tube six times to mix with additives, and keep them at room temperature. **Note:** If an inadequate volume is collected, a redraw of blood sample may be requested.

Collection of Saliva for DNA: With the approval of BASIC Amendment 4, each site will be sent 5 saliva kits for collection of saliva to be used for extraction of DNA when whole blood collection is not possible or contraindicated. It should be understood that the participant is responsible for shipping the kit(s) to Rutgers for processing, whether the kits are mailed to the participant's residence or received at a clinical visit. Sites will be responsible for ordering additional saliva kits as needed. Sites may order up to 10 saliva kits per month, if more than 10 are needed, prior permission from the DCC is required.

Each RUTGERS saliva kit contains the following items:

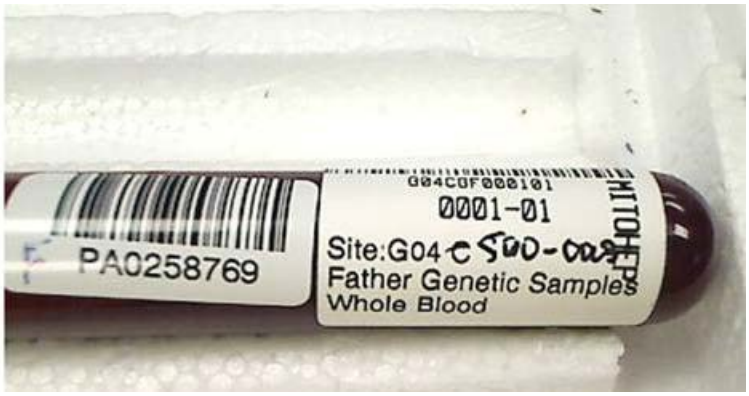
1. Saliva collection kit
2. Saliva collection form
3. Collection and shipping instructions
4. Bio-Hazard bag
5. Bubble Wrap pouch
6. Air bill for shipping to Rutgers
7. Shipping Envelope

Complete instructions for prepping the saliva kit, shipping and tracking the saliva kit in ChiLDReNLink can be found in **Appendix I**.

8.2.2 Rutgers: Shipping

See Appendix J for instructions on completion of Rutgers Collection Form and shipping instructions. All samples should be shipped at ambient temperature in an insulated container on the day of collection by FedEx. Label all samples with the labels provided by the DCC. Refer to the ChiLDReNLink User Guide (Appendix H) for label linking and shipping/manifest instructions.

Rutgers also requires the Participant ID to be written on the label. Please use a permanent marker or ink pen to write the Participant ID on all samples being shipped to Rutgers. This also applies to the parents' whole blood collection tubes. Be cautious not to write over the barcode section of the label. Example from Rutgers pictured below.



Place one copy of your shipping manifest in the outside plastic bag, along with the Rutgers Collection Form. Do not ship specimens on Friday unless the laboratory is notified first.

FedEx: Complete the FedEx air bill. Be sure the shipping label is marked for priority overnight delivery. Whole blood must be shipped on the day of collection. Do **NOT** keep the sample overnight. The account number is already on the air bill. Call FedEx (1-800-GO-FEDEX [1-800-463-3339]) for sample pick-up.

The address of the RUTGERS contact is:
Attn: CommStaff
RUCDR-Infinite Biologics
Nelson Laboratories
604 Allison Road (Rm C125)
Piscataway, NJ 08854
Phone: 848-445-1498

The RUTGERS lab supplies the blood collection tubes and the shipping kits. To request supplies. Please contact commstaff@dls.rutgers.edu.

8.3 Collecting Samples for NIDDK Biorepository

8.3.1 Collecting Tissue

From the participant at the time of transplantation (for the pre-transplant participants only): Liver tissue that is removed as part of the surgical procedure, but is not needed for diagnostic purposes, will be collected for the repository. Hence, when removed as part of the clinical procedure and based on availability after samples needed for diagnosis, the following may be obtained for the repository:

Collect:

- Tissue from the liver
- Unstained paraffin-embedded slides of the liver

Label each vial and slide with the appropriate label:

- Liver tissue (5 sections from explant tissue) and
- 20 unstained paraffin- embedded slides from wedge biopsy

NOTE: The research specimens must be removed from the native liver while it is fresh (not in Normal Saline or Formalin). Specimens should be taken as soon as possible once the hepatectomy is completed. The tissue should be sectioned within 10 minutes after being removed from the participant. Tissue should be taken from the right lobe, and be at least 1 cm deep to the capsule. One approach would be to bisect the liver or alternatively take a large wedge out of the right lobe. An approximately 2 cm X 2 cm X 2 cm piece of parenchyma should be isolated. The piece should be from as representative a section of parenchyma as possible. **From this block of tissue, 5 sections should be taken and placed in 5 cryovials**. Try to make the specimens as large as possible that can fit into the cryovial; 1 piece will be approximately 15 mm X 5 mm. Once placed in the cryovials, specimens should be snap-frozen in liquid nitrogen and transferred to the -70˚C freezer. Alternatively, specimens can be put directly into the -70˚C freezer. Note the time in minutes from harvesting to snap-freezing on the Surgery eCRF.

The remainder portion of this specimen (tissue adjacent to specimens placed in cryovials) should be placed in formaldehyde for processing by pathology. In addition to what else is deemed necessary, the pathologists should specifically interpret this section and cut extra slides from this specimen. Twenty unstained slides should be sent to the repository.

8.3.2 Procedure for Making Slides from the Tissue

- Paraffin-embedded slides will be prepared by the pathologist.
- For best results, all laboratories should use commercially-prepared coated glass slides.

- For each specimen, unstained sections of explant liver tissue cut at 4 microns should be provided; 20 unstained slides should be obtained.
- Label each slide with the slide labels provided by the DCC. Refer to the ChiLDReNLink User Guide for label linking and shipping/manifest instructions.
- Slides should be stored at room temperature in dry boxes. Slide boxes are provided by NIDDK Biorepository.
- Ship to the NIDDK Biorepository every month using the kit provided for the slides.
- NIDDK Biorepository does not supply slides to sites. Sites will supply their own slides.

NOTE: Transplants may be performed at any time during a 24-hour day, making it difficult to have a research member available at the time of the procedure. Please discuss with the transplant team the possibility of snap-freezing the specimen at time of transplant.

NOTE: Consent for these samples of the liver may be requested when the child is listed for transplant. Consent should already be in Study Consent.

8.3.3 Procedure for Snap-Freezing Liver Tissue

Liquid nitrogen is dangerous and needs to be handled appropriately. Do not let it make contact with bare skin. Liquid nitrogen evaporates (boils off) quickly, so it is necessary to check for sufficient liquid nitrogen in a container before using to freeze a sample. Use safety glasses whenever working with liquid nitrogen.

The goal is to freeze the liver sample immediately and to keep it frozen at -70°C or below. Small pieces of tissue can thaw in seconds. Allowing the specimen to thaw or warm can degrade it for use in many research studies.

Because timeliness is critical to proper freezing, it is important to run through all the steps in your mind before proceeding with the actual specimen. A “dry run” or two is often worthwhile. Before beginning, make sure that you have all the necessary supplies and that the tubes are appropriately labeled.

The surgeon will remove a piece of liver at the time of surgery. A portion will be sent to pathology for clinical purposes.

Primary Procedure:
NOTE: Samples should be snap-frozen as soon as possible; ideally this should be done in the operating suite.

Before starting, label the 1.5 ml cryovial with the bar code label provided by the DCC. (Refer to the ChiLDReNLink User Guide for instructions.) Remove any institutional identifiers prior to the following procedure. *For the label to adhere to the vial, attach the label as much in advance as possible of the vial being used (the previous day would be preferable).*

1. Pour liquid nitrogen into a large plastic container.
2. The liquid nitrogen will boil off rapidly, so check that the amount in the container is adequate at the time that you are ready to drop the specimen into the liquid nitrogen.
3. Place each specimen promptly into a labeled 1.5 ml cryovial. This should be done in a manner so that, if the specimen were to drop or spill, it would not fall onto the floor but could be instantly picked up. For example, working on a

tray may be helpful. A pair of forceps may be needed. It is not necessary to wrap the specimen in foil or other material. Just slide the tissue into the vial and cap the cryovial.

4. Drop the cryovial directly into the liquid nitrogen. The specimen will freeze within seconds. During this time, it is important to confirm that there is liquid nitrogen in the container, i.e., that it has not evaporated.
5. Take the liquid nitrogen containing the cryovials to the -70°C freezer.
6. Remove the cryovial with forceps and place it immediately in the freezer.
7. Frozen liver tissue can be sent with batch shipments of serum.

As an **alternate strategy**, pour liquid nitrogen into a 50cc plastic conical test tube secured in a test tube rack (5-10cc). Drop the specimen into the liquid nitrogen in the test tube. Transfer the sample into the labeled 1.5 ml cryovials. Quickly cap the cryovial, and drop the entire cryovial into a larger container of liquid nitrogen; remove the cryovial and place it into -70°C freezer.

Regardless of how the specimen is snap-frozen, once the cryovial is in liquid nitrogen, it should be transported in liquid nitrogen to the -70°C freezer. The specimen should then be retrieved from the liquid nitrogen and quickly placed in the -70°C freezer. Do not let the specimen thaw or warm. If the specimen should thaw during any stage, please note this in the comments section of the surgical eCRF.

If liquid nitrogen is poured into the vial, do NOT seal the cryovial until the liquid nitrogen has evaporated. Otherwise, the vial may explode as the liquid nitrogen thaws.

8.3.4 Collecting and Processing Plasma (Child Only)

Blood will be drawn using an EDTA (purple top) tube according to each hospital's venipuncture procedure.

Collection: Fill the EDTA (purple top) vacutainer.

- Child: 2 ml of blood in a 4 ml vacutainer

Inversion: After collection of whole blood into the EDTA vacutainer, gently invert the vacutainer 8-10 times.

Centrifugation: Blood samples should be centrifuged immediately for best results. If there is a delay, samples should be cooled on wet ice or refrigerated; however, it is best not to keep the samples on ice for more than one (1) hour. Centrifuge the EDTA blood sample at 4°C in a horizontal rotor (swing-out head) for a minimum of 10 minutes at 1,100 RCF (Relative Centrifugal Force) or per your institution's guidelines. The refrigerated centrifuge should be turned on at least 30 minutes prior to use to allow it to cool down.

If blood specimens are collected during "off hours" when Institutional Lab services are unavailable, it is acceptable to spin the specimens in a non-refrigerated centrifuge.

Be sure that there is not any participant-identifying material, except for the supplied labels, on the cryovials that will be sent to the repository.

Aliquots: Aliquot plasma into labeled 1.5 ml or 2 ml cryovials.

- Child: 1.2 ml should be available to be divided into six (6) x 200 µl aliquots.

If there is less volume, fill as many vials as possible with the following volumes. Do **NOT** divide the sample equally into the vials; fill as many vials as possible with the required volume.

Store filled cryovials in -70°C freezers until monthly batch-shipment..

8.3.5 Collecting and Processing Serum

Blood will be drawn using an SST vacutainer, according to each hospital's venipuncture procedure.

Collection: Fill the SST (gold-top) vacutainer.

- Child: 2 ml of blood in an SST vacutainer

Inversion: After collection of whole blood into the SST tube, gently invert the tube 8-10 times. After mixing, store the SST tube upright at room temperature for 30-45 minutes (but not more than 2 hours) to allow time for the specimen to clot.

Centrifugation: Centrifuge SST tube/blood sample at 4°C in a horizontal rotor (swing-out head) for a minimum of 10 minutes at 1,100 RCF or per your institution's guidelines. The refrigerated centrifuge should be turned on at least 30 minutes prior to use to allow it to cool down.

Aliquots: Aliquot serum into labeled 1.5 ml or 2 ml cryovials.

- Child: 1.2 ml should be available to be divided into six (6) x 200 µl aliquots.

If there is less volume, fill as many vials as possible with the following volumes. Do **NOT** divide the sample equally into the vials; fill as many vials as possible with the required volume.

Store filled cryovials in -70°C freezers until monthly batch-shipment to NIDDK Biorepository.

Refrigerated Centrifuges: When bio-samples are collected during "off" hours and a refrigerated centrifuge cannot be utilized it is acceptable to use a non-refrigerated centrifuge for processing the bio-samples. If the use of a refrigerated centrifuge becomes an issue, please contact the DCC Clinical Monitors via children-monitors@arborresearch.org for further instructions.

8.3.6 NIDDK Biorepository: Specimen Supply Kits

NIDDK Biorepositroy will provide one (1) shipping container at a time for vials. Up to three (3) specimen boxes of vials can be shipped within the container. Waybills will be included in the shipping kit.

If additional containers are needed, notify the via email at: niddk.mailbox@precisionformedicine.com and also cc': Eduard.chani@precisionformedicine.com.

Sites may also contact Eduard Chani PhD at 240-415-6052 (office) or 301-318-8218 (mobile). Email correspondence at the above address is preferred.

8.3.7 NIDDK Biorepository: Specimen Labeling

The DCC supplies bar-coded labels for each type of sample to be collected and/or aliquotted. Apply the label lengthwise to the vial and remove all other participant identifiers from the vials.

NOTE: The labels adhere better when placed on the vials an extended duration prior to freezing (the evening before when possible). This 'wait time' enables the temperature of the labels to equilibrate to the vial and form a solid bond. When preparing your cryovials for a visit or procedure, it's best to utilize the "prepped" collection status in ChiLDReNLink. Using the "prepped" status makes the labels easier to unlink if the participant doesn't show for a visit, refuses a blood draw, or the procedure is canceled.

8.3.8.1 NIDDK Biorepository: Specimen Packaging

Assemble the dry ice shipper for vials and package specimens according to instructions in Appendix K.

8.3.8.2 NIDDK Biorepository: Specimen Shipping

All frozen samples collected will be batch-shipped to the NIDDK Biorepository every month, or as needed. All shipments should be sent on Monday through Wednesday, according to study site schedule below:

- Chicago/Houston/Salt Lake City: 1st Mon.-Wed. of each month
- Cincinnati/Philadelphia/Indianapolis/Los Angeles: 2nd Mon.-Wed. of each month
- Denver/Pittsburgh/Toronto/Seattle/Atlanta: 3rd Mon.-Wed. of each month
- San Francisco: 4th Mon.-Wed of each month

Refer to the ChiLDReNLink User Guide for shipping instructions. The database will create an electronic manifest of samples that have been scanned and linked in the system for the Biorespository. Make sure to reconcile the manifest with the samples being shipped. Upon completion of this task in the database, an email notification will be sent to the repository email address: niddk.mailbox@precisionformedicine.com and Eduard.chani@precisionformedicine.com with the following information:

- Date of shipment (in the subject line)
- Shipping tracking number
- Barcode labels of each sample
- Number of specimens being shipped

Complete shipping via FedEx using the instructions in Appendix K. The

address of NIDDK Biorepositoryis:

Precision for Medicine
8425 Precision Way, Suite M
Frederick, MD 21701

The staff at the NIDDK Biorepository will interface with the DCC to acknowledge receipt of the shipment and/or record conflicts with the manifests and/or damage during transit. The repository and/or the DCC may follow up with sites regarding shipment and/or sample queries.

8.4 Samples from Ineligible Participants at the NIDDK Repositories.

If samples from ineligible participants have been collected and sent to NIDDK Biorepository or Rutgers, they cannot be used and will need to be destroyed. Therefore, when an exception/exemption is requested, please do not send samples to the repository until the decision is made about eligibility.

8.5 Lab Supplies

Biosample bar-coded labels will be shipped from the DCC to each study site in advance. Prior to each participant's visit, the CRC should review the participants' biosample repository collection for the expected visit. The labels should be linked and ready for use.

The following supplies are provided by the DCC:

Supply	**Use in Study**
1.5ml cryovials	
	Serum to NIDDK Biorepository
	Plasma to NIDDK Biorepository
	Biopsy to NIDDK Biorepository
Bar-coded labels for samples	Blood and saliva to Rutgers Serum and plasma toNIDDK Biorepository Slide labels and biopsy tissue label to NIDDK Biorepository

Sites are responsible for providing blood collection tubes and slides for tissue collection.

Saliva kits

- Sites may request up to 10 saliva kits monthly from Rutgers. Any more than 10 kits would require approval from the DCC.

9 Adverse Event/Serious Adverse Event (SAE)/Regulatory Bodies Reporting

Adverse Event (AE): An AE is any unfavorable, harmful, or pathological change in a research participant as indicated by symptoms, psychological or physical signs, and/or clinically significant laboratory abnormalities **that occur in association with the study procedures.** This definition includes intercurrent illness, injuries, and exacerbation of pre-existing conditions. Stable pre-existing conditions and elective procedures to address such conditions are not AEs. A change in a laboratory variable is considered an AE, if it was considered by the PIs to be clinically significant (that is, if it institutes a diagnostic evaluation or indicates additional therapy is necessary).

SAE: The term "serious" is based on patient outcomes associated with events that could threaten a patient's life or functioning.

An SAE is defined as any AE that results in any of the following:

- Death
- Life-threatening (Participant was at risk of death as a result of the event; it does not refer to hypothetical risk of death if the event had been more severe.)
- Requires inpatient hospitalization or prolongation of an existing hospitalization
- Results in persistent or significant disability/incapacity

- Congenital anomaly/birth defect
- Is medically significant or requires intervention to prevent one or other of the outcomes listed above

Expected AE: An Expected AE is any AE, the specificity and severity of which is consistent with the current standard of care, or is consistent with the risk information described in the Informed Consent document. The list of Expected AEs is compiled by the Steering Committee (SC) and is included in the protocol and Informed Consent documents.

Unexpected AE: An Unexpected AE is defined as any AE, the specificity and severity of which is not consistent with the current standard of care; or the specificity and severity of which is not consistent with the risk information described in the Informed Consent document or elsewhere in the current application.

Any Expected or Unexpected AE that also qualifies as an SAE, based on the criteria above, is considered an SAE by definition.

Related to Study: The phrase 'related to study' implies causality or attribution to the study procedures. For purposes of defining as an SAE, if a causal relationship cannot be ruled out, then an AE should be considered 'related to the study procedure(s)'. As noted above, it is very unlikely that any AEs will be attributable to this study.

SAE Reporting

Only report SAEs related to the protocol mandated procedures:

- Phlebotomy
- Survey Response

For an event to be considered as an SAE, one or all of the following must apply:

- Death
- Life-threatening
- Persistent or significant disability/incapacity
- Required in-patient hospitalization or prolonged hospitalization
- Congenital anomaly or birth defect
- Important medical events requiring medical or surgical intervention to prevent one of the outcomes listed above

The SAE reporting window for each participant begins with the first study procedure and ends 30 days after last study procedure.

SAEs must be reported to the DCC within 24 hours of the site's awareness of the occurrence. The site should complete the SAE report form in ChiLDReNLink within this time frame. Once you save the form, notification will immediately be sent to the DCC. All SAEs should be recorded during the time frame specified by the local IRB authority.

10 Study Monitoring

Monitoring is the act of overseeing the progress of a clinical trial, and of ensuring that it is conducted, recorded, and reported in accordance with the protocol, Standard Operating Procedures (SOPs), GCP, and the applicable regulatory requirement(s). Monitoring will include a combination of site visits and remote monitoring. Monitoring helps to catch problems and noncompliance before the actions become repetitive. It can identify systemic issues which can be corrected before a study is jeopardized.

Remote monitoring will occur at the DCC, and site-specific information in the form of reports reflecting data completion, integrity, and quality will be produced. These reports will be generated at least monthly and will be shared with the sites and NIDDK.

The DCC will produce reports showing:

- Overall data completion
- Data entry timeliness
- Form completeness
- Database queries comprised of logic checks
- Outstanding queries
- Biosample shipping
- Biosample collection
- Enrollment with consent status (including entire history of consent)
- Protocol deviations
- Visit completion
- Number (%) of queries resolved
- Number (%) of queries per study participant

Regulatory Review

- Other issues identified:
  - Best practices identified
  - Areas for improvement
  - Strategies for improvement
  - Barriers to success at site
  - Regular attendance at study coordinator calls

Site Processes Review

- Team Communication Plan
- Training Plan
- Recruitment Plan
- Retention Plan

The DCC will request a sample of de-identified source documents from the site to check for transcription errors in the database. The DCC staff may conduct site management calls, if needed, to ensure data quality compliance and data query resolution.

10.1 Goals of Monitoring

Proper monitoring helps ensure adequate protection of the rights of human subjects, the safety of subjects involved in a clinical investigation, and the quality and integrity of the data submitted. Ongoing monitoring of a clinical research study will be conducted with the intent to:

- Verify participant consent (for those studies requiring Informed Consent) for study participation has been properly obtained and documented, ensuring compliance with standards for protection of human participants.
- Verify research participants entered into the study meet inclusion and exclusion criteria.
- Verify the study is conducted in compliance with the protocol.
- Verify the accuracy of the data collected.
- Verify all essential documentation required by GCP guidelines are present, current, and

appropriately filed.

10.2 Monitoring Visits

The Clinical Monitor will send a confirmation letter detailing what will be reviewed during the monitoring visit at least 8 weeks prior to the proposed visit. Study sites will need to compile all supporting source documents (medical records, research shadow records, etc.) for participants who will be reviewed. If documentation is kept electronically (i.e., labs), provide a paper version at time of study site visit, or provide the Clinical Monitor with access to electronic records. Each study site should ensure that the site's regulatory binder/folder is up-to-date and available for review.

10.3 Frequency and Content of Monitoring Visits

The DCC will schedule a site visit with each site PI and study research staff every year. During the Site Monitoring Visit, the site's performance on the metrics described above will be discussed. The coordinator(s) and PI must be available for the conduct of the visit to be successful. The agenda for the visit will include such topics as:

- Essential elements of protocol adherence
- Regulatory document requirements
- Completeness or missingness of visits, forms, data, and samples
- Responses to data queries
- eCRFs and source documents

Additional monitoring activities, including more frequent on-site monitoring, may be scheduled at the request of NIDDK, the DCC, or the site PI.

As much as possible, data quality will be the responsibility of the study staff person entering the data. Data quality begins with the design of the CRFs and procedures and incorporates reasonable checks to minimize transcription and omission errors. Importantly, quality assurance measures are the internal validity checks for reasonableness and consistency. These checks may be built into the initial tables and cross tabulations that should reveal any remaining data quality issues.

Routine monitoring will be scheduled at appropriate intervals. Additional visits can be scheduled at the request of NIDDK or the DCC. For each visit, the Clinical Monitor will sign the monitoring log provided in the regulatory binder/file. For multi-day visits at a study site, the Clinical Monitor will sign the log for each day spent at the study site. For review of monitoring log please see Appendix L.

For observational studies, there will be review of 25% or 20 subjects, whichever is greater, monitoring of Informed Consent documents, and inclusion/exclusion criteria.

11 Study Completion and Closeout

Study closeout activities are performed to confirm the PI's study obligations have been met, and post-study obligations are understood. Closeout activities include, but are not limited to:

- Verification that study procedures have been completed, data collected, and study supplies are returned to the responsible party or prepared for destruction
- Review of completed and de-identified Screening and Enrollment Log
- Review of PI correspondence and study files against DCC records for completeness

- Assurance that all data queries have been completed
- Assurance that correspondence and study files are accessible for external audit
- Reminder to PIs of ongoing responsibility to maintain study records and to report any relevant study information to the sponsor or IRB
- Meeting with PIs to ensure they are aware of governing body obligations and requirements for record retention
- Assurance that PI will notify IRB of study completion and obtain copy of notification
- Preparation of a report summarizing study conduct

Appendix A: Summary of Changes

Summary of Changes version 3.2 Corresponding to Protocol Version 5 Amendment 4

Section	Change	Reason for Change
Title Page	Updated version number and date.	Updated versioning due to Amendment number 4.
All sections	Word “subject” changed to “participant” where applicable throughout MOO	To correspond with updating protocols.
All sections	Removed all references to urine collection when speaking of annual collection.	To correspond with amended protocol.
All sections	Removed references to Fisher Biorepository and inserted NIDDK Biorepository.	To reflect update of NIDDK designated Biorepository.
Section 2.4	Removed sentence indicating study is monitored by two separate DSMB Boards.	To reflect current information.
Section 3.1	Amended version of protocol to 5 and Amendment to November 3, 2016	To correspond to correct version.
Section 3.2 Consent Form Finalization and Approval Process	Inserted text at end of second paragraph. “the DCC will send sites' IRB-approved consents to the NIDDK for final review".	Clarified current process of DCC.
Section 3.4 Essential Documents for the Conduct of a Clinical Trial	Removed reference to list of essential documents in first paragraph. Removed paragraph regarding sites submitting annual progress reports to the NIH	To reflect current process of DCC.
Section 4.1 Informed Consent Document	Inserted text on DCC’s process for informed consent review prior to IRB submission and eventual submission to the NIDDK for review and approval once a site receives IRB approval.	Update sites on current process of the DCC.
Section 4.2 Obtaining Informed Consent and Assent	Removed last sentence in first paragraph “Parents/guardians/subjects will be asked to summarize in their own words what participation in this research study involves and whether they are comfortable with the risks benefits of participating in the research study.”	Not a current requirement.

	Inserted following 4th paragraph "Telephone Consents may be used if approved by the DCC and local IRB/REB. Sites will need to follow guidelines set forth by local IRB/REB for use.	Telephone Consents are an option that may be utilized.
Section 6.2 Screening/Recruitment Plan	Inserted text middle of paragraph "be contacted to introduce the study. Methods to contact participants may include sending an IRB approved".	Updated recruitment practices
Section 6.3 Eligibility/Exclusion Criteria Inclusion Criteria	Edited second bullet now reads: "Participants need to be greater than or equal to 6 months of age up to and equal to the age of 20 (participants enrolled at 20 years of age will have one visit)".	To correspond with updated inclusion criteria in amended protocol.
Section 6.4 Exceptions to the Inclusion/Exclusion Criteria	Removed text indicating Exceptions would be completed within ChiLDReNLink system. Inserted text in the first paragraph to indicate current process of notifying DCC with request for exception to criteria. The second paragraph is new and indicates when the Exemption request Form 15 should be completed. The last paragraph indicates the guidelines for completing the Protocol Deviation Form 40 as this needs to be completed when the Exemption Form 15 is completed.	Exception process has been updated by the DCC.
Section 7.1 Visit Descriptions	Native Liver paragraph: removed text and inserted text so paragraph now reads: Participants with a native liver will be seen at baseline and will continue to be followed at annual visits until 20 years of age (+/- 6 months of age) or until transplanted. New text reads: "Once all consented DNA samples are completed participation will end". Transplant paragraph: New text: "The baseline visit has an 18 month window to allow sites time to coordinate the collection of DNA samples and completion of study data."	To correspond with protocol amendment

Section 7.2 Data Collection	Inserted 2nd paragraph "If Biosamples are not collected at the visit, but are able to be collected at a future visit that is within the visit window (example annual follow-up visit windows are =/- 6 months). For these visits, the biosample collection adhoc visit label (processed blood) should be used".	Updated sites on the current practice of the DCC.
Section 7.2.1 Baseline, Eligibility – Form 1	Under first bullet Eligibility inserted text after criteria "is to be reviewed by the PI entering "yes" and date of PI review on the eligibility form signifies PI confirms eligibility for the study". The third square has been removed referencing eligibility criteria and Form 15. Second Bullet: text inserted after not to "have a DNA sample collected and stored at the repository. Also removed text "However, the subject is not eligible if the parents or guardians refuse to have blood collected (plasma and serum) for the biorepository.	Updated sites on the current practice of the DCC. Material is located in earlier locations. Updated language to reflect what was actually happening with biosample collection.
Medical History – Forms 3B, 3C, & Form 24	In first paragraph (Past Medical History 3B) the second sentence "liver and targeted medical history" was inserted. In the 2nd to last sentence "and meeting ChiLDReN Network definitions" was inserted.	Clarified for sites current practice of the DCC.
Sentinel Event Definitions	Material on Ascites, HPS, Nutritional Supplementation (NG or TPN), Cholangitis, GI Bleeding and Esophageal Variceal Hemorrhage, Sepsis, and Bone Fracture was all removed from the main protocol and inserted into Appendix.	Clarified for easier reading, also made for easier updating to Sentinel event definitions.
Physical Exam – Form 7 & 20	Pre-Transplant Subjects Only: Second box the baseline exam now has text stating exam "can occur" at the time eligibility is determined at the site.	Updated to provide sites more flexibility in data capture.
Skinfold Thickness	Inserted "Triceps Measurement" for list of directions that was already provided. Inserted Heading of Subscapular Skinfold Measurement and then 3 boxed bulleted directions for measuring.	Heading was missing. Directions for measuring were missing.

Labs – Form 8 & 23	Inserted two boxed bulleted information points. Under Pre-Transplant Participants Only: Under 2nd bullet point the following was deleted, "cholesterol", "with differential", "total serum lipids", "and bile acids (vitamin levels will only be recorded at baseline, not on follow-up visits)". Last bullet point has been removed.	Information on labs was provided as there was no information Information to correspond with protocol amendment. Information no longer being used.
Old Section 7.2.2 Annual Post-Transplant Follow-up Form 52	Post-transplant Follow-up for Subjects who Undergo Transplant while Enrolled in BASIC: This whole section has been deleted including Peds QL QOL Forms, and Functional Status Assessments.	To correspond with protocol amendment.
New Section 7.2.2 Miscellaneous Protocol Exemption Request	Form 15: This whole section was amended and revised heavily. All text under first bullet deleted and new text inserted so first bullet now reads " This form is completed by the site after the email request has been submitted to ChiLDReN-Monitors@arborresearch.org requesting a protocol exemption. 2nd bullet (B1) was revised B2 and B3 were deleted B4 – B5 was amended to read "Enter the name of the investigator requesting the exemption and the email address". This text is original text the rest of the text has been deleted. C1 has been inserted C2 – C3 has been inserted Z1 has been inserted B5 – B7 was removed. The last bullet was amended and text inserted to further elaborate on what would need to be done	The process has changed, the updated information is necessary for the process to be carried out properly.

	if samples had already been collected and sent to the Repository and/or other labs.	
Final Status – Form 35	Pre-Transplant Participants Only Definitions: Inserted 4 definitions Lost to follow-up, Other early termination, Completed Study and Transplant. Inserted Note: Post-transplant (those transplanted during the study) participants who reach 20.5 years of age should be exited regardless of DNA collection status. The reason provided should be Completed Study. Inserted text FORM IS NOT COMPLETED FOR THE FOLLOWING PARTICIPANTS! Text was also deleted at end of paragraph. Post-transplant participants now reads"have only one visit (baseline) which has an 18 month window to complete all the study procedures and their study participation ends. Missing Visits header and text was inserted.	Updated language to make more concise and clear on how to Final Status participants.
Protocol Deviation – Form 40	Removed first two paragraphs of text and inserted two new paragraphs.	Updated sites on current practice of DCC.
Major Protocol Deviations	Removed the word Major in title and in title under heading.	Updated sites on current practice of DCC
Minor Protocol Deviations	Removed the heading First paragraph has now been amended to read "Protocol deviations also includes those situations which results in noncompliance with the study protocol or GCP. Three paragraphs of text have been inserted below bullets to include directions on completing Protocol Deviation Form 40. Also included are instructions in the last paragraph regarding submitting Deviation Form to your local IRB.	Updated sites on current practice of DCC. New language makes practice easier to understand.
Section 7.3 Schedule of Evaluations	Table 2 and 3 have been deleted and updated tables have been inserted.	To correspond with amended protocol.

Section 7.4.3 Visit Windows	Under Transplant Visit: inserted text in middle of sentence after NIDDK repository and "lab data, and if more than 3 months since the las annual follow up visit". Out of Window Visits: removed text in middle of paragraph after visit, "unless a subject in the pre transplant cohort received a liver transplant. If this occurs the visits are then generated from the date of transplant (±6 months visit window)" In Hospital at Time of BASIC visit: In the middle of this paragraph the second sentence text after missed visit that reads "and complete a deviation report explaining the circumstances" has been deleted.	Updated sites on current practice of DCC.
Section 8.1 Schedule for Specimen Collection from the Child	The 2nd bullet point has been amended to read: "Annual Follow Up visits through age 20 or until transplanted" Blood for cell lines has been removed Blood amounts drawn have been updated	To correspond with the amended protocol. Decreased amounts in amended protocol.
Section 8.1.1 Timetable for Collection of Specimens from the Participant	Urine has been removed Saliva has been added In the footers text has been added: "If whole blood collection for DNA extraction is not possible or contraindicated saliva may be obtained for DNA extraction purposes. 2 ml of saliva will be obtained in a saliva collection kit. Post-Transplant Collection text was added 1. If whole blood is not obtained at the Baseline visit (only visit) try to obtain within the next year. 2. If unable to obtain within the next year, send a saliva kit to the participant prior to the end of the 18 month window. This entire process can be completed within the 18 month window.	Urine is no longer being collected. Saliva is added to correspond to the protocol amendment.

Section 8.1.2 From Each Parent at Baseline (or When Convenient)	Amount of blood being taken is amended and saliva inserted. 3. Number of EDTA vacutainers changed	To correspond with the amended protocol.
Section 8.2 Collecting Genetics/DNA for Rutgers	Removed Genetics in title. Title now reads: "Collecting DNA for Rutgers"	More accurate title to match amended protocol.
Section 8.2.1 Rutgers: Specimen Collection and Processing	Amended first sentence and now reads: Collection: Collect the blood specimen into the vacutainers provided by Rutgers". Inserted text: "DNA may be collected via whole blood or, in the event that whole blood collection is not possible or contraindicated by saliva". Information on saliva collection was added for child and parents as well as sentence on where saliva will be sent. Under Processing: In the first paragraph the following Note was inserted "if an inadequate volume is collected a redraw of blood sample may be requested". Collection of Saliva for DNA text with information on how to order saliva kits and what the saliva kits contain was added.	To correspond with the amended protocol. To correspond with the amended protocol and the addition of saliva kits.
Section 8.2.2 Rutgers: Shipping	In the first paragraph clarified that samples should be shipped on the day of collection.	Updated sites on current DCC practice.
Section Title 8.3 Title Change	Removed Fisher Bioservices and inserted NIDDK Biorepository Precision for Medicine.	Reflected change of NIDDK Biorepository.
Section 8.3.2 Procedure for Making Slides from the Tissue	In the 5th bullet a sentence was added "Slide boxes are provided by Precision for Medicine". Last bullet sentence added "Precision for Medicine does not supply slides to sites. Sites supply slides". Under Note: Removed last sentence beginning with "Please document on the shipping manifest form in the comments section the amount of time that the specimen remained at room temperature prior to freezing and/or sectioning".	Updated on instructions from Precision for Medicine.

	2nd Note: Added "Consent should already be in Study Consent".	
Section Title 8.3.4 Title Change	Fisher BioServices removed and Collecting and Processing Plasma inserted	Reflect change of NIDDK Biorepository.
Section 8.3.5 Title Change	Fisher BioServices was removed and Collecting and Processing Serum inserted Fisher BioServices Repository removed and Precision for Medicine inserted in last sentence. Paragraph added in regards to Refrigerated Centrifuges and biosamples being collected during "off" hours and the procedure to follow.	Reflect change of NIDDK Biorepository.
Section 8.3.6 Fisher BioServices: Urine Collection and Processing (Child Only) (Change Title)	This section was removed. Text on aliquoting of urine was removed as well.	To correspond with amended protocol
Section 8.3.7 Fisher BioServices: Specimen Supply Kits (Change Title)	Title change to remove Fisher BioServices and insert Precision for Medicine. Inserted paragraph stating Precision for Medicine will provide a slide shipper for shipping slides. Removed email address for requesting additional supplies from NIDDK Repository and inserted niddk.mailbox@precisionformedicine.com address to use to request supplies from Precision. Inserted phone numbers for study lead Eduard Chani at Precision for Medicine.	Reflect change of NIDDK Biorepository
Section 8.3.8 Fisher BioServices: Specimen Labeling (Change Title)	Title change to remove Fisher BioServices and insert Precision for Medicine First paragraph was amended and now reads: "The DCC supplies bar-coded labels for each type of sample to be collected and/or aliquotted. Apply the label lengthwise to the vial. Remove all other participant identifiers from the vials. Text added to end of NOTE: "When preparing your Cryovial for a visit or procedure its best to utilize the "prepped" collection status in ChiLDReNLink. Using the "prepped" status makes	Reflect change of NIDDK BioRepository Clarification needed for applying barcode labels. Helpful information/reminders

	the labels easier to unlink if the participant doesn't show for a visit, refuses a blood draw or the procedure is canceled".	for working directly in ChiLDReNLink
Section 8.3.8.2 Fisher BioServices: Specimen Shipping (Title Change)	Fisher BioServices removed and Precision for Medicine inserted. Fisher BioServices Repository removed and Precision for Medicine inserted. In the first paragraph changed wording so that shipments may be sent Monday through Wednesday. In list of sites for shipping deleted St. Louis. The address of Fisher Repository was deleted and the address of Precision for Medicine was inserted.	Updated sites on current DCC practice St. Louis does not participate in the BASIC protocol.
Section 8.4 Title Change	Removed the Repository (Fisher and Rutgers) and inserted Precision for Medicine and Rutgers.	
Section 8.5 Lab Supplies	Text removed at the end of first paragraph QOL surveys. Saliva was inserted and urine was removed in the table. Information on Saliva kits was inserted. Fisher changed to Precision for Medicine	To correspond with the amended protocol.
#9 AE/Serious Adverse Event (SAE)/Regulatory Bodies Reporting	Under SAE Reporting heading: removed Height/Weight Measurement as needing to be reported as an SAE	Measurements are for clinical care and not research related.
#10 Study Monitoring	Added heading Site Processes Review and four bullet points.	Site processes was added to monitoring reports in 2017
Appendices Appendix F Sentinel Events	Ascites – the sentence regarding "may be confirmed by a successful abdominal paracentecis was removed.	Updated to be the current practice.

	HPS – section was amended. Cholangitis – The original A. has been removed. The former B. is now A. and the text "with positive culture (blood or liver)" has been removed. The former C. is now B. GI hemorrhage – text removed "causing a drop in the subject's hematocrit >5%" Same text removed under Gastric Variceal Hemorrhage. Text inserted *"Endoscopy must be completed for diagnosis unless done within the past 6 weeks for a previous bleed. Spontaneous Bacterial Peritonitis – text inserted "Diagnosis of SBP is made requires documentation of the presence of one of three criteria. Removed sentence "Ascitic fluids should be inoculated into aerobic and anerobic blood culture bottles at the patient's bedside.	
Appendix I Saliva Kit Instructions	This Appendix is new	Updated to reflect Saliva Collection with amended protection.
Appendix K NIDDK Biorepository Shipper Assembly	This Appendix is new	Updated to reflect new Biorepository.

APPENDIX B: BASIC Protocol Amendment (Version 5.0, Amendment 4)

Biliary Atresia Study in Infants and Children (BASIC) Protocol BASIC, Version 5.0, Amendment 4:

Appendix B BASIC Protocol Amendmen

APPENDIX C: Consent Templates

Sample Consent Template for Older Children and Adults:

BASIC Post-TXP
Consent Older Kid &

Sample Consent Template for Older Children and Adults:

BASIC Pre-TXP
Consent Older Kid &

Sample Consent Template for Infant and Parents:

BASIC Post-TXP
Consent Parent FIN

Sample Consent Template for Infant and Parents:

BASIC Pre-TXP
Consent Parent FINA

APPENDIX D: Certificate of Confidentiality

Appendix D: Certificate of Confidentiality

DEPARTMENT OF HEALTH & HUMAN SERVICES

Public Health Service
National Institutes of Health

National Institute of Diabetes and
Digestive and Kidney Diseases
Bethesda, Maryland 20892

August 4, 2014

Our Reference: Confidentiality Certificate DK-09-016, Amendment #1

John Magee, M.D.
Section Head
Section of Transplant Surgery
Department of Surgery
The University of Michigan Medical School
2924F Taubman Center
Ann Arbor, MI 48109

Dear Dr. Magee:

This letter amends the Confidentiality Certificate protecting the identity of research subjects in your project entitled, **"Childhood Liver Disease Research and Education Network (ChiLDren)-Data Coordinating Center."**
Please note that the expiration date has been extended to **June 30, 2019**. This will enable the investigators to complete their research.

Please attach this amendment to the original Certificate as well as other pertinent materials.

If you determine that the research project will not be completed by the new expiration date, **June 30, 2019**, it will be necessary to submit a written request for an extension of the Certificate **three months prior to the expiration date**. Any such request must include the justification for the extension, documentation of the most recent IRB approval, and the expected date for completion of the research project. In addition, IRB approval must be maintained throughout the length of the study. Approval must be current and unconditional, or conditioned only upon the issuance of a Certificate of Confidentiality and documented by a letter or form signed by an authorized IRB representative.

Correspondence should be sent to:

Francisco O. Calvo, Ph.D.
Chief, Review Branch, NIDDK, NIH
6707 Democracy Blvd, Room 752
Bethesda, MD 20892-5452

Sincerely,

Gregory G. Germino, M.D.
Deputy Director, NIDDK

Gregory G. Germino -A

Digitally signed by Gregory G. Germino -A
DN: c=US, o=U.S. Government, ou=HHS, ou=NIH, ou=People, 0.9.2342.19200300.100.1.1=001433804 7, cn=Gregory G. Germino -A
Reason: I am approving this document
Date: 2014.08.18 13:17:19 -05'00'

Cc: James Ashton-Miller, Ph.D.

APPENDIX E: Site Screening and Enrollment Log

ChiLDReN 18January2018

Participant Screening Log
(Use only for participants who did not consent to participate)

Principal Investigator:_____

Study SiteName/Number: _____

Protocol: PROBE ☐ **BASIC** ☐ **LOGIC** ☐ **MITOHEP** ☐ **FORCE** ☐

A screening log is an essential document that records all individuals who entered screening and details the reasons why an individual was not enrolled in a study. It should be completed separately for each study.

	Screening Date	Gender	Age of Subject	Race/Ethnicity W: white; B: black H: Hispanic; A: Asian; O: other	**Reason for Exclusion/Comments**
1		M☐ F☐		W☐ B☐ H☐ A☐ O☐	
2		M☐ F☐		W☐ B☐ H☐ A☐ O☐	
3		M☐ F☐		W☐ B☐ H☐ A☐ O☐	
4		M☐ F☐		W☐ B☐ H☐ A☐ O☐	
5		M☐ F☐		W☐ B☐ H☐ A☐ O☐	
6		M☐ F☐		W☐ B☐ H☐ A☐ O☐	
7		M☐ F☐		W☐ B☐ H☐ A☐ O☐	
8		M☐ F☐		W☐ B☐ H☐ A☐ O☐	
9		M☐ F☐		W☐ B☐ H☐ A☐ O☐	
10		M☐ F☐		W☐ B☐ H☐ A☐ O☐	
11		M☐ F☐		W☐ B☐ H☐ A☐ O☐	
12		M☐ F☐		W☐ B☐ H☐ A☐ O☐	
13		M☐ F☐		W☐ B☐ H☐ A☐ O☐	
14		M☐ F☐		W☐ B☐ H☐ A☐ O☐	
15		M☐ F☐		W☐ B☐ H☐ A☐ O☐	
16		M☐ F☐		W☐ B☐ H☐ A☐ O☐	
17		M☐ F☐		W☐ B☐ H☐ A☐ O☐	
18		M☐ F☐		W☐ B☐ H☐ A☐ O☐	
19		M☐ F☐		W☐ B☐ H☐ A☐ O☐	
20		M☐ F☐		W☐ B☐ H☐ A☐ O☐	
21		M☐ F☐		W☐ B☐ H☐ A☐ O☐	
22		M☐ F☐		W☐ B☐ H☐ A☐ O☐	
23		M☐ F☐		W☐ B☐ H☐ A☐ O☐	
24		M☐ F☐		W☐ B☐ H☐ A☐ O☐	

Version 03

ChiLDReN ENROLLMENT LOG – FOR SITE USE ONLY

Protocol: PROBE ☐ BASIC ☐ LOGIC ☐ MITOHEP ☐ FORCE ☐

NAME	MEDICAL RECORD #	CONSENT DATE (if consented)	SUBJECT ID (if consented)	SCREEN/ BASELINE DATE	INELIGIBLE (Specify reason)

Page___ of ____ DO NOT SEND TO THE DCC

Versio

Childhood Liver Disease Research Network

(ChiLDReN)

BILIARY ATRESEIA STUDY IN INFANTS AND CHILDREN (BASIC)

SENTINEL EVENTS Definitions

ASCITES

Ascites is the presence of excess fluid in the abdominal cavity. Physical assessment should be by an experienced physician. Ascites is diagnosed by the presence of shifting dullness, ballottable fluid, bulging flanks, or a fluid wave on physical exam. Diagnosis is typically based on physical examination together with an ultrasound.

HEPATOPULMONARY SYNDROME (HPS)

Diagnosis of HPS requires documentation of both:

- **Deoxygenation:** pulse oximetry level of 97% provided a sensitivity of 96% and a specificity of 76% for detecting mild hypoxemia (pO2 <70 mm Hg) and
- **Intrapulmonary vascular dilation:** 2D transthoracic contrast echocardiography is the most commonly used technique. Agitated saline, which creates microbubbles visible on echocardiography, is used as a contrast agent. A positive test for intrapulmonary vasodilation occurs when delayed visualization of intravenously- administered microbubbles are observed in the left heart (bubbles are seen in the left heart after the third heartbeat, usually between the 3rd and 6th heart beat after injection).

NUTRITIONAL SUPPLEMENTATION (NG or TPN)

A. Nasogastric tube (NG tube) is a tube that is passed through the nose and down through the nasopharynx and esophagus into the stomach.
B. Total Parenteral Nutrition (TPN) is intravenous feeding that provides all daily nutritional requirements.

CHOLANGITIS

A. Cholangitis

Fever >38°C in a child with no other obvious source of infection with:

1. Acholic stools in a child who previously had stool pigmentation
2. Right upper quadrant pain/tenderness
3. Elevation of direct bilirubin by 25% and at least >1.0 mg/dl above previous level baseline
4. Positive bacterial culture of blood and liver

B. Possible cholangitis

Fever >38°C in a child with no other obvious source of infection with at least two of the following:

1. Acholic stools in a child who previously had stool pigmentation
2. Right upper quadrant pain/tenderness
3. Elevation of direct bilirubin by 25% and at least >1.0 mg/dl above previous level baseline
4. Rise in 2 or more of aspartate aminotransferase (AST), alanine transaminase (ALT), alkaline phosphatase, or gamma-glutamyl transpeptidase (GGTP) to 1.5X the upper limit of normal or >25% above baseline values if previously elevated
5. Clinical and biochemical (improvement in CB level and GGT levels) improvement (such as defeveresence, decreasing WCC, serum CB or GGT levels) in response to treatment with antibiotics

GI BLEEDING and ESOPHAGEAL VARICEAL HEMORRHAGE

GI hemorrhage:

Hematemesis, hematochezia, or melena, with either:

A. **Esophageal Variceal Hemorrhage**:
GI hemorrhage and documentation of actively bleeding esophageal varices by esophagoscopy* OR identification of esophageal varices and no other identifiable cause of hemorrhage.

OR

B. **Gastric Variceal Hemorrhage**:
Hematemesis, hematochezia, or melena, with documentation of actively bleeding gastric varices by endoscopy*.

*Endoscopy must be completed for diagnosis unless done within the past 6 weeks for a previous bleed.

SEPSIS

Sepsis is an overwhelming immune response to infection when immune chemicals released into the blood to combat the infection trigger widespread inflammation. This results in impaired blood flow, which damages the body's organs by depriving them of nutrients and oxygen.

SPONTANEOUS BACTERIAL PERITONITIS (SBP)

Diagnosis of SBP is made requires documentation of the presence of one of three criteria.

A. Diagnosis of SBP is made when the polymorphonuclear cell count in ascitic fluid is ≥250/mm3, and the ascitic fluid bacterial culture is positive.

B. The diagnosis of culture negative SBP is defined as any instance of negative ascitic fluid culture with an ascitic fluid neutrophil count of ≥250 neutrophils/mm3.

C. Bacterascites is defined as any instance of positive ascitic fluid culture with ascitic fluid neutrophil count of <250 neutrophils/mm3.

The interval between intra-abdominal operation and diagnosis of SBP should be at least 4 weeks. Secondary bacterial peritonitis, i.e., ascitic fluid infection caused by a surgically-treatable intra- abdominal source, should be excluded.

BONE FRACTURE

A bone fracture occurs when there is a break in the continuity of the bone. Document the site of the bone fracture.

APPENDIX G: Endoscopy Form 41

Subject ID: _____
Date of Visit: _____

ChiLDReNLink: BASIC

Pre-Transplant Form 41 GI ENDO

C1 Date of GI Endoscopy
Month Day Year Today

C2 Indication for endoscopy (check one):
- -- (selected)
- Screening (no previous episodes of bleeding)
- Surveillance (follow up of therapy)
- Ongoing therapy of varices
- Other, specify:
- Evaluation of GI bleeding

C3 Esophageal varices found:
- -- (selected)
- None
- Yes

Identify SMALL or LARGE and the GRADE, noting that the grade should reflect the largest size of varices identified, if more than one varix.

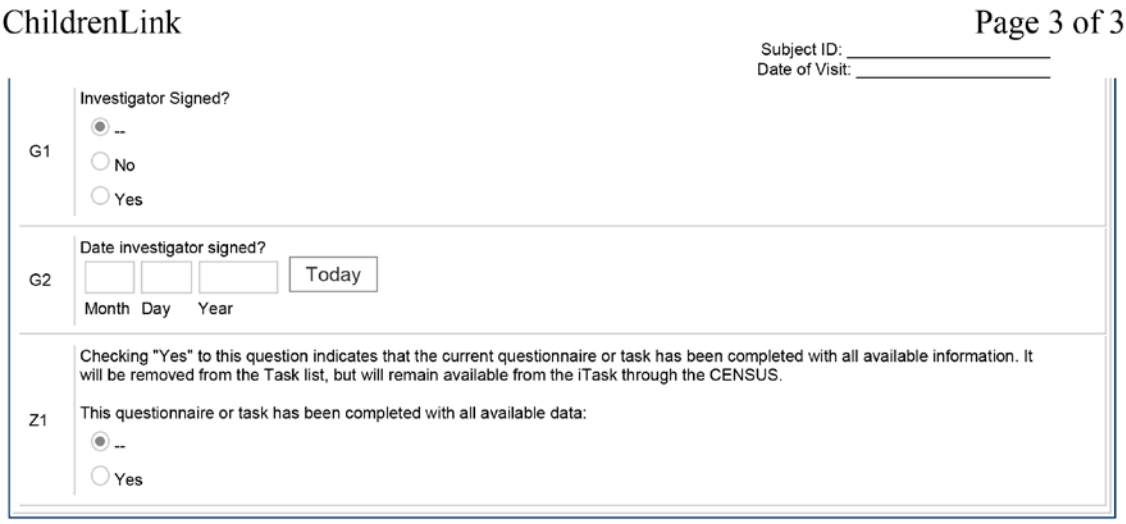
C4 Small varices
- -- (selected)
- No
- Yes

C5 If yes, choose grade:
- -- (selected)
- Grade I (Small)
- Grade I-II (Small to Medium, Flat)
- Grade II-III (Flatten with Insufflation)

C6 Large varices
- -- (selected)
- No

Subject ID: _____
Date of Visit: _____

	○ Yes
C7	If yes, choose grade: ◉ -- ○ Grade III (Large) ○ Grade III-IV (Very large, Protuberant) ○ Grade IV (Filling entire lumen, do not flatten with insufflations)
D1	Esophageal findings? ◉ -- ○ None ○ Yes
D2	What esophageal findings were reported? (check all that apply) ☐ Red markings (any) ☐ Active bleeding
D3	Which intervention(s) were performed? (check all that apply) ☐ None ☐ Band Ligation ☐ Sclerotherapy
E1	Stomach findings? ◉ -- ○ None ○ Yes
E2	What stomach findings were reported? (check all that apply) ☐ Portal Gastropathy ☐ Gastric varices
E3	Which endoscopic interventions were performed? (check all that apply) ☐ None ☐ Banding ☐ Glue injection ☐ Other, specify:
E4	Other interventions reported? ◉ -- ○ None ○ Yes
E5	Which interventions were reported? (check all that apply) ☐ Porto-systemic Shunt ☐ Non-selective Beta Blocker ☐ TIPPS ☐ Other, specify:



ChildrenLink

Page 3 of 3

Subject ID: _____
Date of Visit: _____

G1 Investigator Signed?
--
No
Yes

G2 Date investigator signed?
Month Day Year Today

Z1 Checking "Yes" to this question indicates that the current questionnaire or task has been completed with all available information. It will be removed from the Task list, but will remain available from the iTask through the CENSUS.

This questionnaire or task has been completed with all available data:
--
Yes

Endoscopy Form Instruction Sheet
Lee M. Bass, MD

Approved in January 2014 and now in the data collection materials as CRF 41, the endoscopy data collection form is designed to collect the retrospective endoscopy information. As this involves some interpretation of the endoscopy reports, it is best that an MD is the person interpreting the reports and marking the CRF.

The form is relatively easy to fill out. I suggest that each site ask their study coordinators to obtain the date of each endoscopic procedure for the patients in the BASIC cohort and pull the endoscopy report (If not already in your EMR (Oftentimes the GI tech in your procedural area can assist with pulling the reports). The site investigator can then sit down with the reports, mark off the CRF's and the coordinators can upload them to the database appropriately.

Going forward, the investigators are reminded that any endoscopy performed on any patients in the basic cohort should be recorded on CRF 41.

Following are some specific instructions for PI's and Study coordinators regarding the sections on the endoscopy form.

- Section B: Indication for Endoscopy
    - Screening implies that the patient has no history of GI bleeding and no history of therapy on their varices.
    - Ongoing therapy of varices is a patient with previous treatment of esophageal varices who is having the endoscopy in order to obliterate their varices (Generally 4-6 weeks after previous endoscopy)
    - Evaluation of GI bleeding
    - Surveillance (follow up of therapy)- This refers to patients who previously had successful treatment of varices and are having follow up endoscopic evaluation.
- Section C: Findings- Esophageal Varices
    - Clinicians are to record the largest varices present. For example if there are several grade I and one Grade III would say no to small varices(C1) and yes to the larger varices(C2)
    - Grade II-III varices: The medical literature has demonstrated in the BEST studies only fair inter-observer variability of grading of varices.(D'Antiga et al. JPGN 2015) Grading the difference between large and small varices has been shown to be more reliable than numbered grading. Our grading scale will essentially reflect this.
    - As it is particularly difficult to grade the size of varices retrospectively. The goal is to determine the clinician's best estimate of whether the varices fit into the large or the small category. This will be a subjective best estimate by the clinician. One suggestion is if the varices are unable to be flattened by insufflation or if there are positive red markings to grade those varices

as large and if those findings are not there, to grade those varices as small.

- Section F: Interventions (Shunts, TIPPS etc)
    - This information may be recorded on the endoscopy form but this is not consistent. The goal of this is to relate the findings to a timely intervention following the endoscopy. These details are generally captured in other CRF's if you are unable to find on the endoscopy form.

If there are specific questions or problems with the form, please do not hesitate to contact me lbass@luriechildrens.org

Thank you

Lee Bass

Appendix H: ChiLDReNLink User Guide

Please see the ChiLDReN study website for the ChiLDReNLink User Guide.

(https://childrennetwork.org/)

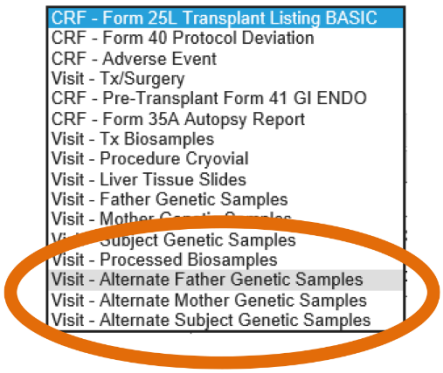
APPENDIX I: Saliva Kit Instructions

ChiLDReNLink Saliva Kit Process

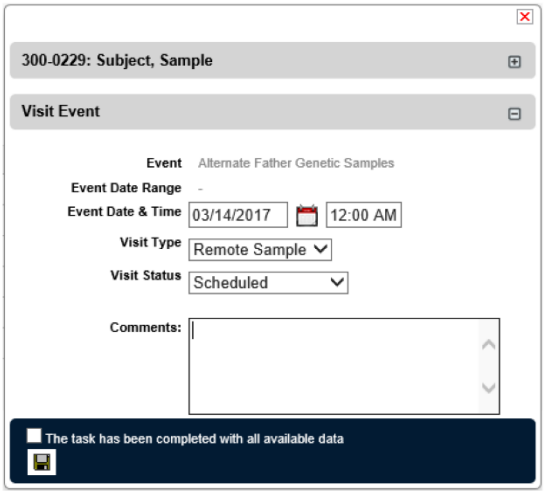
After receiving IRB approval and guidance from the DCC, follow these steps to work with saliva kits for a participant or their parents. You will need to mark the kit as shipped in ChiLDReNLink, but no manifest is sent to the repository.

Step 1 – Send Saliva Kit to Parent or Participant

1. Select the appropriate Ad hoc Visit – Alternate Genetic Samples from the dropdown list on the iTask page for the participant.
2. Choose from one of three new ad hoc Visits – Father, Mother, Participant



3. Enter the Event Date & Time as the date you are sending (or prepping) the kit to the participant or parent.
4. Select ***Remote Sample*** from the *Visit Type* dropdown list.
5. Select ***Scheduled*** from the *Visit Status* dropdown list and click the Save button.



6. Locate the visit in the iTask list and click the calendar icon to open the visit popup.

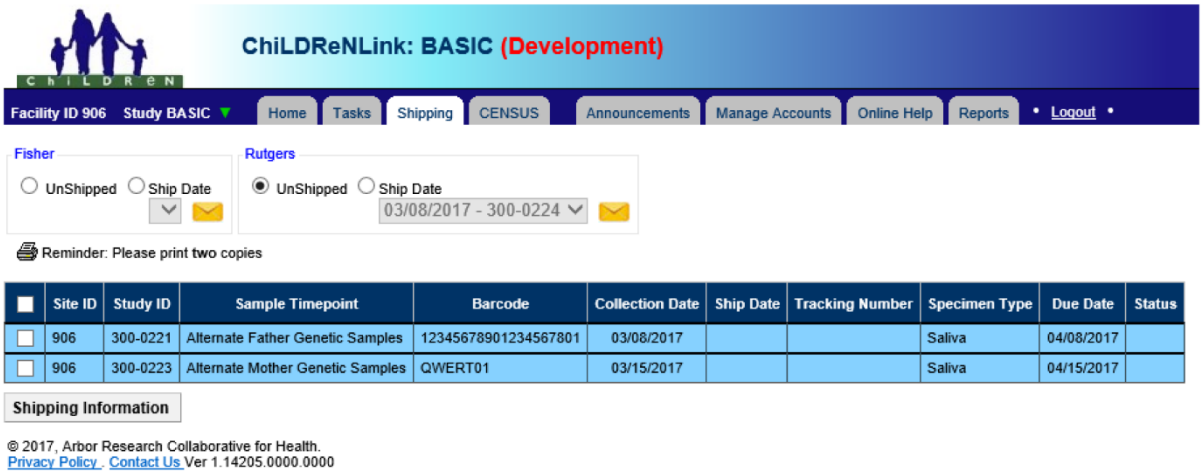
7. Expand the Sample Collection area and scan the saliva kit barcode.

   Note: The barcode will be on the pre-packaged label included with the saliva kit. The barcode is a 12-character text string. Labels will not be printed from ChiLDReNLink.

There is no need to open or work with the Sample Label Worksheet, because there are no ChiLDReNLink labels being used. (The worksheet is not available.) If needed, you may click the trashcan icon to unlink the saliva kit barcode from the participant.

Note that the saliva sample will display in the *Unshipped* list from the Rutgers box on the *Shipping* tab.

8. Locate the saliva kit in the Unshipped list for Rutgers and mark it as shipped.



Step 2 – Parent (or Participant) Sends Sample to Repository

1. The parent or participant will send the saliva kit directly to Rutgers using the FedEx shipping label included with the kit.
2. When you hear that the kit has been shipped, display the Shipping tab and mark the Saliva DNA sample as shipped. No manifest is needed.

   Note: The parent or participant may tell you themselves that the kit has shipped. You can also check online with the shipping carrier's website, using the tracking number from the waybill to confirm that Rutgers has received the saliva kit.

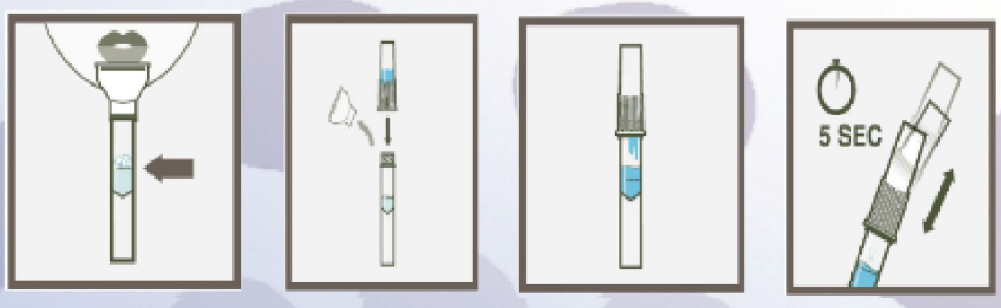
ChiLDReN

SALIVA SAMPLE COLLECTION & SHIPPING INSTRUCTIONS

RUCDR INFINITE BIOLOGICS

SAMPLE COLLECTION:

1. Do NOT eat, drink, smoke, or chew gum for 30 minutes before collecting the sample.
2. Fill the tube with saliva to the black wavy line (not including the bubbles). Do not overfill.
3. Remove the funnel from the tube.
4. Screw enclosed cap TIGHTLY until the blue solution in the cap empties into the tube.
5. Shake the tube for 5 seconds.



SALIVA COLLECTION FORM:

1. Complete and attach ID labels to the tubes, but do not cover the barcode on the tube.
NO HIPAA identifiers.
3. On the Saliva Colection Form, fill out the folowing fields:
    a. **Subject Code** - the patient's unique ID number as assigned by the Project leadership
    b. **Collection Date**
    c. **Gender** - if applicable
    d. **Courier Tracking #** - tracking number for package
4. Double check the ID information on the tube(s) matches the RUCDR Collection Form.

PACKAGING INSTRUCTIONS:

1. Place the container in the biohazard bag and seal the bag, then place both the container and the collection form in the yellow cushioned envelope. Seal the envelope.
2. Samples are assembled for shipment at room temperature.

SHIPPING INSTRUCTIONS:

1. Notify RUCDR - Inifinite Biologics via RUCDRLIMS by logging onto web: http://www.rucdr.org/lims.htm.
2. **VERY IMPORTANT: PLEASE SCHEDULE THE SAMPLES TO BE SHIPPED OVERNIGHT**
3. Call Federal Express (1-800-GO-FEDEX) to schedule a pick-up. Be sure to give FedEx the zip code of the shipping address, not that of the destination. **Do not put mailer in FedEx drop box.**

APPENDIX J: Rutgers Genetic Collection and Shipping

CHILDREN'S NETWORK
BLOOD SAMPLE COLLECTION & SHIPPING INSTRUCTIONS

RUCDR
INFINITE BIOLOGICS

SAMPLE COLLECTION:

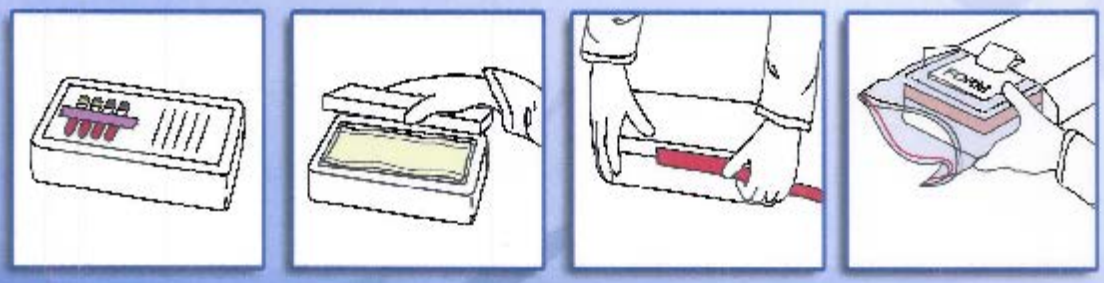
1. Complete and attach ID labels to the tubes, but do not cover the barcode on the tube. **NO HIPAA identifiers.**
2. Collect blood specimen in the 2 pediatric yellow (ACD) or 2 adult purple (EDTA) tubes provided. **Be sure to invert each tube gently 8 to 10 times to mix blood with additives and keep them at room temperature.**
3. Complete the enclosed collection form.
4. Double check the ID information on the tube(s) matches the RUCDR Collection Form.

REQUIRED PACKAGING COMPONENTS:

One Model 470 Safety Mailer (body & lid)
One 2-1/2" x 9" pre-cut section of absorbent material
One roll of waterproof tape
One press-lock plastic bag
One corrugated shipping carton with locking tabs

PACKAGING INSTRUCTIONS:

1. Place tubes in styrofoam mailer and secure with lab tape.
2. Place absorbent pad on top of secured tubes and close styrofoam mailer.
3. Seal styrofoam box with red waterproof tape.
4. Place sealed styrofoam mailer into plastic bag and collection form outside of plastic bag, then place all contents into cardboard shipping box.



SHIPPING INSTRUCTIONS:

1. Notify RUCDR - Infinite Biologics via RUCDRLIMS by logging onto web: http://www.rucdr.org/lims.htm.
2. Call Federal Express (1-800-GO-FEDEX) to schedule a pick-up. Be sure to give FedEx the zip code of the shipping address, not that of the destination. **Do not put mailer in FedEx drop box.**

WWW.RUCDR.ORG

APPENDIX K: NIDDK Biorepository Shipper Assembly

Appendix : NIDDK Biorepository: Assembling the STP 320 Repository Shipper

1. Upon receipt of the empty shipping kit from the NIDDK Biosample Repository, remove the "EMPTY PACKAGING" cardboard piece from the outer box.
2. Place up to 81 x 2ml cryovials in each specimen box. When packing vials, place them in the specimen boxes left to right, top to bottom. Group vials together by patient and visit.
3. Place each specimen box and an absorbent sheet inside a plastic biohazard bag. Seal the bag.
4. Place each plastic biohazard bag inside a white Tyvek envelope. Seal the envelope.
5. Place the Tyvek envelopes in the cardboard inner box. If only two specimen boxes are being shipped, fill the rest of the space inside the cardboard inner box with packing material (e.g., bubble wrap) or an empty specimen box to prevent movement during shipment. Close and tape the inner cardboard box and set it in the middle of the cooler.
6. Completely fill the space between the inner cardboard box and the inner walls of the cooler with dry ice pellets.
7. Place the lid on the cooler. Place the "EMPTY PACKAGING" cover and shipping log on top of the cooler lid.
8. Close and tape the outer cardboard box.
9. Place a checkmark in the block on the outer cardboard box next to "BIOLOGICAL SUBSTANCE, CATEGORY B". Do not cover this marking with labels.
10. Affix a label with your name and return address to the side of the box in the "Shipper:" block.
11. Affix the repository address label to the side of the box in the "Consignee:" block.
12. Affix the dry ice label below the repository address label. Enter the weight of dry ice on the label in kilograms.
13. Affix the "UN3373 BIOLOGICAL SUBSTANCE, CATEGORY B" label to the right of the dry ice label.
14. Use the pre-printed Fed Ex air bill to ship specimens to the NIDDK Repository:
    a. Section 1: Fill in your name, return address, phone number and the date. Leave "Sender's FedEx Account Number" blank.
    b. Section 6, Special Handling: Check "Yes, Shippers Declaration not required". Check the "Dry Ice" block; enter "1" and the weight of dry ice in kg.
    c. Section 7: Enter "1" under "Total Packages" and the total weight of the package.
    d. Follow the peel-and-stick instructions on the back of the air bill. As shown, affix the air bill to the side of the box adjacent to the labeled side.
15. Call Federal Express at 1-800-GO-FEDEX (1-SOD-463-3339). Give them the account number on the preprinted FedEx air bill (in Section 7, Payment) and your pickup address. FedEx will dispatch a courier to pick up the package. Please schedule shipments Monday through Wednesday to avoid weekend shipment delays. Do not ship samples on Friday; the repository is closed on weekends.
16. Send a shipment notification to the repository via email at niddk.mailbox@precisionformedicine.com and CC Eduard.chani@precisionformedicine.com on the day the package is picked up by FedEx. Include the 12-digit FedEx tracking number in the notification.
17. Contact Precision for Medicine via email at niddk.mailbox@precisionformedicine.com or Email or call Eduard Chani at Eduard.chani@precisionformedicine.com or office (240-415-6052) or mobile (301-668-3416).

APPENDIX L: Site Monitoring Log

Site Monitoring Log

Instructions: Routine monitoring will be scheduled at appropriate intervals, with more frequent visits occurring at the beginning of the study. For each visit, the monitor will sign the visit log provided in the regulatory binder/file and indicate the purpose for the visit. For multi-day visits at a site, the monitor will sign the log for each day spent at the site. The site personnel will sign the last column verifying the visit.

Principal Investigator:	Site Name:

Date(s) of Visit:	Purpose (specify study):	Specify Study	Signature of Monitor or Other Site Visitor	Signature of Site Personnel
__/__/__				
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All persons making study related visits must sign this log. This form should be kept in the regulatory file