Undiagnosed Diseases Network Manual of Operations

December 9, 2015

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Table of Acronyms

Acronym	Definition
ACMG	American College of Medical Genetics and Genomics
BCM	Baylor College of Medicine
bp	Base pairs
BWA	Burrows Wheeler Aligner
CAP	College of American Pathologists
CC	Undiagnosed Diseases Network Coordinating Center
CIRB	Central Institutional Review Board
CLIA	Clinical Laboratory Improvement Amendments
COI	Conflict of Interest
CR	Continuing Review
CRC	Clinical Research Center
CS	Undiagnosed Diseases Network Clinical Site
CSF	Cerebrospinal fluid
CSL	Clinical Services Laboratory
dbGaP	Database of Genotypes and Phenotypes
DOB	Date of birth
EEG	Electroencephalogram
EMG	Electromyography
FFQ	Food Frequency Questionnaire
FIPS	Federal Information Processing Standards
FISMA	Federal Information Security Management Act
FWA	Federalwide Assurance
gDNA	Genomic DNA
GSL	Genomic Services Laboratory
HA	HudsonAlpha Institute for Biotechnology
HIPAA	Health Insurance Portability and Accountability Act
HITECH	Health Information Technology for Economic and Clinical Health Act
HRPP	Human Research Protections Program
HPO	Human Phenotype Ontology
ICD	International Classification of Diseases
ICF	Informed Consent and Assent Forms
IRB	Institutional Review Board
LIMS	Laboratory Information Management System
MMA	Mercy Medical Angels
MRI	Magnetic Resonance Imaging
MRS	Magnetic Resonance Spectroscopy
NCBI	National Center for Biotechnology Information
NCV	Nerve Conduction Velocity
NGS	Next Generation Sequencing

NHANES	National Health and Nutrition Examination Survey
NHGRI	National Human Genome Research Institute
NHGRI-IRP	National Human Genome Research Institute-Intramural Research Program
NIH	National Institutes of Health
NIST	National Institute of Standards and Technology
NORD	National Organization for Rare Disorders
OMIM	Online Mendelian Inheritance in Man
ORDR	Office of Rare Diseases Research
OSC	Office of Strategic Coordination
PBMC	Peripheral Blood Mononuclear Cell
PCC	Patient Care Coordinator
PCP	Primary Care Physician
PCR	Polymerase chain reaction
PE	Paired-end
PHI	Personal Health Information
PI	Principal Investigator
PII	Personally Identifiable Information
QC	Quality Control
QWES	Quick Whole Exome Sequencing
SC	Undiagnosed Diseases Network Sequencing Core
SNOMED	Systematized Nomenclature of Medicine
SNP	Single Nucleotide Polymorphism
SRC	Scientific Review Committee
TAT	Turnaround time
UDN	Undiagnosed Diseases Network
UDNCB	Undiagnosed Diseases Network Central Biorepository
UDN NIH PO	Undiagnosed Diseases Network National Institutes of Health Program Official
UDP	Undiagnosed Diseases Program
UDPICS	Undiagnosed Diseases Program Integrated Collaboration System
UUID	Universal Unique Identifier
WES	Whole Exome Sequencing
WGS	Whole Genome Sequencing
WGL	Whole Genome Laboratory

I. Network Overview and Operating Procedures

A. Network Overview

The Undiagnosed Diseases Network (UDN) consists of 7 Clinical Sites (CSs), a Coordinating Center (CC), 2 DNA Sequencing Cores (SCs), a Model Organisms Screening Center, a Metabolomics Core, and a Central Biorepository.

The CC is located at the following institution, with the following PIs:

- Harvard Medical School, Boston, MA - Isaac Kohane, MD, PhD; Alexa McCray, PhD; and Rachel Ramoni, DMD., ScD.

The CSs are located at the following institutions, with the following PIs:

- Baylor College of Medicine, Houston, TX - Brendan Lee, MD, PhD
- Duke University (with Columbia University), Durham, NC – David Goldstein, PhD and Vandana Shashi, MBBS, MD
- Harvard Teaching Hospitals (including Boston Children's Hospital, Brigham and Women's Hospital, and Massachusetts General Hospital), Boston, MA - Joseph Loscalzo, MD, PhD
- National Institutes of Health (NIH), Bethesda, MD - William Gahl, MD, PhD, and Cynthia Tifft, MD, PhD
- Stanford Medicine, Palo Alto, CA - Euan Ashley, MD; Jonathan Bernstein, MD; and Paul Fisher, MD, Stanford University
- University of California Los Angeles, Los Angeles, CA - Katrina Dipple MD, PhD; Stanley Nelson, MD; Christina Palmer, PhD; and Eric Vilain, MD, PhD
- Vanderbilt University Medical Center, Nashville, TN – John Newman, MD, PhD and John Phillips III, MD

The SCs are located at the following institutions, with the following PIs:

- Baylor College of Medicine, Houston, TX - Christine Eng, MD
- HudsonAlpha (with Illumina), Huntsville, AL - Howard Jacob, PhD

The Model Organisms Screening Center is located at the following institutions, with the following PI:

- Baylor College of Medicine (with University of Oregon), Houston, TX – Hugo Bellen, DVM, PhD

The Metabolomics Core is located at the following institutions, with the following PIs:

- Battelle Pacific Northwest Laboratories (with Oregon Health & Science University), Richland, WA – Thomas Metz, PhD and David Koeller, MD

The Central Biorepository is located at the following institution, with the following PIs:

- Vanderbilt University Medical Center, Nashville, TN – Joy Cogan, PhD and John Phillips III, MD

The purpose of this cooperative research Network is to establish a national network added to and building upon the NIH Undiagnosed Diseases Program (NIH UDP). The objectives of this program are to: 1) improve the level of diagnosis and care for patients with undiagnosed diseases through the development of common protocols designed by a community of investigators; 2) facilitate research into the etiology of undiagnosed diseases, by collecting and sharing standardized, high-quality clinical and laboratory data including genotyping, phenotyping, and documentation of environmental exposures; and 3) create an integrated and collaborative research community across multiple clinical sites and among laboratory and clinical investigators prepared to investigate the pathophysiology of these new and rare diseases and share this understanding to identify improved options for optimal patient management.

The major funders of the UDN are:
The NIH Common Fund, which is managed by the Office of the Director/Office of Strategic Coordination (OSC).

B. Cooperative Agreement Responsibilities

The administrative and funding instrument used for the UDN is the cooperative agreement, an "assistance" mechanism (rather than an "acquisition" mechanism), in which substantial NIH programmatic involvement with the awardees is anticipated during the performance of the activities. Under the cooperative agreement, the NIH purpose is to support and stimulate the recipients' activities by involvement in and otherwise working jointly with the award recipients in a partnership role; it is not to assume direction, prime responsibility, or a dominant role in the activities. Consistent with this concept, the dominant role and prime responsibility resides with the awardees for the project as a whole, although specific tasks and activities may be shared among the awardees and the NIH as defined below.

NIH staff have substantial programmatic involvement that is above and beyond the normal stewardship role as described below:

The NIH Project Scientist(s) have substantial scientific and programmatic involvement during the conduct of this activity through technical assistance, advice, and coordination. However, the role of NIH staff is to facilitate and not to direct the activities. It is anticipated that decisions in all activities are reached by consensus of the UDN and that NIH staff are given the opportunity to offer input to this process. The Project Scientist(s) will participate as members of the Steering Committee and will have one vote. The Project Scientist(s) have the following substantial involvement:

- Participating with the other Steering Committee members in addressing issues that arise with UDN planning, operation and analysis. The Project Scientist(s) assist and facilitate the group process and do not direct it.

- Serving as a liaison, helping to coordinate activities, including acting as a liaison to other NIH Institutes/Centers, and as an information resource for the awardees. The Project Scientist(s) also help coordinate the efforts of the UDN with other groups conducting similar efforts.
- Attending all Steering Committee meetings as a voting member and all working group meetings, assisting in developing operating guidelines, quality control procedures, and consistent policies for dealing with situations that require coordinated action. The Project Scientist(s) are responsible for working with the grantee(s) as needed to manage the logistic aspects of the resource.
- Reporting periodically on Network progress to the NIH UDN Working Group (a trans-NIH Common Fund working group made up of staff from multiple NIH Institutes and Centers) and through it to the NIH Common Fund and to the National Advisory Council of Human Genome Research Institute.
- Serving on subcommittees of the Steering Committee, and Working Groups as appropriate.
- Assisting awardees in the development, if needed, of policies for dealing with situations that require coordinated action.
- Providing advice in the management and technical performance of the award.
- Assisting in promoting the availability of the data and related resources developed in the course of this program to the scientific community at large.
- Participating in data analyses, interpretations, and, where warranted, co-authorship of the publication of results of studies conducted through the program.
- Other NIH UDN Working Group staff may assist the awardee as designated by the UDN NIH Program Official (The NIH official responsible for the programmatic, scientific, and/or technical aspects of the grant).

Collaborative Responsibilities:

Close interaction among the participating investigators is required, as well as significant involvement from the NIH, to develop and operate the UDN. Principal investigators participate in in-person Steering Committee meetings on a quarterly basis during the first year of Network operation and subsequently three times per year; during months in which there are not in-person meetings, there are monthly conference calls as needed to share information on data resources, methodologies, analytical tools, as well as data and preliminary results. Key co-investigators and pre- and post-doctoral trainees, especially those who are members of under-represented minority groups or those from different but related disciplines, are also eligible to attend these meetings.

All Awardees agree to work collaboratively to:

- Assist in refining a common approach to patients with undiagnosed diseases.
- Work collaboratively with other UDN investigators to provide for secure, accurate and timely data submission.
- Participate in presenting and publishing new processes and substantive findings.
- Participate in the governance of the UDN as a member of the Steering Committee.

- Interact with other relevant National Human Genome Research Institute (NHGRI) and NIH activities, as needed, to promote synergy and consistency among similar projects.

Additionally the Clinical Site Awardees agree to work collaboratively to:

- Participate in network-wide processes for patient selection and assignment to a specific Clinical Site for evaluation.
- Identify 10 previously unidentified diseases Network-wide per year in FY16 and FY17.
- Fulfill all principal investigator (PI) primary responsibilities laid out in RFA-RM-13-004.

Additionally the CC Awardee agrees to work collaboratively to:

- Share statistical experience and expertise across the UDN and provide advice on statistical methods design.
- Participate with the current NIH UDP investigators to refine and adapt current single center activities to the requirements of the Network.
- Fulfill all PI primary responsibilities laid out in RFA-RM-12-020.

Additionally the DNA SC Awardees agree to work collaboratively to:

- Fulfill all PI primary responsibilities laid out in RFA-RM-13-018.

Additionally the Model Organisms Screening Center Awardee agrees to work collaboratively to:

- Fulfill all PI primary responsibilities laid out in RFA-RM-14-016.

Additionally the Metabolomics Core Awardee agrees to work collaboratively to:

- Fulfill all PI primary responsibilities laid out in RFA-RM-15-001.

C. Steering Committee Policies

Guideline: A Steering Committee composed of PIs from all sites (including the CC, CSs (including the NIH-UDP), the other Core Laboratories (including the DNA SCs, Model Organisms Screening Center, Metabolomics Core, and Central Biorepository), and the NIH Project Scientist(s) will be responsible for the scientific direction of the Network, as set forth in the FOAs RFA-RM-12-020, RFA-RM-13-004, RFA-RM-13-18, RFA-RM-14-016, and RFA-RM-15-001. The Steering Committee is responsible for the scientific direction of the Network.

Policies:

- The Steering Committee is responsible for policy decisions regarding the Network, and for the discussion and resolution of procedural issues that affect the operation and status of the network as a whole.
- The UDN Steering Committee will be the operational group through which the NIH UDN Working Group interacts with the UDN
- The Steering Committee will have monthly conference calls.
- The Steering Committee will meet in person quarterly during the first year and three times per year or as needed subsequently.
- The minutes for all Steering Committee discussion will be documented and posted on a CC website (viewable to Steering Committee members).

- The voting members of the UDN Steering Committee include the Principal Investigator(s) of each CS, the PI (s) of the CC, the PI (s) of each Core Laboratory (including the DNA SCs, Model Organisms Screening Center, Metabolomics Core, and Central Biorepository), and the collective NIH IC Project Scientists. Each site has one vote (multiple PIs may all be members of the Steering Committee, but collectively have one vote for their site) and the NIH Project Scientists group collectively has one vote.
- The Steering Committee may add additional members, and other government staff may attend the Steering Committee meetings as desired.

STEERING COMMITTEE
Co-Chairs: Euan Ashley, MD and William Gahl, MD, PhD

Members:
Clinical Site PIs (1 vote for each CS):
1. Baylor College of Medicine - Brendan Lee
2. Duke University (w/ Columbia)- David Goldstein and Vandana Shashi (contact)
3. Harvard Teaching Hospitals - Joseph Loscalzo
4. NIH - William Gahl
5. Stanford Medicine - Euan Ashley (contact), Jon Bernstein, and Paul Fisher
6. UCLA - Katrina Dipple, Stanley Nelson, Christina Palmer, and Eric Vilain (contact)
7. Vanderbilt University Medical Center - John Newman and John Phillips III (contact)

Coordinating Center PIs (1 vote):
8. Harvard Medical School - Isaac Kohane, Alexa McCray, and Rachel Ramoni

Core PIs (1 vote for each core):
9. Baylor College of Medicine - Christine Eng
10. HudsonAlpha (w/ Illumina)- Howard Jacob
11. Baylor College of Medicine (w/University of Oregon) – Hugo Bellen
12. Battelle Pacific Northwest Laboratories (w/Oregon Health & Science University) – David Koeller and Thomas Metz (contact)
13. Vanderbilt University Medical Center – Joy Cogan (contact) and John Phillips III

NIH IC Project Scientists (1 collective vote):
14. Anastasia Wise

D. Election of UDN Steering Committee Co-Chairs

Guideline:
The position of Chairperson of the Steering Committee of the UDN will be filled by Co-chairs who serve overlapping terms. The first 2 Co-chairs will be selected by the NIH UDN Working Group. Subsequent Co-chairs will be selected by a vote of the UDN Steering Committee.

Principles:
1. The term of the position of Chair will be 1-2 years in duration.
2. The individual holding the position of Chair must be a current member of the UDN Steering Committee.

3. The Chair must be either the Principal Investigator of one of the CS or a Core Laboratory or the CC.

E. UDN Executive Committee

The UDN Executive Committee consists of the 2 Co-chairs of the UDN, the PIs of the Coordinating Center, and the NIH Project Scientists. The Executive Committee meets weekly to review and monitor UDN progress.

F. Other Network Committees

Guideline: The Steering Committee may establish working groups as needed to address particular issues, which will include representatives from the program and the NIH and possibly other experts. The UDN Steering Committee will have the overall responsibility of assessing and prioritizing the progress of the various working groups and other needed subcommittees of the working groups.

Working Group Governance:

- Any individual or group proposing a new UDN working group will present their idea to the UDN Steering Committee. A formal vote of the UDN Steering Committee is needed to create a new working group.
- Volunteers for chair or co-chairs of the new working group will be solicited when the new working group is proposed. A formal vote of the UDN Steering Committee is needed to confirm the chair or co-chairs.
- Co-chairs are not required for all working groups, but may be recommended by the UDN Steering Committee.
- Working group co-chairs may come from the same site.
- If there are no volunteers, or only one, the UDN Steering Committee may recommend a site or type of site that may be a good fit for the working group and one of the UDN Steering Committee co-chairs will solicit the site(s) for a recommended chair.
- Any UDN working group proposing to close will present their idea to the UDN Steering Committee. A formal vote of the UDN Steering Committee is needed to close a working group.

Active Committees:
Billing: Chairs: Katrina Dipple (UCLA) and Vandana Shashi (Duke)

Biosamples and Biorepository: Chairs: Jordan Orange (BCM CS), Ed Silverman (Harvard CS), and Joy Cogan (Vanderbilt)

Case Review Committee: Ashok Balasubramanyam (BCM CS) and Katrina Dipple (UCLA)

Clinical Protocols: Chairs: Cyndi Tifft (NIH UDP) and Katrina Dipple (UCLA)

UDN Utility and Utilization: Chairs: John Mulvihill (NIH Program) and Tina Hambuch (Illumina)

Genetic Counseling: Chair: Ingrid Holm (Harvard CC and CS), Allyn McConkie-Rosell (Duke), and Christina Palmer (UCLA)

Model Organisms: Hugo Bellen (BCM MOSC) and May Christine Malicdan (UDP)

Publications and Research: Chairs: Rizwan Hamid (Vanderbilt) and Vandana Shashi (Duke)

Sequencing: Chairs: Christine Eng (BCM Seq) and Howard Jacob (HudsonAlpha)

G. Implementing and Revising the UDN Manual of Operations

- Working groups have been established to develop chapters for the UDN Manual of Operations.
- Chapters of the Manual of Operations are ratified by the UDN Steering Committee.
- Working groups have the authority to make decisions regarding implementation of ratified chapters of the Manual of Operations that are assigned to the working group for implementation.
- If a working group cannot resolve an implementation decision internally, the UDN Steering Committee will be consulted.
- Working groups will consult with other relevant working groups on implementation decisions that involve multiple areas of expertise. A cross-working group liaison may be assigned to facilitate these interactions.
- All working groups will make their agendas and minutes available to other working groups.
- Working groups that would like to recommend: 1) a change to a ratified Manual of Operations chapter that affects network-wide operations, or 2) addition of a new chapter, should recommend the change to the UDN Steering Committee for ratification.
- Groups that would like to recommend a change to the UDN network-wide IRB protocol or consents should recommend the change to the UDN Executive Committee, who will determine the need for a Steering Committee vote.
- Ratified changes to the Manual of Operations will be submitted by the UDN working group recommending the change to the CC for the Manual of Operations to be updated.

II. UDN Collaborative Clinical Sites

The UDN is open to Collaborative Clinical Sites that agree to the criteria for participation described below.

Criteria for Participation in the UDN are:

1. Each participant will inform the UDN NIH PO and the UDN Steering Committee about his/her group's plans for a UDN Collaborative Clinical Site.
2. Each participant will specify the sequencing, laboratory, and clinical evaluation plans for his/her proposed Collaborative Clinical Site.
3. Each participant is expected to contribute significantly to the project, bringing his/her particular expertise to bear on accomplishing the goals of the UDN in a timely manner. Participation in the UDN should consist of more than submission of data to the UDN and should include substantial intellectual contributions to the Network.
4. Each participant will adhere to UDN data sharing and publications policies, guidelines and agreements.
5. Each participant will take part in group activities, including attending UDN Steering Committee meetings and working group calls and contributing to the products of these groups.
6. Each participant will agree that s/he will not disclose confidential information obtained from other members of the UDN.
7. Additional criteria may be added upon recommendations of the UDN Steering Committee, External Scientific Advisors, and the NIH UDN Working Group.

Affiliate Membership application process:

An investigator who is interested in applying to be a UDN Collaborative Clinical Site should complete the UDN Collaborative Clinical Site Application form and return it to the CC Staff for appropriate dissemination. Items that should be included in the application and that will be used to evaluate acceptance into the Network are:

1. A concise plan, including DNA sequencing, other laboratory, and clinical evaluation plans proposed and a rationale for how the proposed Collaborative Clinical Site addresses the goals of the UDN. (Maximum length 3 pages, font 11, single spacing)
2. Evidence that the proposed Collaborative Clinical Site's research has received appropriate Institutional Review Board (IRB) approvals and is consistent with participants' informed consent.
3. Evidence of funding to conduct the proposed research as a Collaborative Clinical Site.
4. An agreement to abide by the UDN Data Sharing and Use Agreement and data submission policies, along with all relevant UDN Policies and Procedures.
5. An agreement to participate fully in UDN activities, including attending UDN Steering Committee meetings and working group calls and contributing to the products of these groups.

Applications will be reviewed by the UDN Steering Committee, UDN program staff, and the UDN External Scientific Advisors to determine whether a Collaborative Clinical Site will be accepted into the Network.

Evaluation for Collaborative Clinical Site applications will include a determination that:

- The clinical evaluation plan is appropriate for the UDN;
- The applicant has sequence data available or funding available to sequence their patients; and
- The applicant has the requisite expertise to participate in the Network.

The participation of Collaborative Clinical Sites will be reviewed yearly by the UDN Steering Committee, UDN program staff, and the External Scientific. A limited number of Collaborative Clinical Sites may be approved and acceptance may be limited to one-year after which an assessment will be conducted for continuation. At this point in time Collaborative Clinical Site applications are not being accepted by the UDN.

III. Clinical Protocol

A. Introduction

This Clinical Protocol component of the Manual of Operations "provides preliminary protocols and operating guidelines that will define an initial framework for common approaches to patient selection, data collection, laboratory investigation, and diagnosis, and serve as a base for further refinement by UDN investigators." (From RFA-RM-12-020).

I. Background of the UDP (see Appendices 1-3 for additional information).

Delivery of medical care to patients with rare and yet-to-be described diseases can be fraught with repetitive, inconclusive efforts at diagnosis as patients and their families go from physician to physician in hopes of finding answers. The Office of Rare Diseases Research (ORDR) notes that 6% of individuals seeking their assistance have an undiagnosed disorder and as many as 15% remain in the undiagnosed category for at least 5 years as physicians labor to define cause and pathophysiology. To address these issues, the NIH UDP was established in May 2008, as a joint venture of the NIH ORDR, the National Human Genome Research Institute Intramural Research Program (NHGRI-IRP), and the NIH Clinical Research Center (CRC). The goals of the UDP are to:

1. Provide answers for patients with undiagnosed diseases;
2. Generate new knowledge about disease mechanisms;
3. Assess the application of new approaches to phenotyping and the use of genomic technologies;
4. Identify potential therapeutic targets, if possible.

II. UDN Clinical Protocols Working Group:

The UDN Clinical Protocols Working Group developed this Clinical Protocol as part of the Manual of Operations and with input from the UDN Steering Committee will continue to refine it. The Working Group currently consists of the members listed below. Should there be a need to vote on matters within the working group, each site, the NIH Program, and the CC, will cast a single vote, for a total of 9 votes. Co-chairs of the Working Group are: Cyndi Tifft (NIH UDP) and Katrina Dipple (UCLA).

- CC: Ingrid A. Holm, MD, MPH (primary representative); Catherine Brownstein, PhD, MPH; Rachel Ramoni, DMD, ScD; Beth Rayworth; Kim Splinter, MS
- CSs:
  - Baylor College of Medicine: Carlos Bacino, MD (primary representative); Ashok Balasubramanyam, MD; Paolo Moretti, MD
  - Duke Medical Center: David Goldstein, PhD; Vandana Shashi, MD, MB BS (primary representative); Young-Hui Jiang, MD, PhD; Kelly Schoch, MS; Rebecca Spillman, MS
  - Harvard Medical School (Brigham and Women's Hospital, Boston Children's Hospital, Massachusetts General Hospital): David Sweetser, MD, PhD (primary representative); Ed Silverman, MD, PhD; Richard Maas, MD, PhD; Joan Stoler, MD; Calum MacRae, MD, PhD; Meredith Hanna; Wen-Hann Tan, MDNIH UDP: Cyndi Tifft, MD, PhD (primary representative); David Adams, MD, PhD; Bill Gahl, MD, PhD; Camillo Toro, MD
  - Stanford Medical Center: Jon Bernstein, MD, PhD (primary representative); Matthew Wheeler, MD, PhD

- University of California, Los Angeles Medical Center: Katrina Dipple, MD, PhD (primary representative); Stan Nelson, MD; Christina Palmer, PhD; Eric Vilain, MD, PhD
- Vanderbilt Medical Center: John Newman, MD (primary representative); John Phillips, MD; Rizwan Hamid, MD, PhD; Amy Robertson, MD

B. Detailed UDN Clinical Protocol

I. Study Design

In this study, individuals with undiagnosed diseases, and their family members when applicable, will be investigated. Applicants will apply to the UDN through a secure website managed by the CC, called the Gateway, and will be assigned to a CS based on an assignment algorithm. The CS will collect and review the applicant's medical records and will make a recommendation to accept or reject the applicant. Final approval to accept will be given by the UDN Case Review Committee (see Appendix 4: Case Review Committee of the UDN). Accepted applicants will typically be evaluated at the CS to which they were assigned; however, applicants may be reassigned to a different site based on presenting problems and the expertise of the site. Enrolled individuals will undergo a comprehensive medical and family history, physical examination, laboratory testing, imaging studies, consultations, and biological specimen collection, typically over the course of up to a five-day evaluation. Follow-up visits may occur if indicated. (See Appendix 5: ClinicalTrials.gov Record for a publicly available summary of the protocol.)

II. Triaging and accepting applicants into the UDN

1. Types of referrals:
    a. Applicant initiated: applicants (or their legal guardians) may learn about the UDN from a variety of sources, including the UDN website, publicity, or from another patient.
    b. Healthcare provider initiated:
        i. Healthcare providers not associated with the UDN may learn of the UDN from sources including the UDN website, publicity, colleagues, or medical conferences or publications.
        ii. Healthcare providers from CSs may refer their own patients for evaluation.
2. UDN application:
    a. Individuals (or their legal guardians) will register and apply to the UDN through the Gateway managed by the CC.
        i. The website will include:
            1. Information about the UDN and the application process
            2. A link to the Gateway
        ii. The Gateway will include:
            1. A disclosure statement – in order to be considered for participation in the UDN, individuals (or their legal guardians) will be required to either electronically sign or verbally agree to a disclosure statement allowing the UDN to store the applicant data that will be used to: a) assign the applicant to a CS for review, and b) collect characteristics of people who apply to the UDN. If an applicant (or legal guardian) does not speak English, a translator will be used to facilitate the verbal consent process.

iii. Once the applicant (or legal guardian) provides consent, either the applicant (or legal guardian), their referring provider, of a CC representative will enter the following data into the Gateway, which will be stored in the UDN database:
    1. Applicant's name, date of birth, gender, self-described race and ethnicity (for children <18 years the names of their parents will be required).
    2. Applicant's mailing address, contact information (email address, phone number). For children the addresses and contact information for both parents will be required. If parents are divorced or separated, they must provide information regarding who is legally permitted to sign a consent for medical research on behalf of the child. If parents are separated or divorced they must also both be willing to: (1) provide family history information, and (2) submit DNA samples for genomic analysis. If an adult applicant is unable to consent, the name and contact information of the individual with legal power of attorney who is able to consent on the applicant's behalf must be provided.
    3. Evaluation history
    4. UDN site preference
    5. Travel limitations
    6. Referring provider's name and contact information (mailing address, email address, phone number, fax number).
    7. Applicant's chief complaint, identification of the system most involved (i.e. cardiac, gastrointestinal), and symptom onset.
    8. Environmental exposures (this information does not refer to the Environmental Exposures Questionnaire, which is administered after acceptance)

iv. Either the applicant (or legal guardian), their referring provider, or a CC representative will also upload to the Gateway a referral letter (see Appendix 6: Example Referral Letters) summarizing the following:
    1. Pertinent medical problems
    2. Prior diagnoses
    3. History of evaluations and tests
    4. Medications
    5. Family history
    6. Review of systems
    7. Physician's diagnostic impressions

v. If an applicant (or legal guardian) does not speak English, a translator will be used to facilitate the application process.

vi. If an applicant does not have access to the Internet, a paper application can be requested through the CC. Completed paper applications will be mailed to the CC for data entry and CS assignment.

vii. Applicants (or their legal guardians) will be instructed to refrain from sending additional information, including any records, until assigned to a CS. If an applicant (or legal guardian) or their referring provider sends materials to the CC, it will hold them until the applicant is assigned a CS, at which time the materials will be sent to the CS.

3. Applicant triaging:
    a. The application will be assigned to a CS for triage to determine if the applicant is appropriate for acceptance into the UDN at the assigned site. The assignment to a site will be based on an assignment algorithm that takes into account the individual's

location, pertinent medical problem(s), site workload and site preference. If a given site is the closest and/or best suited to see a given subject but has already reached enrollment quota, the case would likely be assigned to a different site.

b. If an application is deemed inaccurate or incomplete by a CS (example: referral letter not written by a healthcare provider), the CS will contact the CC. The CC will reach out to applicants accordingly.
c. The CS will make initial contact with the applicant within 30 days of application submission.
d. The CS will gather the information needed to make a decision regarding acceptance (see Appendix 7: Suggested Triage Methods). Typically this will involve collection of medical records from the applicant. If the applicant (or legal guardian) has difficulty obtaining the medical records (including imaging and pathology materials), the site may also contact the referring provider for more information.
e. The CS will review the applicant's records and referral letter and make a recommendation regarding acceptance into the UDN. (See Appendix 8: Applicant Review Form.)
f. The UDN Case Review Committee (see Appendix 4: Case Review Committee of the UDN) will meet on a regular basis (weekly or biweekly) to:
    i. Finalize decisions (at least initially) for all cases at the CSs that have been recommended for acceptance.
    ii. Assign an applicant to a different CS if it feels that the applicant may be more appropriate for another CS based on expertise.
    iii. Review challenging cases

4. Guidelines for applicant selection:

Since few individuals can be accepted into the UDN each year due to limited resources, preference will be given to applicants for whom there is the greatest potential to provide a diagnosis or generate new knowledge about disease mechanisms.

a. More likely to be accepted:
    i. The applicant does not have a diagnosis that explains the objective findings.
    ii. The applicant (or legal guardian) agrees to the storage and sharing of information and biomaterials in an identified fashion amongst the UDN sites, and in a de-identified fashion to research sites beyond the network.
b. Less likely to be accepted:
    i. The applicant has a diagnosis that explains the objective findings.
    ii. Review of the records suggests a diagnosis and further evaluation is deemed unnecessary.
    iii. The applicant is too seriously ill to travel safely to the CS.
c. Preference may be given to individuals with one or more of the following characteristics:
    i. Novel clinical findings
    ii. Previous evaluations that have been non-diagnostic
    iii. A genetic diagnosis that has a poorly defined phenotype and no molecular mechanism
    iv. Multiple family members affected
    v. An objective laboratory or imaging clue to pursue
    vi. If a genetic origin is considered, biological parents are available to obtain blood for DNA sequencing (these families will be the most informative for gene discovery)
    vii. The individual is a member of an under-represented minority group

5. Application outcomes:
    a. Applicant and site appropriate for acceptance

b. Applicant appropriate for acceptance but reassigned to a different site
c. Applicant requires further testing or evaluation and may be reconsidered following receipt of the results
d. Applicant not appropriate for UDN
    i. A diagnosis was identified based on the review.
    ii. A potentially beneficial referral was identified based on review. An evaluation at the UDN may not be necessary to make a diagnosis.
    iii. The applicant is more appropriate for an expert site outside of UDN such as:
        - A research protocol at the NIH or elsewhere.
        - An expert at an academic medical center or elsewhere.
    iv. The applicant is not appropriate for the UDN and no alternative can be identified.

In all cases the referring provider and the applicant will be informed of the decision, generally within 60 days after receipt of all the medical records. The CSs will send out the disposition letters whether they are accept or not accept. (See Appendix 9: UDN Generic Letters.) When individuals are not accepted into the UDN, their application information will be stored securely and indefinitely in the database managed by the CC.

III. Sequencing prior to the clinical visit (optional)

In some cases, it may be useful to have the results of genetic testing (whole-exome/whole-genome sequencing (WES/WGS)) from the enrolled proband and relevant family members prior to the clinical evaluation. Genetic testing will only be performed on individuals accepted into the UDN. The sequencing will be done at the Baylor College of Medicine and/or HudsonAlpha. The decisions regarding timing of sequencing and WES versus WGS will be made on a case-by-case basis as clinically indicated, and left to the discretion of the CS responsible for the evaluation.

If review of the proband's presenting medical problem(s) and medical records suggest that performing sequencing prior to the clinical evaluation would be beneficial and aid in diagnosis, the CS will follow the following protocol:

1. Informed consent: Informed consent will be obtained over the phone or by videoconferencing (or in person if reasonable) from the enrolled proband, parent, or guardian. The consent form will be sent (by mail or email) to the proband (or legal guardian) prior to the remote or in-person consent. The PI, an associate investigator, a genetic counselor, or a project coordinator trained in consenting will be available to answer questions and obtain consent. Consent will be obtained at this time for the entire study, including for 1) obtaining blood for DNA extraction and sequencing, 2) any research studies performed as part of the evaluation, 3) obtaining other samples (blood, urine, etc.) during the evaluation for research, and 4) the collection of all of the clinical and research data by the UDN for research use. The signed consent forms will be sent to the CS responsible for the clinical evaluation and uploaded to the Gateway managed by the CC. The CS will also record the consent version signed by the proband and proband's preferences in the Gateway managed by the CC. Genetic counseling will also be provided by a physician or genetic counselor to all probands during the consent process. This genetic counseling will include a discussion of the types of results an individual may or may not receive (including primary, secondary, and incidental findings), the likelihood of receiving these types of results, individual preferences for types of results returned, and limitations of the genetic testing including false negative and false positive results. Probands will be given the choice of learning secondary and incidental findings related to conditions with treatment or management options. . "Secondary findings" are findings that the laboratory will look specifically for. "Incidental findings" are findings

discovered by chance during the genetic testing process. Genetic counseling aids were developed to supplement this session.

2. Assent: Assent will be obtained over the phone (or in person if reasonable) in the presence of a parent or guardian for all children ages 7-17 years old who are not decisionally impaired. Assent will be given for the entire study, including assent for 1) obtaining blood for DNA extraction and sequencing, 2) any research studies performed as part of the evaluation, 3) obtaining other samples (blood, urine, etc.) during the evaluation for research, and 4) the collection of all of the clinical and research data by the UDN for research use. The signed assent form will be sent to the CS responsible for the clinical evaluation and uploaded to the Gateway managed by the CC. The CS will also record the assent version signed by the proband and in the Gateway managed by the CC.

3. Family history: Once a signed consent form has been received from the proband (or legal guardian), a family history will be obtained over the phone (or in person if reasonable) to identify other family members of interest related to the proband's phenotype.

4. Enrollment of family members: Family members will be recruited through the proband, i.e., the researchers will ask the proband (and/or legal guardian) for permission to contact the family members. Priority will be given to those family members who would be most informative for sequencing analysis. Informed consent will be obtained over the phone or by videoconferencing (or in person if reasonable) from interested family members. The consent form will be sent (by mail or email) to the family member prior to the remote or in-person consent. The principal investigator, an associate investigator, a genetic counselor, or a project coordinator trained in consenting will obtain consent and be available to answer questions. Consent will be obtained for the collection of blood for DNA extraction and sequencing and for the collection of all of the clinical and research data and pertinent lab specimens for research use. The signed consent forms will be sent to the CS responsible for the clinical evaluation and uploaded to the Gateway managed by the CC. The CS will also record the consent version signed by the family member and family member's preferences in the Gateway managed by the CC. Genetic counseling will also be provided by a physician or genetic counselor to all family members. This genetic counseling will include a discussion of the types of results an individual may or may not receive, the likelihood of receiving these types of results, individual preferences for results returned, and limitations of the genetic testing including false negative and false positive results. A search for secondary findings will not routinely be performed in family members. However, incidental findings may be discovered by chance during the genetic testing process. During the consent, family members will be given the choice of learning incidental findings if they are identified. Genetic counseling aids were developed to supplement this session.

5. Collection of blood for DNA extraction: Once a signed consent or assent form has been received from the proband and family member(s), a kit will be sent to the proband and family member(s) for obtaining DNA. The kit will include:
    a. Tubes for blood collection
    b. An order form
    c. Directions for payment by the CS (direct costs for the collection and shipping of samples will be covered by the CS)
    d. An addressed shipping container for the blood to be sent back to the CS.

    It is expected that the blood collection will be completed with the assistance of the proband's local healthcare provider or local laboratory.

6. DNA extraction and sequencing: For probands selected for pre-admission sequencing, DNA will be extracted at the CSs and sent to one of the SCs, where the sequencing will be performed and analyzed prior to the evaluation in order to have the analysis available by the time the plan for the proband's admission is finalized. Significant variants will be identified using standard programs for assessing pathogenicity, Mendelian segregation patterns, allele frequencies, and databases of benign variants. Through this process, secondary and incidental findings may be identified. Findings intended to be reported to probands or participating family members for use in clinical decision making will be confirmed by Sanger sequencing in a Clinical Laboratory Improvement Amendments (CLIA) certified laboratory.

7. Return of sequencing results: If possible, results of genetic testing will be disclosed to the proband (or legal guardian) or family member during an in-person genetic counseling session at the CS. However, since some participants may have to travel a significant distance to get to the CS, results of genetic testing may need to be disclosed to the participant (or legal guardian) in a genetic counseling session over the phone. This may occur when blood is drawn for sequencing as part of the clinical evaluation or when the participation of a non-local family member is limited to a blood sample for DNA analysis and an incidental finding has been identified.

   CLIA-certified results of genetic testing will be provided to probands (or legal guardians) during a genetic counseling session with a qualified physician or genetic counselor. Genetic results related to the indication for testing will be returned to all probands (or legal guardians) and their referring providers.

   If a primary finding is identified, it will be listed on the proband's genetic testing report. If other family members undergo genetic testing, the report may include information about inheritance and other family members that carry the primary finding. A parent of a proband may therefore learn information about his/her own genetic status or the status of his/her relatives when receiving his/her child's results. If this report is shared with relatives, they may also learn information relevant to their own genetic results. These results will be discussed during a genetic counseling session with a qualified physician or genetic counselor.

   The SCs will report secondary findings, i.e., variants that are medically actionable in the genes recommended for such reporting by the American College of Medical Genetics and Genomics. In addition, both cores will report other secondary findings beyond the currently recommended 56 genes, provided these additional findings meet the threshold of having a defined medical treatment or specific management guidelines for disease surveillance. Stringent criteria for interpretation of variants in these medically actionable genes will be applied; reported secondary variants will either be previously reported as pathogenic or expected to be pathogenic based on the usual molecular mechanism associated with the gene. As a further measure to ensure consistency between the SCs, the SCs will communicate and reach consensus on the reporting of each secondary variant that they propose to report.

   During the pre-test genetic counseling/informed consent session, probands (or legal guardians) will be given the option of receiving secondary and incidental genetic results that are unrelated to the indication for testing, including results related to: (1) medical conditions with treatment or management options and (2) carrier status (only applicable for adult probands). The proband's genetic testing results will be shared with family members only if the proband (or legal guardian) provides permission.

Analyses to identify secondary and incidental findings will not be performed on family members. However, incidental findings may be discovered by chance during the sequencing process. During the pre-test genetic counseling/informed consent session, family members will be given the choice of learning incidental findings if they are identified. If a secondary or incidental finding is discovered in a proband and a family member is interested in learning if he/she also has this finding, the family member will be referred to a clinical genetics program for genetic counseling and testing. As this counseling and testing would not be done as part of the UDN, cost associated with this clinical follow-up would be billed to insurance.

There will be cases where the results of the sequencing performed prior to the clinical evaluation point to a likely diagnosis and in these cases, the CSs will be strongly encouraged to continue with the complete evaluation of the individual. The evaluation would allow the site to collect phenotypic data about the condition, provide counseling, and make suggestions about management. Exceptions would be cases where the diagnosed condition is common enough that established management standards exist and the presenting phenotype is a typical presentation of that disorder. If the clinical presentation varies from the typical clinical presentation of a well-recognized disorder, then phenotyping of the proband would still be appropriate.

IV. Planning the evaluation

Once a proband has been enrolled into the UDN and assigned to a CS, the site will work with the proband (or legal guardian) and the local team to create a plan to maximize the efficiency of the evaluation.

1. Information gathered by the CS prior to the evaluation (some of this information may have already been collected as part of the applicant selection process):
    a. Recent medical records (including consultation reports)
    b. Previous tests and results
    c. Pathology data/slides
    d. Imaging/radiography results
    e. Review of the medical history and review of systems
    f. Family history (including name, age, and contact information of family members)
    g. Medication list (including doses, schedule)
    h. Contact information for the proband's relevant physicians
    i. Environmental assessment
    j. Nutritional assessment
    k. Proband needs for travel and admission (including ventilators, mobility issues, etc.).
    l. Optional: administer surveys and perform interviews
2. The CS will create a plan for evaluation, that will include:
    a. Determining the lead physician.
    b. Determining clinical tests, procedures, consults, or research studies to be performed, including a determination of whether additional IRB approvals will be required.
    c. Scheduling dates for the proband evaluation – typically evaluations will occur over several days (expected to be five sequential days in most cases).
    d. Determining the sequence and schedule of tests, procedures, and consults
    e. Arranging a "sedation day" if indicated (especially important for children to maximize the efficiency and minimize the number of times a child needs to be sedated for a procedure or test).
    f. Arranging travel based on medical needs.

V. Evaluation

1. Schedule recommendations:
    a. Day 1:
        i. Informed consent (see Section III.1 Informed consent above for details)– if the proband (or legal guardian) has not already provided consent (see Section III.1 Informed consent above), consent will be obtained for: 1) drawing blood for DNA extraction and sequencing, 2) any research studies performed as part of the evaluation, 3) obtaining other samples (blood, urine, etc.) during the evaluation for research, and 4) the collection of clinical and research data. If not already provided, assent will be obtained for all non-decisionally impaired children 7-17 years of age. Following the consent, the CS will record the consent version signed by the proband and proband's preferences in the database managed by the CC. Probands may also be consented for other research projects at this time.
        ii. Initial visit with the primary care team including the lead physician:
            1. Review the medical history, review the family history, and perform a physical examination.
            2. Genetic counseling may occur if results of genetic testing are available.
            3. Surveys and interviews for research may be administered.
            4. The goals of the visit and schedule will be reviewed. Changes in the schedule based on this initial visit will be made.
    b. Days 1-5: All tests, procedures, and consultations will take place. Clinical investigations during the evaluation may include: laboratory testing, imaging studies, and biological specimen collection. Genetic counseling may occur if results of genetic testing are available. Specialized research studies, such as proteomics, metabolomics, and functional studies, may also be performed to elucidate underlying mechanisms of disease. Surveys and interviews for research may also be administered and consultations/counseling sessions may be recorded (if proband and/or family members give permission). Surveys and interview guides will be submitted to the IRB for review prior to their use. For individuals who did not undergo genetic testing prior to the evaluation, if it is determined during the visit that genetic testing is clinically indicated, blood will be drawn, DNA will be extracted and sent to a Sequencing Core for sequencing.
    c. Day 5: The team will meet with the proband and family to summarize the evaluation and make plans for follow-up.
2. Clinical diagnostic studies: Clinical diagnostic studies will be performed as clinically indicated and within the standards of accepted medical practice.
3. Specialized research studies: Specialized research studies may be performed as deemed relevant.
4. Biological specimens:
    a. Clinical specimens: Clinical specimens will be collected as medically indicated and at the discretion of the CS where the proband is being evaluated. Recommendations for certain clinical tests to be sent to specific facilities are presented in Appendix 10: Suggested Sites for Testing.
    b. Research specimens: Recommendations for research specimen collection are presented in the Biospecimens section of this document (see section XI).
5. Environmental studies: Environmental data will primarily be collected for clinical purposes through the use of a comprehensive questionnaire derived from the PhenX toolkit and National Health and Nutrition Examination Survey (NHANES) survey questions. The

environmental survey will be completed on line for each proband. In addition, the following may be obtained:

a. Food Frequency Questionnaire (FFQ)
b. Medications
c. Assessment of environmental exposures: Questionnaire, saliva or specific tissue collection for methylome analysis

VI. Unanticipated non-genetic medical information

During the course of this study, it is possible that unexpected medical information will be discovered that is important to the proband's health care. This information will be provided to the proband's health care provider. At the time, the proband (or legal guardian) will be given the option of learning this information and referrals will be provided as needed.

VII. Change in clinical stability

If, during the course of the UDN evaluation, the proband has a significant change in clinical stability requiring escalation of care or initiation of new treatments not covered by the research protocol, the proband may be offered completion of the UDN protocol at a later date. Probands and their caregivers, as well as referring providers, will be apprised of this change of condition necessitating active treatment rather than research-based investigation. If the proband's condition does not allow discharge from the CS at the scheduled completion date, care will be assumed either by the referring provider or appropriate clinical team members at the CS. Payment for further acute care will be provided by the patient's insurance company.

VIII. Terminating subject participation

During the UDN study, if a participant (or legal guardian) does not comply with study procedures or does not follow instructions given by UDN investigators, the participant's involvement in the protocol may be terminated.

IX. Clinical evaluation wrap-up

1. At the conclusion of the evaluation, and prior to discharge, the lead physician and other members of the care team as appropriate will meet with the proband and family to:
    a. Summarize the results of the clinical evaluation (clinical and research tests performed, procedures performed, consultations provided, results of testing received, and pending test results)
    b. Provide genetic counseling as indicated
    c. Make recommendations for follow-up with the medical home team
    d. Provide clear instructions about how to contact UDN team members if additional questions or concerns arise.
    e. Answer any questions the proband or family may have.

    This wrap-up will be facilitated using a structured wrap-up form (see Appendix 11: Wrap-up Template).
2. The wrap-up form and a short letter highlighting the key findings and follow-up recommendations will accompany the transfer of records to the referring healthcare provider and other providers designated by the proband or family.
3. The wrap-up form and a narrative summary of the evaluation will be uploaded to the Gateway.
4. All consultation and laboratory study reports pending at the time of discharge will be included in a revised wrap-up report sent to the proband, referring provider, and any other

care providers the proband has designated. If additional revisions occur, updated and revised wrap-up reports will be issued.

X. Return Visits to the UDN Site

Follow-up visits to the CS are not generally expected but may occur under at least two circumstances: (1) the CS requests additional phenotyping of the proband or family members to clarify or inform "affected" status or further interrogate candidate genes, and (2) a diagnosis has been made and the family returns for delivery of results.

C. Post-evaluation Activities and Follow-up

I. Transitions of Care

1. Background

Transitions of care programs are designed to promote the safe and timely passage of patients between levels of health care and across care settings. In the context of patient management, suboptimal transitions of care may result in: readmissions, adverse drug events, use of higher-intensity setting of care, decreased functional status, reduced quality of life, unnecessary repetition of tests or procedures, avoidable costs, and/or additional stress on patients, families, and caregivers. Suboptimal transitions of care are a risk for patients undergoing diagnostic evaluations performed by the UDN due to the potential for poor communication between providers; inadequate patient, family, and/or caregiver understanding of findings and follow-up needs of the patient; incomplete diagnostic evaluations; and a lack of clear understanding of which results are returned and which are pending. The purpose of the transitions of care plan within the UDN is to avoid these outcomes.

2. Best practices

Best practices at the CSs will include:

1. Providing a written summary of the diagnostic work up to the family upon departure (see section VIII. Clinical evaluation wrap-up)
2. Confirming that probands have made it home safely (via text message, email, or phone call).
3. Being available to families and caregivers in case any clinical issues arise.
4. Maintaining open lines of communication

3. Obtaining participant feedback

The CC and/or other UDN investigators will remain in contact with patients and families after discharge from the CS, which may include contacting patients shortly after the visit to assess satisfaction with the UDN visit and understanding of recommendations, as well as contacting patients periodically to assess clinical and research status. Any survey instruments and/or interview guides created will be vetted by the CC and Genetic Counseling and Testing Working Group and appropriate IRB approval will be obtained before initiating these activities. (See Appendix 12: Patient Follow-up Surveys).

4. Patient, Family, Caregiver Advisors

The CC plans to bring together a group of advisors as a network-wide resource for the UDN. Patients, family members, and caregivers are often extremely knowledgeable and can offer unique perspectives and valuable feedback. For example, these individuals can be involved in the development of patient-oriented discharge materials and advising on how best to partner with advocacy groups. The CC will be the conduit for requests for input from this group so that these volunteers will not be over-burdened and will be adequately supported for their effort. The CC will attempt to have the group members represent different genders, ages, incomes, geographic locations, CSs, and types of conditions.

II. Transitions to basic science

In most cases, the transition of patient data and/or sample to basic science will occur through the CS to which the patient was assigned, or through cross-site UDN collaborations. We expect that there will be referrals to the basic science community based on candidate genes or other diagnostic information, and it will be important to track these activities. A research follow-up plan will be developed by the CC (See Appendix 13: Research Inventory Form for an example). The goals of the plan will be to keep track of all research activities that the patient, samples, and data were involved in. In cases where there are no leads to pursue, the case could be reassessed again at two years after evaluation in order to determine if a research plan is indicated.

References- Post-evaluation Activities and Follow-up

1. Rennke, Stephanie, et al. "Hospital-Initiated Transitional Care Interventions as a Patient Safety Strategy: A Systematic Review." *Annals of Internal Medicine.* 2013; 158(5 Part 2): 433-440.

2. Centers for Medicare and Medicaid Services. http://cms.gov. Accessed April 15, 2014.

3. Garvey KC, Wolpert HA, Rhodes ET, Laffel LM, Kleinman K, Beste MG, Wolfsdorf JI, Finkelstein JA. Health care transition in patients with type 1 diabetes: young adult experiences and relationship to glycemic control. *Diabetes Care.* 2012 Aug;35(8):1716-22. 22.

IV. Data Standards

A. Background

The success of the UDN depends on the collection and subsequent sharing of well-described proband data. In order for the UDN data to be comparable and maximally useful, information about probands and their families must be captured in a uniform way. Several well-established standards have already been adopted by the undiagnosed and rare diseases community. These will be adopted by the UDN and are described in the first part of this section. The second part of the section describes the standard processes and data that are needed in order to track probands' progress through the UDN.

B. Data Standards for Demographic, Phenotypic, and Genotypic Characteristics

Demographics

Term	*Description/Notes*	*Likely Source*	*Data Format*
Address and zip code		CC Gateway	Text (with address autocomplete option)
Date of birth		CC Gateway	Date
Gender	Male, female, other	CC Gateway	Structured data
Race	Select CDC race categories (American Indian or Alaska Native; Asian; Black or African American; Native Hawaiian or Other Pacific Islander; White)	CC Gateway	Structured data
Ethnicity	Select CDC ethnicity categories (Not of Hispanic, Latino, or Spanish Origin; Mexican, Mexican American, or Chicano; Puerto Rican; Cuban; South American; Central American; Spaniard; Latin American; Dominican)	CC Gateway	Structured data

Phenotyping

Term	*Description/Notes*	*Likely Source*	*Data Format*
Human Phenotype Ontology Terms	See below for specific details about this standard	CSs	List of structured records with HPO ID and associated metadata (onset, etc) for each term

The Human Phenotype Ontology (HPO) is a resource for connecting genomic data with disease data, and provides links to diseases listed in OMIM and other disease databases [1].

The consistent annotation of UDN data with HPO terms will allow identification of probands who share the same or similar disease phenotypes across all CSs and, ultimately, more broadly with other large-scale efforts, including phenotype comparisons across model organisms [2]. Phenotypic data for UDN participants will be collected using the PhenoTips tool [3], which has fully integrated the HPO.

The HPO defines and organizes thousands of terms and relationships that characterize phenotypic variation and is regularly updated in response to requests from the research community, including the NIH UDP. The ontology has three sub-ontologies that cover 1) the mode of inheritance, 2) the onset and clinical course of the disease, and 3) phenotypic abnormalities, describing a wide-range of abnormalities across all body systems. HPO is cross-referenced to the Unified Medical Language System, the Medical Subject Headings, and other terminologies, including Orphanet's signs and symptoms.

Family History and Pedigree

Term	*Description/Notes*	*Likely Source*	*Data Format*
Pedigree	Family and individual unique IDs, gender, 'affection' (unknown, unaffected, affected) status	CSs	Text file (PED file)
Parental ethnicities	Include ethnicities of all four grandparents in pedigree. If Native American, ask tribal affiliation.	CSs	Text (with address autocomplete option)
Health conditions found in family		CSs	Text

Other Data useful for Research and Clinical Evaluation

Term	*Description/Notes*	*Likely Source*	*Data Format*
Photographs, videos, medical reports	Photographs to aid dysmorphology and other assessments	CSs	Folder system or binary db object
Interpreted Variants	Selected, highly-annotated variants of potential interest for disease causation	CSs	Text
Environmental Exposures Survey	Specific data gathering instrument	CC Gateway	Electronic survey
Timing of evaluation	Date evaluation started/ended	CSs	Date
Master clinician		CSs	Text
Specialists consulted		CSs	Structured data
Symptom onset	Prenatal/neonatal period, childhood, adulthood (>18 years)	CSs	Structured data

Prenatal/perinatal history (only for probands with prenatal/neonatal or childhood onset)	Maternal age at birth, paternal age at birth, maternal pregnancy history (G/P/SAB/Stillbirth), multiple gestation, assisted reproduction (conception after fertility medication, IVF (ICSI, typical fertilization), gestational surrogacy, donor (sperm, egg), prenatal genetic test/screen and results, prenatal movements (normal, decreased), gestation at delivery, delivery mode (NSVD, c-section secondary to), APGAR scores (one minute, five minutes) Exposures could be imported from Environmental Exposures Questionnaire (ex. medications, alcohol, tobacco, substance use, chemicals, xrays, hyperthermia, illness)- if yes, specify the exposure, timing of exposure, dose (if applicable), and notes	CSs	Structured data, text
Adverse events	Use cancer AE schema – Common Terminology Criteria for Adverse Events (http://evs.nci.nih.gov/ftp1/CTCAE/CTCAE_4.03_2010-06-14_QuickReference_8.5x11.pdf), related, date reported to NHGRI	CSs	Text

Genetic Data

Term	*Description/Notes*	*Likely Source*	*Data Format*
FASTQ Files	Unaligned; two per sample (read 1 and read 2)	SCs	FASTQ [text]
BAM Files	Aligned to GRCh37 (hg19)	SCs	BAM [binary]
VCF Files	Annotation standards have not been established yet. Proposed (incorporated) metadata to include ancestry, race, relationship to proband, ID of proband.	SCs	VCF [text]
Coverage metrics		SCs	CSV [comma separated values]
Description of analysis pipeline	Names and versions of all pipeline components (software, algorithms, platforms, etc.)	SCs	TBD (likely XML)

Date sequence data uploaded to UDN database and date data entered into dbGaP		SCs, CC	Date
Candidate genes/variants		SCs and CSs	Text
Clinical sequencing report		CSs	PDF
Results communication	Primary care provider, proband (or legal guardian) and date	CSs	Structured data and date
Segregation studies		CSs	Structured data
Functional study information	Laboratory name, laboratory address, candidate gene, allele, variant, modification, species, cell type, phenotyping protocol, genetic background (inbred, outbred, etc), phenotyping data, peer review/publication	CSs	Structured data, text

Minimum standards for genetic data will be consistent with National Center for Biotechnology Information (NCBI)'s dbGaP requirements (http://www.ncbi.nlm.nih.gov/gap). This includes family sequence variant files in .VCF format, BAM files associated with each .VCF file, and PED files. (Ethnicity and race data and phenotype data in HPO format will also be submitted to dbGaP.) The Sequencing section of this manual provides further detail about standards for WES and WGS.

Sequencing sample data

Term	*Description/Notes*	*Likely Source*	*Data Format*
Prior sequencing	Whole exome, whole genome, N/A	CSs	Structured data
Laboratory name, address, CLIA number where DNA extracted		CSs	Text
Type of sequencing	Whole exome, whole genome, targeted variant sequencing	CSs	Structured data
Test requested	Proband only, duo, trio, quad, other	CSs	Structured data
Date DNA extracted		CSs	Date
Method of extraction		CSs	Text
DNA quantity and quality		CSs	Text
Accession number	Sample identifier at CLIA lab performing extraction	CSs	Text

Relative information	relationship to proband, affection status	CSs	Structured data
Date DNA sent to and received by SC		CSs and SCs	Date
Tracking number		CSs	Text
Sample barcode(s)		CSs	Text

Biosample Data

Term	*Description/Notes*	*Likely Source*	*Data Format*
Biosample IDs and locations	UDN ID, sample type, if obtained in fasting state, number of tubes collected, date and time of collection, date and time of processing, number of tubes aliquots made and stored, shipping tracking number, issues with sample collection	CSs	Structured data to allow searching

C. Standards for Tracking the Progress of Probands through the UDN

When first applying to the UDN, prospective probands will be assigned a UDN UUID that will be used to track them throughout their involvement in the UDN.

Upon acceptance, UDN UUIDs will also be assigned to participating family members. Process and workflow data about both prospective and enrolled probands will be tracked and recorded in a series of structured documents, as shown below.

Application

Term	*Description/Notes*	*Likely Source*	*Data Format*
Date of Application, name of patient, birthdate, gender, address, phone number, email address, referring physician, name and address of primary contact		CC Gateway	As needed per item
Date of onset of primary condition		CC Gateway	Date
Healthcare provider referral		CC Gateway	Uploaded file (PDF, doc, etc)

letter			

Initial assignment to CS

Term	*Description/Notes*	*Likely Source*	*Data Format*
Name of CS, date sent to site by CC		CC Gateway	As needed per item
Date reviewed by CS		CSs	Date
Date selected to UDN Case Review Committee		CC Gateway	Date

Application Review

Term	*Description/Notes*	*Likely Source*	*Data Format*
Name of CS, date assigned		CC Gateway	As needed per item
Name of responsible investigator		CSs	Name
Narrative summary on applicant's condition	Provide a narrative summary (150-200 words) on the applicant's condition. If applicable, include: history of present symptoms, date symptoms first noted, past medical history, previous diagnoses/comorbidities (using ICD terms if possible), prior procedures and surgeries.	CSs	Text
Prior pertinent evaluations	Please indicate the applicant's pertinent prior evaluations. If applicable, please include: prior positive or negative test results and prior genetic testing (especially sequencing).	CSs	Text
Provisional diagnosis/working plan	ICD or SNOMED or OMIM. The NIH Office of Rare Diseases maintains a list of rare disease names (http://rarediseases.info.nih.gov/gard/) that may also be useful for those diagnoses that are not already available in other established	CSs	Text

	terminologies.		
Category of primary condition		CSs	Structured data
Number of family members affected		CSs	Structured data
Family members available for analysis	All/some/none/unknown	CSs	Structured data
Date accepted		CC Gateway	Date
Date not accepted		CC Gateway	Date
Reason not accepted	Not Accepted with Recommendations - Specific testing; Not Accepted with Recommendations - Seek expert care; Not Accepted - Diagnosis identified; Not Accepted - UDN would likely not be able to find a diagnosis; Not Accepted. Insufficient records made available to UDN site	CSs	Structured data

Consent

Term	*Description/Notes*	*Likely Source*	*Data Format*
Date participant (or legal guardian) provided consent and assent		CSs	Date
Remote or in-person consent and assent	Record if consent/assent occurred remotely or in-person; if remotely-record the date the consent/assent form was received by the CS	CSs	Structured data
Consent/assent form version		CSs	Structured data
Signed informed consent document		CSs	Scanned PDF
Preferences	Probands: Photographs of face and body, video/voice recordings, skin biopsies, return of secondary/incidental genetic results (medical conditions that have treatment or management options for all probands and carrier status for adult probands) Family members: voice recordings, return of incidental genetic results	CSs	Structured data

	discovered by chance during the testing process		

Enrolled Proband Data

Term	*Description/Notes*	*Likely Source*	*Data Format*
Cost of stay	Best estimate of real cost of all clinical work done for proband. Some debate about this—may need further discussion.	CSs	Unknown
Costs billed to insurance		CSs	Unknown
Follow up after discharge	Includes provider information; Discuss in context of transitions of care. May be specific survey tools, etc. involved in this process; Captured in follow-up surveys designed by CC	CSs	TBA
Complications after discharge if applicable	Captured in follow-up surveys designed by CC	CSs	Text
Withdrawn	Date and reason for withdrawal	CSs	Structured data
Death	Date, cause, circumstances if happened during UDN visit, date reported to IRB	CSs	Structured data

References - Data Standards

1. Köhler S et al. The Human Phenotype Ontology project: linking molecular biology and disease through phenotype data. Nucl. Acids Res. (1 January 2014) 42 (D1): D966-D974. http://www.human-phenotype-ontology.org/.

2. Mungall CJ, Gkoutos GV, Smith CL, Haendel MA, Lewis SE, Ashburner M. Integrating phenotype ontologies across multiple species. Genome Biol. 2010 Jan 8;11(1):R2.

3. Girdea M, Dumitriu S, Fiume M, Bowdin S, Boycott KM, Chénier S, Chitayat D, Faghfoury H, Meyn MS, Ray PN, So J, Stavropoulos DJ, Brudno M. PhenoTips: patient phenotyping software for clinical and research use. Hum Mutat. 2013 Aug;34(8):1057-65. http://phenotips.org/.

V. Technology and Data Management

A. Privacy, Security Quality, and Compliance

In order to provide prompt and effective data management across a geographically diverse and highly specialized network, it is clear that the UDN data network, systems, and applications will need to store, manage, and protect personally-identifiable information (PII) and personal health information (PHI). This necessitates that primary engineering, policies, and procedures are strongly driven and governed to ensure the necessary security and compliance.

I. 'Above the line' and 'Below the line' Technologies, Processes, and Systems.

'Above the Line' refers to all technologies, processes, and systems that are operated under the responsibility of the CC.

'Below the Line' refers to all technologies, processes and systems that are operated within each CS and Core. It is fully understood that each CS has preexisting processes, systems and novel technology capabilities, and the CC does not dictate which systems or processes a given CS chooses to use as long as that decision does not impair or threaten the overall security and compliance posture of the UDN data and technology network.

A few examples of 'Below the Line' systems include:

1. EMR (Electronic Medical Record) systems
2. LIMS (Laboratory Information Management Systems), such as UDPICS from the NIH UDP
3. Local document and record management systems
4. Local bio-bank and clinical laboratory systems

II. Security Controls at the CC

1. *Physical Controls:* For physical records (paper, photographs, pen drives etc.), the CC and the CSs will employ appropriate physical access controls (e.g., locked cabinet in a locked room).
2. *Computer systems Controls.* Electronic security will consist of multiple levels of protections.
    a. For computer systems containing personally identified information (PII) and personal health information (PHI), security controls that are compliant with HIPAA, National Institute of Standards Technology (NIST) guidelines, and the Federal Information Security Management Act (FISMA) will be utilized and a proper Federal Information Processing Standard Publication (FIPS) 199 assessment will be performed prior to commissioning of these systems.
    b. For computer systems that contain PII but not PHI, appropriate roles-based access and security controls will be used and a 3rd-party security assessment will be performed and documented.
    c. For computer systems that contain neither PII nor PHI, these systems will also employ roles-based controls and will conform to the information security and compliance standards of Harvard University.

III. UDN Technology Security and Compliance Policies

As proper security privacy and compliance can only be accomplished via an integrated approach of people, policies, processes, and technologies, a comprehensive approach is required. This approach will minimally require:

a. Selection of regulatory standards, strategy and compliance approach
b. Information asset and data security training
c. Uniform roles-based access strategies
d. Technology monitoring and logging
e. Independent verification of procedural and technology controls

IV. Technology and Compliance Services and Coordination

The UDN will provide advice and access to information security and privacy expertise via a set of pre-qualified partners and internal resources that can help with security and privacy assessments, vendor qualification, and procedural document control.

V. Auditing

All key 'Above the Line' technology will be assessed via a risk-based approach to determine security, privacy, and compliance requirements. For systems containing high-sensitivity data and technologies (i.e., the Gateway), will be wrapped into FISMA Moderate compliant structures. 'Below the Line' components and technologies will need to take precautions such as having up-to-date virus scanners, disk encryption, workstation-level authentication and lockouts on all machines accessing Above-the-Line systems.

IV. Technology Standards

Technology Standards will be essential to enable automated communications and rapid transmittal of data as well as for the essential elements of technology resilience, security and privacy. An ongoing set of technology standards will be developed, managed and governed by a standards oversight committee or working group

B. UDN Data Flow Process

Since the UDN will operate as a real-time knowledge network, it is essential that appropriate and essential data flow securely and privately through each required step.

Step 1. Patient application and initial response. The process is initiated when the patient (or her/his representative) applies to the UDN through the secure UDN Gateway. The patient provides contact, medical, and demographic information and is assigned a UUID and a UDN ID. The UUID ensures that there is a universally unique ID. The UDN ID is a simplified ID that humans can use in communication with each other. Once registered, the patient receives a confirmation of application as well as an initial referral to a CS. This referral, based primarily upon geography, may be automated, but may involve some input from a medical manager to better distribute patients to the various clinical sites. As this step involves the capture of PII and, possibly, PHI, the portal is built and administered in a manner that is compliant with HIPAA and FISMA regulations. Any physical data or correspondence that accompanies the application process for the UDN will be managed with compliant physical document management controls.

Step 2. Referral and data transfer to a CS. This step is initiated when the patient is contacted by the CS and assembles her/his medical records for admission review at that center. The patient's medical records include current and past reports, laboratory studies, radiographic studies, etc. These data will be stored securely and indefinitely under the oversight and policies of the individual CSs. The records, in this form, will not be stored on the Gateway.

Step 3. Evaluation. All data obtained, created, or managed during the inpatient or outpatient evaluation will be the responsibility of the CSs and will be managed in such a way that ensures the security and privacy according to the guidelines of each institution. These data will serve as the permanent record and be subject to appropriate records retention policies. All UDN-wide agreed-upon "Above-the-Line" data elements will be collected and entered into the UDN Gateway.

Step 4. Post visit reporting and review. Agreed upon UDN-wide "Above-the-Line" data elements from the CS evaluation will be transmitted to the CC via secure electronic transfer and will be archived in a FISMA-compliant repository. These data will be structured data and will be kept in the Gateway's database (as opposed to just uploading scans of records). An API enabling easier input will be made available. A separate copy of the data/documents, most likely representing a subset of the data including copies of test and procedure results, will be transferred to the home care team.

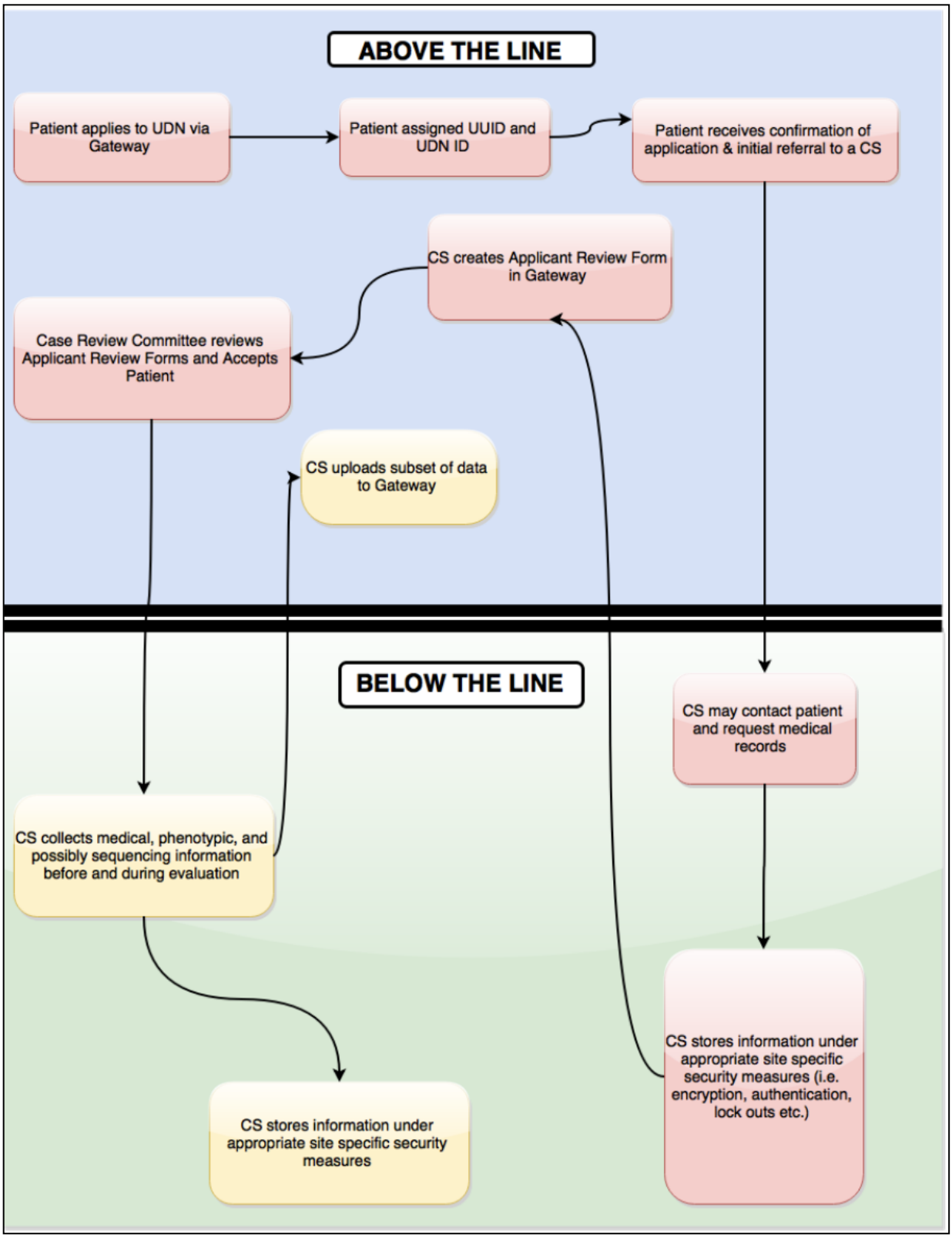


Figure 1. UDN Data Flow Process

C. Sequencing Data

The SCs will ensure the privacy and security of all PII and any PHI collected or generated during the sequencing process. The resulting genetic data will be transferred securely to the UDN Gateway, which will serve as the center of record for these data. The nature and management of the sequencing data provided by the SCs is discussed in detail in the Sequencing section (see Section VI: Sequencing) of this manual.

D. UDN Feature Request Process

The UDN Gateway will evolve over the course of life of the UDN. A major part of this evolution will be driven by requests for additional features. This feature request process described here applies to all UDN Gateway feature requests, both large and small. The CC will begin accepting feature requests upon the public launch of the UDN Gateway.

Definitions:

1. Feature requester: Individual or group who is making the request for the feature.
2. CC project manager: Individual at the CC who communicates with the Site Coordinators, SCs, and CC team members to complete project related activities.
3. CC technology team: Team at the CC that produces and manages the UDN Gateway.

Feature request process:

1. Feature requester completes the feature request form (see Appendix 14: Feature Request Form) and sends the request to the CC project manager.
2. The CC project manager logs the feature request in the CC queue.
3. The CC technology team assesses the feasibility of the request from a technical and compliance standpoint. This may necessitate asking for additional information from the requester.
    a. If the request is infeasible on a technical or compliance basis, the CC project manager will convey this information to the feature requester and will remove the feature request from the queue.
4. The CC technology team assigns an approximate time to complete the feature request.
5. The feature request is preliminarily prioritized by the CC, after which the Executive Committee vets the prioritization for presentation to and approval by the Steering Committee.
6. The CC technology team executes the feature requests in order of priority.

Note: If a feature requester is able to provide funding for additional programming and support resources, the requested feature may be able to be addressed more quickly. To determine the resources required, the feature requester should speak with the CC technology team. “Showstopper”/Critical bugs will always jump the queue and be priority. These sorts of issues aren’t classified as “features” and have a different handling process. They will come through the UDN Help Desk and be issued to the technology team for immediate resolution.

VI. Sequencing

A. Flow of samples to SC

Sample Collection and DNA Extraction

1. CSs arrange for blood sample collection before or during the clinical evaluation. If collected off-site, blood samples for DNA should be shipped to the CS.
2. The CS arranges DNA extraction and quality control (QC). DNA samples submitted for sequencing should meet the following conditions:
    a. WES: at least 6ug of 50-200ng/ul DNA
    b. WGS: at least 10ug of 50-200ng/ul DNA
3. Additional DNA is stored at the CS with other biospecimens collected during the clinical evaluation.

Shipping After Determination of Exome or Genome Sequencing

1. CS prepares DNA samples for shipment to the appropriate SC. DNA samples should be sent as complete families (including all family members that will be included in the analysis) excepting clinically urgent samples that warrant prioritized sequencing. Urgency is at the discretion of the CS.
2. CS completes a sequencing request form in the Gateway for each DNA sample being sent for sequencing.
3. CS enters and releases updated phenotype information (patient application review and PhenoTips) in the Gateway for use by the SC in their analyses.
4. CS enters shipping information (date DNA sent and tracking number) in the Gateway and ships samples.
    a. Please note that the Gateway provides alerts for shipping of UDN samples, but shipment tracking needs to occur at the CS/SC level.
5. Gateway sends an automated email to alert the appropriate SC of sample shipment and available phenotypic data.
6. SC acknowledges receipt of samples by entering date DNA received in the Gateway.
7. If a submitted DNA sample does not pass QC at the SC or is otherwise deemed unacceptable, the SC will contact the CS site directly via phone or email to request a replacement.
8. Sample labeling discrepancies will be addressed on a case-by-case basis at the discretion of each SC.

B. Flow of clinical information to SC

1. CSs will organize the collection of blood specimens and DNA extraction entirely below the line. The CC and SCs will not know about or track the DNA specimens until they are shipped to the SCs. The CSs are encouraged to collect all specimens for a family before sending them, but additional family members may be added at a later date if necessary.
2. Typically, a CS will send an aliquot of DNA extracted in a CLIA-certified lab and keep the remainder of the DNA for future procedures, developed by the Biorepository working Group.
3. Samples could be sent either before or after the in-person evaluation of the study proband. In either case clinical information and a pedigree (including the relationships of all submitted family members to the proband) should be added to the PhenoTips

instance on the Gateway as soon as possible for samples submitted for sequencing. This information will be used by the SCs for their analysis.

4. DNA samples submitted for sequencing must be labeled with patient name, date of birth, and the initials “UDN”.
5. Other required information for sample submission includes:
    a. Gateway consent form
    b. Gateway sequencing form
        1. Lab name, address, CLIA number where DNA Extracted
        2. Sequencing core
        3. Type of sequencing and rationale
        4. Test requested (proband only, duo, trio, quad, other)
        5. Date DNA extracted
        6. Date DNA sent to sequencing core
        7. Tracking number
        8. Method of extraction
        9. DNA quantity
        10. DNA quality
    c. Affection status of family members

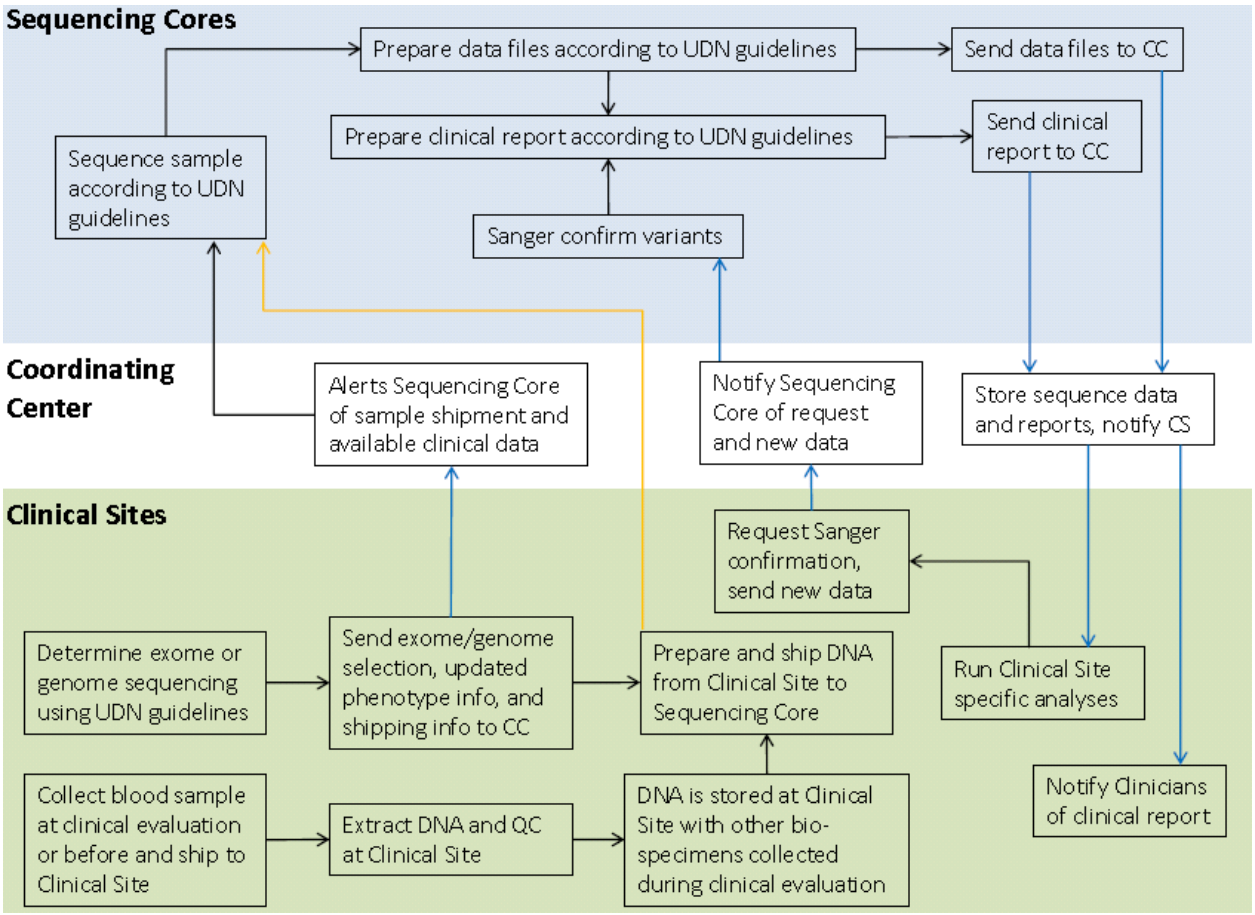


Figure 2. Sequencing Flow Diagram

C. Exome-Genome Sequencing

WES - Baylor College of Medicine (BCM)

This section covers sample intake, library preparation, whole exome capture, and sequencing at Baylor College of Medicine. This section will describe the sample flow from DNA sample receipt to production of WES data, including appropriate quality control and assurance procedures.

Sample Intake

DNA samples are received at the Whole Genome Laboratory (WGL). A visual inspection of the sample tubes is conducted. Sample tube label is compared with information entered in the Gateway to ensure consistency and completeness of the Gateway data, consent forms, proper sample labeling, and sample tube integrity. Samples will be accepted if no discrepancies are found, sample labels match, and no tube damage is observed. If any of the above criteria is not met, Baylor will notify the referring CS.

Once accepted, samples are accessioned into the WGL Laboratory Information Management System (LIMS) system. Sample information in the Gateway is entered into the WGL LIMS database. Each sample is assigned an internal six-digit lab number, as well as a six-digit family number in LIMS. 1D bar code labels with patient specific information (unique identifier) including patient name, DOB, lab number and family number are attached to the stock DNA tube. Subsequently samples are aliquoted from the stock tubes into 2D barcode tubes. The samples in 2D bar code tubes will be processed for exome sequencing. Before sample transfer, the record for a sample is first opened in LIMS, then the 1D barcode label on the stock tube of the sample is scanned and the LIMS automatically verifies if the sample ID in LIMS matches that on the label. Then, the 2D bar code on the aliquot tube is also scanned to link the two bar codes in LIMS before sample transferring occurs. These steps are to ensure the chain of custody remains intact during sample transfer.

Sample QC

DNA samples are then screened to quantify DNA as well as determine DNA quality. To determine DNA concentration and purity, the samples are evaluated using the Quant-iT PicoGreen dsDNA assay on the BioTek Synergy 2 microplate reader.
Passing criteria include:

- ✓ The R-squared value for the standard curve must be ≥99.9%DNA concentration
- ✓ Sample contains 1ug of DNA

To verify DNA integrity and relative size, the same dilution of sample is loaded on a 0.8% E-Gel.
Passing criteria include:

- ✓ Gel image is clear and shows no DNA degradation

If a sample does not meet the criteria above, the CS is notified.

Pre-capture Library Preparation

In order to meet UDN sequencing objectives, we use our quick whole exome sequencing (QWES) protocol. QWES is an optimized version of the standard Illumina (ILM) library preparation workflow that reduces library construction time to 5-6 hours.

Library construction is a completely automated process on the Span-8 Biomek NXP with an incorporated LIMS tracking system. Before starting library preparation, all primers and adapters lots are validated and the appropriate dilutions are prepared. Negative (H_2O) and reagent blank controls are included Robot operator closely monitors each transfer step. Pre-capture library preparation involves the following steps:

Normalization and shearing

DNA samples are normalized to 750 ng total. Samples are loaded into Covaris microtubes in 50 ul aliquots and sheared to approximately 250-500 bp using the Covaris E220 ultrasonicator. Shearing efficiency is assessed using a 2.2% flash gel. Fragments should range from 100-600bp with average of 250-500bp. If the majority of sheared fragments is larger than 800 bp, the sample is re-sheared.

End repair

Fragmented DNA samples are treated with NEBNext® END REPAIR Module (catalog#: E6050L) at 20°C for 20 minutes to make blunt ended DNA. Then 1.8X Beckman SPRI beads (Agencourt AMPure XP Solid Phase Reversible Immobilization magnetic beads) and 70% Ethanol are used for cleanup. Treated fragmented samples are eluted with 40 ul elution buffer while SPRI beads remaining in the solution.

3' Adenylation

The treated DNA samples are incubated with NEBNext® dA-Tailing Module (catalog#: E6053L) at 37°C for 20 minutes to incorporate a non-templated dAMP on the 3' end of a DNA fragment. The binding buffer (BB) made with Polyethylene Glycol (PEG) and 5 M Sodium Chloride (NaCl) (final concentration of PEG and NaCl: 20% and 2.5 M respectively) is applied to help dA-Tailing fragment DNA binding back to SPRI beads while the rest of solution is discard. 70% ethanol then is used for cleaning up the DNA bounded SPRI beads. DNA samples again are eluted with 40 ul elution buffer while SPRI beads remaining in the solution.

Ligation

Post dA-Tailing, DNA Samples are ligated with Illumina multiplexing paired-end (PE) adapters by using Invitrogen Expresslink ligase (catalog#: A13726101) and buffer at room temperature for 20 minutes. The same binding buffer is used to allow ligated DNA binding back to SPRI beads and 70% ethanol is used for cleaning up the SPRI beads. SPRI beads are removed post 40 ul elution buffer added.

Enrichment

Ligated DNA samples are enriched for total 6 cycles with 2X KAPA HiFi HotStart Ready Mix PCR kit (catalog#: KK2612) and Illumina PE PCR primers. AB GeneAmp PCR System 9700/Veriti are used for amplification Enrichment.

Post-enrichment QC

The enrichment PCR efficiency analysis is performed on 2.2% FlashGel by checking the product intensity. The FlashGel analysis is preformed after 6 cycles and is re-run if the band is too weak. Additional PCR cycles can be added for samples with low yield.

- ✓ No more than 9 cycles total can be run for samples
- ✓ If the amount of the post-PCR product is insufficient after a total of 9 cycles, the whole process needs to be repeated.
- ✓ The negative (H_2O) and reagents blank control should give no product

To check size distribution and quantify the final library, the sample is run on an Agilent Bionalyzer 2100 DNA 7500 Chip.

- ✓ The library sizes should range from 200 – 750 bp (Majority are 250-550bp) with the peak ranging from 250 - 350 bp.
- ✓ The yield of library should be more than 1.5 ug. If pre-capture library yield is lower than 1ug, the library preparation is repeated. No adaptor dimer and free primers are visible. See Figure 3 for an example of a passing pre-capture library.

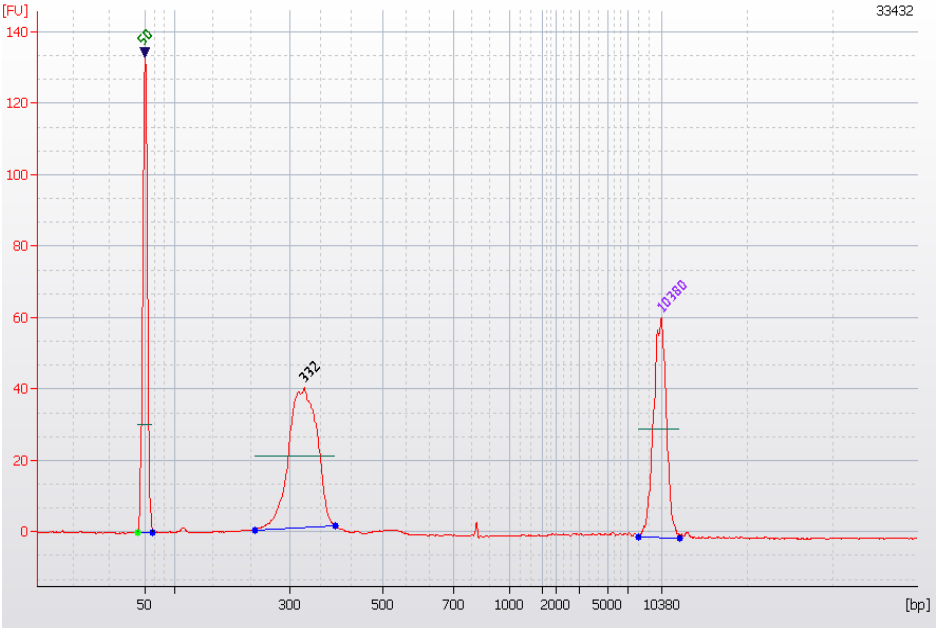


Figure 3: Illumina paired end DNA library on Agilent Bionalyzer 2100

Whole Exome Capture

The whole exome capture utilizes the NimbleGen liquid capture on HGSC VCRome 2.1, that targets approximately 34Mbp of genomic DNA including all coding exons of currently known disease genes (OMIM, HGMD, and GeneTests). To enhance the coverage of clinically relevant disease genes, the currently developed spike-in probe set (Exome 3 – PKV2) is used in 1:1.25 equimolar ratio with the VCRome exome capture design in combination with the QWES protocol.

Solution capture is initiated by combining 1.5 ug of the pre-capture library, 40ul of 1mg/ml human Cot1 DNA, and adding 0.65 ul of each 1,000uM Hybridization enhancing (HE) oligos. Full-length hybridization enhancing oligos are used to augment capture efficiency.

This mixture is dried down in a DNA vacuum concentrator on high heat setting and re-suspended in Hybridization buffer and Formamide. The mixture is denatured for 10 minutes and VCRome probe with Panel Killer V2 combined in 1:1.25 ratio are added. The mixture is incubated at 56°C for 16 hours. The following day, captured DNA is washed and recovered. Post-Capture PCR amplification is performed using KAPA HiFi HotStart DNA polymerase with total 12 cycles.

Final library QC

FlashGel

Capture efficiency is checked using a 2.2% FlashGel. If the intensity is too low, the capture process needs to be repeated. Only primer bands should be seen for negative and reagent blank controls

Bioanalyzer

To assess size distribution and quantify the final post-capture product, libraries are loaded onto DNA 7500 Chip for assessment on the Agilent 2100 Bioanalyzer. The majority size should be around 300-400 bp with the concentration above 20 nmol/L. See Figure 4 for an example of a passing final library.

qPCR

Capture efficiency evaluated by SYBR green-based qPCR with known four loci assays. Successfully enriched capture libraries have an average delta Ct of the four loci >6 with delta CT of the individual assay is >5.

Post-capture libraries must pass all three QC checks to proceed to cluster generation and sequencing. For each sequencing run, three individual barcoded libraries are normalized to 10nM and pooled into one pool.

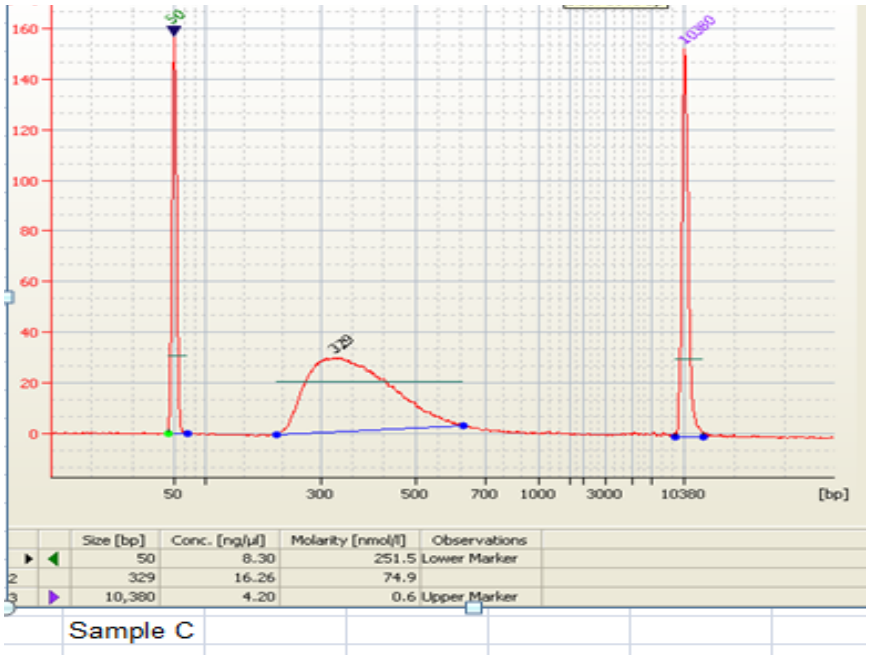


Figure 4: Example of Agilent Bioanalyzer result of final post-capture library

cBot Cluster Generation

Post-capture libraries are denatured prior to loading on the cBot. Denatured PhiX control is spiked-in into lanes 1 and 2 for HiSeq 2500 Rapid runs. The optimal library concentration for cluster generation is 10nM. Library and flow cell information are entered into WGL LIMS system prior to starting a cBot run.

After the cBot run is completed, each strip is checked to ensure that correct volumes were drawn for each lane. If volume of any reagent tube or DNA strip tube is not equal to the others (most frequently more volume is left), the reagent and/or DNA was not delivered properly for that lane.

Sequencing

The HiSeq 2500 is employed for sequencing in rapid mode (27 hr cycle time) to generate 100bp paired-end reads in a format of 3 samples per lane to generate 10-12 GB per sample. Target coverage for proband and parental samples is >100x. The WGS LIMS system is utilized to track the run set up, status and quality metrics. Pertinent metrics and passing thresholds are provided in the tables that follow.

The run will stop after imaging is complete for the first cycle and generate a first base report. The first report will be confirmed by manual review and the run will resume if all the metrics correlate with the determined standards.

G intensity	>6000
Cluster density	>400k/mm2

The performance of the run is monitored, and the metrics below are recorded to assess quality at a particular step of the sequencing run, evaluate library quality and concentration, detect any potential sequencing reagents and/or optical issues

Cluster density at cycle 5	900-1100 k/mm2
Phasing/pre-phasing at 25 cycles	<0.3/0.7%
Passing filter rate	>80%
PhiX error rate	<3%
Q30	>80%

After the run is complete, a comprehensive set of post-sequencing production metrics are continuously monitored and are reviewed at weekly meetings to facilitate timely troubleshooting to maintain overall pipeline performance. Overall run performance is evaluated by metrics from the off-instrument software (Casava) and from mapping results generated by the Mercury analysis pipelines using Burrows Wheeler Aligner (BWA) software to ensure that production standards are met

Pass filter	>80%
Aligned reads	>80%
Error rate	<4%
Unique reads	>90%

The capture analysis is incorporated in the Mercury analysis pipeline and provides metrics to gauge the overall quality of the capture process. This pipeline reports:

- Proportion of the aligned reads that map to the targeted region, which is relative to the effective enrichment of the capture
- Distribution of coverage across the targeted bases; specifically, the fraction of targeted bases covered at 1x; 10x, 20x, 40x

The complexity of the capture library is assessed by calculating the number of alignment reads that occur from PCR duplicates. If needed, these reads can be removed from the analysis.

Key metrics that have been developed and are reviewed in weekly meetings are presented in the table below

Reads Aligned to target	>50%
Target bases covered at >20x	>90%
Target bases covered at >40x	>80%
Mean coverage of target bases	>100x

As an additional quality control measure, samples are also analyzed by SNP array. SNP data is compared with WES data to ensure correct sample identification and to assess sequencing quality. The data is analyzed using an automated pipeline that produces concordance and contamination scores.

Concordance	>90%
Contamination	<5%

WGS – HudsonAlpha Institute for Biotechnology (HA)

This section covers sample intake, library preparation, and whole genome sequencing at HudsonAlpha Institute for Biotechnology. This section will describe the UDN sample flow from DNA sample receipt to production of WGS data, including appropriate quality control (QC) and quality assurance procedures.

DNA Samples are received by the Genomic Services Laboratory (GSL) at HudsonAlpha Institute for Biotechnology. A visual inspection of the samples and subsequent accessioning is performed by two GSL employees. Accessioning includes entering the samples into a project in the LIMS of the Clinical Services Lab (CSL) and assigning two identifiers to the samples. The first identifier is the CSL identifier and is formatted as a project number, submitter's initials, and a unique sample number incrementing up from sample 0001 (i.e. C1001-SL-0001). The second identifier is a unique identifier created by the LIMS using the date of accessioning and another digit, which indicates the order in which samples were accessioned. These identifiers deidentify the sample and are used to track the samples through all handling performed by the CSL. This accessioning is performed by the HAIB CSL laboratory manager or a designee.

If the submitted sample is DNA from a CLIA-certified laboratory, it proceeds to QC procedures as described below. If the submitted sample is blood, DNA is isolated in conjunction with its accessioning and another unique sample identifier for the DNA is created by adding a '.1" to the GSL ID (i.e. C1001-SL-0001.1). DNA is extracted from 1ml of whole blood on the QIAsymphony instrument using the Blood_1000_V7_DSP protocol. This protocol yields on average of 10-18ug of gDNA from 1 ml of whole blood.

Sample information in the Gateway is reviewed by the HAIB UDN Project Manager to ensure consistency and completeness of the Gateway sequencing and consent forms, proper sample labeling, and sample tube integrity. Coordination between the UDN HAIB Project Manager and the CSL laboratory supervisor ensure that sample labels are cross-checked with information in the Gateway. Sample tubes will be visually compared to information in the Gateway for accuracy prior to accessioning by the CSL laboratory supervisor or an assignee.

Samples will be accepted if no discrepancies are found, sample labels match, and no tube damage is observed. If any of the above criteria is not met, HudsonAlpha will notify the referring CS.

Sample QC

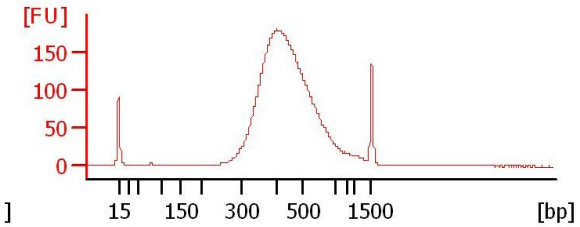
All DNA samples are evaluated for concentration by fluorometric assay (Qubit® or Picogreen®) and for integrity by agarose gel. Ideally, there will be at least 500ng of intact, high quality DNA available to enter the GSL whole genomic sequencing (WGS) library preparation. For fragmented samples or other non-traditional DNA preparations, such as low input samples, QC may be performed with the Agilent Bioanalyzer or Caliper GX. Samples not meeting the minimum requirements for clinical WGS may still proceed into library preparation, but as research samples, not clinical.

Library Preparation

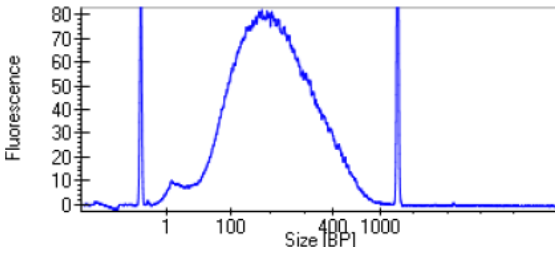
DNA samples are normalized to 1000ng of DNA in 50ul of water. All gDNA samples require fragmentation in a random manner to create the fragments that will become the inserts in the final library. The Covaris L-series and E-series instruments are used to shear the DNA to a final

insert size of ~350bp. This longer insert size improves overall library performance and allows the longer sequencing read lengths on the Illumina HiSeq X platform (150bp) to be efficiently used without producing a significant number of over-lapping reads. QC is performed after sonication to ensure that yield and fragment size are within expected ranges. Library preparation is then performed using a proprietary GSL methodology with key QC steps performed throughout. QC is performed during preparation, after ligation, to assess yield. After the library preparation is complete, final library yield, fragment size, and fragment distribution are measured. Finally, real-time PCR quantitation to determine the molar fraction of the sequenceable library is performed. Yields are determined with fluorescent measurements (PicoGreen) and fragment sizes and distribution will be determined with either the Agilent Bioanalyzer or the Caliper LabChip GX, depending on batch size. Example traces for sonicated gDNA and final sequencing library are below. The final libraries are diluted to 3 nM stocks for use in clustering and sequencing.

Post sonicated gDNA QC



Post library prep QC



Sequencing

Once a library passes QC, the production sequencing on the HiSeq X will be performed. Clustering and sequencing will be performed as per standard Illumina protocols for HiSeq X sequencing. Each UDN sample is sequenced by itself in one lane, plus also sequenced in a pool of 3 samples across a final lane. One lane of sequencing on the HiSeq X instruments will generate approximately 30X coverage of the human genome when duplicates removed from consideration. The UDN grant specifies a minimum of 40X coverage. Therefore, for a given trio of samples, a pool of the 3 samples is run across a fourth lane to supply an additional ~10X coverage per sample, yielding 40X total. Approximately 360 million paired-end reads, each 150 bp in length, will be generated for each sample, with typical flow-cell runs lasting ~3 days each. Over 105 Gb of sequence per sample is generated per lane and a 40X UDN WGS sample will receive a minimum of 150 Gb of data.

D. Analysis

Overview

- The steps in the analysis of WES or WGS data can generally be divided into 4 phases: primary analysis, secondary analysis, tertiary analysis, and interpretation. Secondary analysis can be further subdivided into read mapping and variant calling phases.
- Best-of-breed standards in analysis of WES or WGS sequence data will be followed (as defined in this manual and agreed to across all SCs, CSs, and the CC).
- Annual review of current methodologies, with an aim of identifying and potentially incorporating advances of note in analytical approaches supporting interpretation of sequencing data will be performed by the SCs.
    - Any alterations that are considered for inclusion will be shared with the Sequencing Working Group.
    - Those prioritized will be implemented leading to revision of the analysis steps outlined in this document.
- Each CS may conduct analyses on UDN cases sequence data as they see fit but the SCs will undertake primary, secondary, and tertiary analysis of the sequencing data.
    - The purpose of this is to provide consistency in the format and quality of the data provided and to create maximal utility for the widest range of consumers of these data.
    - For example, this method ensures that sites without existing clinically certified variant annotation and prioritization pipelines will have access to richly annotated data.
    - SCs will share sequencing data with CSs through the Gateway.

Sequencing Files in the UDN Gateway

Output files from various stages in the analysis process derived from various applications will be uploaded to the Gateway by the SCs. Files will also be stored locally according to clinical data retention policies in place at the SCs. As currently defined these are:

- Standard compliant format FastQ files
- Standard compliant BAM files
- Standard compliant VCF files
- Annotated variant files
    - A tab delimited text file format
    - SCs will work to ensure that the format and data encapsulated in this file is equivalent at both sites
- A spec sheet listing software versions and patches, analysis tools, and annotation repositories will be provided, along with exact parameters used in the analysis.
    - This will allow sites to unambiguously determine the exact steps for reproduction of analysis and, perhaps allow for case based additional optimization of analysis parameters at capable clinical sites. Format of this file TBD; one suggestion is an XML spec sheet for unambiguous representation and downstream automation.
    - In addition, where applicable (for example as the header of the generated VCF file), a human verifiable description of the applications, version, and reference datasets used will be encapsulated in the output files themselves.
- An interpreted clinical report will also be provided by the SCs.

    - The format of this report will follow the existing industry standards for clinical sequencing reports.
    - Clinical reports will include the following report sections:
        - Lab contact information and general test information
        - Patient name and date of birth
        - Indication for testing
        - Primary findings (pathogenic, likely pathogenic, and variants of unknown clinical significance) in tabular format
        - Secondary and incidental findings in tabular format
        - Interpretation of findings – textual discussion of the relevance of the findings given the clinical presentation of the proband
        - Specific recommendations
        - A description of the methods used
        - Limitations for both the sequencing technology and analytical processes
        - References
- Secondary findings will not be sought or reported for family members, however, incidental findings discovered by chance during the testing process may be returned to family members at the discretion of the SC and CS. Each family member will receive a report containing one of three possible results:
    - Incidental finding identified
    - No incidental finding identified
    - Family member opted out of receiving incidental findings

Primary Analysis

Primary analysis (demultiplexing) will be performed on the HiSeq instrument workstation according to Illumina guidelines. Software used for primary analysis is described in Table 1.

The primary analysis steps at each site will be equivalent, although they may have version differences reflecting the software update timetables in place at the SC. The primary analysis software version used will be listed in the spec sheet provided by the SCs. Changes and updates will be appropriately communicated to the CSs.

Secondary Analysis Background

It is important to note that the secondary analysis steps performed at the SC will not necessarily be identical across all steps. They will, however, be comparable and clinically appropriate as defined by their existing usage in the CLIA and College of American Pathologists (CAP) accredited clinical laboratories at both SCs.

The secondary analysis steps at each SC will be equivalent although they may have version differences reflecting the software update schedules in place at the SCs. Each SC will perform clinically appropriate validation of all datasets and algorithms/software applications in use within its clinically validated analysis pipelines. Significant pipeline component changes will undergo re-validation at the discretion of the SCs.

Secondary Analysis – Read Mapping

Secondary analysis step 1 (mapping and realignment) will be performed at each SC using the methods and tools described Table 2.

GRCh37/hg19 (b37d5) will serve as the alignment template until it is superseded and adopted by leaders in the field. Alignment of reads to GRCh37/hg19 (b37d5) will be performed without truncation of the data. More specifically, duplicates will be marked but not removed from the dataset.

Secondary analysis Step 2 (variant calling) will be performed at each SC using the methods and tools described Table 2.

Tertiary Analysis – Variant Annotation

It is important to note that the variant annotations produced by each SC will not necessarily be identical but they will be equivalent. The reason behind this is that to make them identical would require re-working the clinical pipelines in place at the SCs. This activity was not planned for or funded. The plan is to use the existing clinically validated pipelines at the two sites. The SCs will liaise closely to ensure equivalency and will continue to work towards a unilateral set of variant annotations. The specific variant annotations and the tools used to produce them are outlined in Tables 3 and 4. The SCs will, to the extent possible, ensure that the data is labeled the same in the datasets produced from each CS.

Coverage analysis

In addition to primary, secondary, and tertiary analysis, the SC will also provide a summary of the sequencing coverage for each sample. This report will detail the coverage from the sample run at the gene model, transcript, and exon levels. This will provide the CSs with an indication of regions of likely importance that are not well covered. In cases where a trio or an extended pedigree is submitted for sequencing this coverage report will be provided for both the proband and the other individuals in the pedigree. The CSs and CC will be appraised of any updates to the coverage algorithms made by the SCs.

BCM uses ExCID, an in-house developed tool, to assess capture efficiency and coverage at desired cutoffs. Files from this tool can be viewed in any genome browser to see the actual coverage on genes of interest. This will also report a summary of poorly covered regions.

HA assesses coverage with GapMine v 3.0.1, a software package developed the Medical College of Wisconsin for coverage at the gene, transcript, and exon level. Files can be reviewed in any genome browser. The coverage at these levels and for specific gene lists are provided as a report if requested. Coverage gaps at a defined depth threshold are also reported.

Interpretation

The SCs will also provide an interpreted clinical report. A systematic process will be followed in accordance with ACMG guidelines as published (https://www.acmg.net/docs/Standards_Guidelines_for_the_Interpretation_of_Sequence_Variants.pdf) to determine the clinical significance of each variant considered for reporting.

Analysis output delivery turn around times

Initial analysis (to end of tertiary analysis phase) will be completed within a 2 week turnaround time (TAT) at both SCs.

Preliminary clinical reports are typically available 7-8 weeks after raw data is uploaded to the Gateway. All variants included in clinical reports are confirmed by Sanger sequencing. CSs may request additional Sanger sequencing of variants identified during their analysis. Sanger sequencing is performed by the SCs. The SCs will Sanger confirm up to 8 variants per case. Unused Sanger confirmations may be distributed among each clinical site's case cohort (ex. 25 cases = up to 200 Sanger confirmed variants). The turnaround time for final clinical reports will depend on the timing of CS data analysis.

Requesting Release of Sequencing Data

Each SC will follow its existing institutional policy for fulfilling raw data requests. Raw data will only be released after a final report has been returned to the CS and test results have been communicated to the participant.

Baylor College of Medicine will release WES data to physicians and qualified researchers at the patient's request. See http://bmgl.com/media/wysiwyg/bmgl/pdfs/ExomeDataReleasePacketv6.pdf for more information.

HudsonAlpha will release WGS data to patients, physicians, and qualified researchers. Interested patients should contact the Clinical Services Laboratory directly at clinical@hudsonalpha.org or (256) 327-9413.

Table 1. Primary analysis tools used by each SC.

Phase	Step	BCM	HA
Demultiplexing	**Bcl2Fastq**	bcl2fastq-1.8.3	bcl2fastq v 2.15.0.4

Table 2. Analysis steps and applications/algorithms/platforms used for secondary analysis at each SC.

Phase	Step	BCM	HA
Read mapping	Alignment	bwa v0.6.2	BWA-mem v0.7.12
	Fixmate, Sort & Index	Picard v1.8.4	SAMbamba v0.5.4
	Mark duplicates	Picard v1.8.4	Picard v1.136
	Realignment and	GATK v2.5.2	GATK v3.3
Variant calling	SNV	Atlas2-SNP v1.4.3	GATK v3.3
	INDEL	Atlas2-Indel v1.4.3	GATK v3.3

Table 3. Tertiary analysis tools used by each SC.

Phase	Step	BCM	HA
Variant annotation	Annotation	Cassandra v15.4.29	CarpeNovo v 6.0.0
Variant prioritization	Trio analysis	Trio Afterburner v2	CarpeNovo v 6.0.0

Table 4. A depiction of the set of variant annotations and tools used by each SC.

Annotation	BCM	HA	Notes
Annovar	yes	no	commercial product
Alamut HT	no	no	commercial product
splice sites	yes	yes	
near splice site	yes	yes	BCM: +/- 5bp , HA +/- 6bp donor & 25bp acceptor
protein coding flag	yes	yes	
syn change flag	yes	yes	
non-syn change flag	yes	yes	
AA change	yes	yes	
Sift prediction	yes	yes	
Polyphen2 HVAR prediction	yes	yes	
Polyphen2 HDIV prediction	yes	yes	
Mutation Taster prediction	yes	yes	stand-alone
Condel prediction	no	no	BCM uses dbNSFP (PMID: 25552646)
MutationAssessor prediction	yes	yes	
AlignGVGD	no	no	web service; not in pipeline
stop gain flag	yes	yes	
stop loss flag	yes	yes	
start loss flag	no	yes	
frameshift flag insertion/deletion/indel	yes	yes	
non-frameshift flag insertion/deletion/indel	yes	yes	
location: intron/exon	yes	yes	
location: 5'UTR/3'UTR/Intergenic/Promotor	yes	yes	
HGNC appropriate Gene Symbol	yes	yes	RefSeq
Transcript ID	yes	yes	
COSMIC	yes	no	BCM: additional HA: scheduled for 2015 inclusion
HGMD ID	yes	yes	
HGMD variant level association	yes	yes	
HGMD gene level association	yes	yes	
OMIM ID	yes	yes	
OMIM variant level association	yes	yes	
OMIM gene level association	yes	yes	
ClinVar ID	yes	yes	
ClinVar metadata (various; to be clarified)	yes	yes	
dbSNP ID	yes	yes	
dbSNP AF	yes	yes	
1000 Genomes AF	yes	yes	
ESP EVS AF	yes	yes	
Mappability score	yes	yes	

VII. Data Sharing

The success of the UDN will depend on the collection and subsequent sharing of well-described data. This UDN Data Sharing Policy is consistent with the goals of the NIH Data Sharing Policy.[1] The NIH states "Data should be made as widely and freely available as possible while safeguarding the privacy of participants, and protecting confidential and proprietary data."[2]. Data from the UDN are expected to be handled so as to increase the value of the significant public investment in the creation and operation of the UDN. The CC is committed to best practices in data standardization and will develop efficient mechanisms for sharing and dissemination of the data generated by the UDN.

This document outlines the minimum requirements for sharing the data that are collected in the course of participation in the UDN. The document is organized as a set of questions and answers.

Q: What data will be shared within the UDN?
A: All clinical, biospecimen, and sequencing data that are generated by the UDN effort will be shared in a secure and compliant manner within the Network. These identified data will be referred to as "UDN data" throughout this document. UDN data include data generated both in human subjects' research and in laboratory research.

Q: Who will have access to the UDN data?
A: Any UDN investigator. UDN investigators who acquire UDN data must use the data responsibly and must monitor the use of the data by members of their laboratories. (See Appendix 15: UDN Data Sharing and Use Agreement.)

Q: Will UDN data be shared with investigators who are not currently part of the UDN?
A: Possibly, if there are complementary initiatives with goals that are consistent with the UDN, as for example, would be the case if the NIH awarded grants that are scientifically related to the work of the current UDN. Also for the diagnosis of individual probands, if there are useful experts outside the UDN, these can/should be involved on an as needed basis.

Q: Will UDN data be shared more broadly in public databases?
A: Yes, in de-identified form. Data resulting from UDN efforts will be deposited in dbGaP, which is maintained by the NCBI at the NIH. De-identified data may also be deposited in other public databases, registries, and repositories, such as PhenomeCentral, the NIH Global Rare Diseases Registry, and be shared with other existing or emerging rare and undiagnosed diseases research efforts.

Q: How will the rights of individual research subjects be protected?
A: Research participants will give consent to have their data shared, according to a UDN agreed upon informed consent process. Each subject in the database will be associated with a UUID that will be used as the primary identifier for all data associated with that participant. Role-based access and physical security controls that are fully aligned with the sensitivity of the data

[1] NIH Data Sharing Policy. http://grants.nih.gov/grants/policy/data_sharing/.
[2] Final NIH Statement on Sharing Research Data, February 26, 2003. http://grants.nih.gov/grants/guide/notice-files/NOT-OD-03-032.html.

at each point of use and access will be employed. De-identified data shared outside of the UDN will not reveal individual identifying information, consistent with the HIPAA Privacy Rule[3].

Q: How will institution-specific intellectual property regulations and restrictions be addressed?
A: The CC will work with the principal investigators at each of the CSs and Cores to develop an approach that is consistent with the data sharing policy described in this document.

Q: What is the publication/authorship policy for UDN collaborative activities?
A: There is a separate Publications section within the manual that describes these policies. If broad data release is required as a condition of publication by the authors or the publisher, the Publications Working Group should be contacted as soon as possible prior to making any commitments to ensure that the data release is feasible.

Q: What is the commitment of each UDN investigator?
A: UDN investigators agree to:

1) Further the mission of the UDN: to create new knowledge regarding the biochemistry, physiology, and mechanisms of undiagnosed diseases and improve diagnostic and management options for patients afflicted with them.
2) Acknowledge that in pursuit of this mission, common UDN goals may supersede individual goals. Specifically, in the interest of rapid progress, UDN investigators commit to:
    a. Model a collaborative, open, interdisciplinary spirit, characterized by mutual trust and respect across disciplines, individuals, areas of expertise, institutions, and by demonstrating interest and engagement beyond their own specific domains.
    b. Ensure that data generated at individual sites are comparable and additive by adhering to UDN data standards.
    c. Make data contributions to the UDN in a timely manner.

Q: What is the role of the CC?
A: To facilitate, monitor, and report on the effective and timely sharing of data within the UDN and beyond.

[3] HHS - Office for Civil Rights - HIPAA. http://www.hhs.gov/ocr/hipaa/

VIII. Publications and Research

One parameter of UDN success will be the number and quality of its publications and presentations. The purpose of this document is to establish a framework, which facilitates and streamlines collaborative manuscript submission, as well as antecedent work, like meeting abstracts and presentations. The UDN Publications and Research Working Group ("UDN Publications and Research Committee") will oversee the activities set out herein on behalf of the UDN Steering Committee, and report to it. Changes to the policy described herein, which are expected from time to time, must be approved by the UDN Steering Committee. The UDN Publication Policy applies to a proposed publication if the results are the product of research that the NIH UDN prime or sub-award funded.

A. Scope

I. To facilitate manuscript submission.

II. To provide input in abstract submission and scientific presentation (when requested).

III. To help the CC with content for the UDN website and, if required, social media.

IV. To maintain an up-to-date list of all UDN presentations, abstracts, publications and proposals. The CC will assist in tracking and coordinating projects.

V. Notwithstanding anything to the contrary in this document, the scope of UDN Publications Committee activity does not include evaluation of the scientific merit of any publication produced as a result of UDN participation.

B. Manuscript: Authorship Review and Submission

I. Authors (First, Middle and Senior) will be determined by the type, scope and site of project. First author will take primary responsibility for the manuscript. Given the nature of the UDN's work, shared first or last authors should be remembered as an option.

II. UDN will be acknowledged at the end of the author list, as "Members of UDN". The UDN member list would include PIs and Master Clinician of each site and the members of the UDN Publications and Research Committee. A UDN membership list will be provided by the UDN Publications and Research Committee and may be different on a case-to-case basis.

III. Generally it is expected that authors would make contributions to any or all of the following including but not limited to the concept, design, acquisition and analyses of data, drafting of manuscript, editing and revision of manuscript.

IV. All manuscripts will be reviewed and approved by the UDN Publications and Research Committee prior to submission to any journal (see Appendix 16: Publications and Research Reference Sheets for details). Approval shall be for purposes of satisfaction of the points in this section alone and, for purposes of clarity, this means that UDN Publications and Research Committee shall not withhold approval of a manuscript on the basis of its scientific merit.

V. The UDN Publications and Research Committee will resolve all authorship disagreements.

VI. All UDN papers (network-wide and local) should include a statement such as "Research reported in this manuscript was supported by the NIH Common Fund, through the Office of Strategic Coordination/Office of the NIH Director under Award Number(s) [xxxxx]. The content is solely the responsibility of the authors and does not necessarily represent the official views of the National Institutes of Health".

C. Manuscript Proposal−Submission and Approval (Concept sheet)

To initiate the process that will lead to a publication, all UDN investigators are invited to submit firm, mature concepts for papers to the Publication Committee. The proposed lead (first) Author submits a completed Manuscript Concept Sheet (see https://hms.az1.qualtrics.com/SE/?SID=SV_3CyZOKOuiEvnCx7), which is reviewed and approved by the Committee before substantial drafting begins. A voting member of the Steering Committee must endorse the concept proposal. If more than one person submits the same or similar topic, the Committee helps decide who will assume the project lead.

The Concept Sheet is submitted to the CC for administrative processing. The CC forwards the proposal to the UDN Publications and Research Committee for review. It is expected that the approval process will not take more than two weeks.

For Special Issue Journals, the journal organizer will submit a Concept Sheet for the entire Issue, listing proposed articles and lead authors along with Concept Sheets for each article. Once the main Concept Sheet and all abbreviated Concept Sheets have been submitted, the CC forwards the proposal as a Special Issue Journal packet to the UDN Publications and Research Committee for review.

D. Abstracts and Presentations:

I. An abstract submission will not require approval from the UDN Publications and Research Committee.

II. Abstracts and presentations should mention UDN as "Members of UDN" in the authors list as well as UDN grant number.

III. All presentations should be sent to the CC so that they may be posted to the UDN web site. All abstract citations should be sent to the CC so that the UDN bibliography can be updated accordingly.

IV. If there is a NHGRI or Common Fund co-author, final versions of the abstract must be submitted to the PO for review and approval.

E. Database of Publications and Concepts:

The responsibility of the UDN Publications and Research Committee will be to advise the CC in management and population of the database of concepts, presentations, abstracts and publications submitted, finished and in process.

F. Start of UDN publication:

The UDN Publications and Research Committee will develop a manuscript or multiple manuscripts, which will describe, define, and introduce the network to the medical and scientific community.

G. End of Funding Cycle Publications:

The Publication Committee will be responsible for stimulating the preparation of manuscripts, which describe the UDN experience towards the end of the first four-year funding cycle.

Concept Sheet:

The investigator submitting the concept sheet will have sole possession of the idea. Other groups can contribute but the original submitter of the sheet will take the lead. Please note that the concept sheet **has** an **expiration date** of 6 months but can be extended for an additional 6 months if needed upon submission of an updated concept sheet. (See Appendix 16: Publications and Research Reference Sheets and the following *Concept Sheet Link:* https://hms.az1.qualtrics.com/SE/?SID=SV_3CyZOKOuiEvnCx7.)

IX. Website and Social Media

The purpose of this section is to outline the UDN plan for the creation and maintenance of a unified public-facing UDN web and social media presence.

The UDN will have a public-facing website, which will be created and maintained by the CC, with directional input from the UDN Steering Committee. We anticipate that the content will include success stories, descriptions of the CSs and core laboratory sites of the network, publications, information for potential applicants, and information for researchers, among other content.

In addition, the CC will cultivate a social media presence that will begin with a Twitter account and may expand to include other social media forums, and the CC will proactively solicit information about publications, presentations, abstracts and scientific publications on a semi-annual basis.

All members of the UDN will be invited to submit content suggestions for the website.

X. Metrics

One of the core functions of the CC is to monitor each component of the network (CC, CSs, SCs, other Cores) as well as the network as a whole. The rationale for this measurement is two-fold: (1) to encourage understanding and continuous improvement of components of the network and (2) to codify the expectations of our funders in order to ensure that our efforts are aligned with expectations.

The NIH Program-defined measures are listed below: the NIH Program may update these measures over the course of the network. The CC may calculate a range of other measures to assess the performance of the network. A table of potential metrics is shown in Appendix 17: Proposed UDN Metrics.

It is anticipated that these metrics will be compiled and reported quarterly to the Steering Committee during the first two years of the UDN, after which the frequency of evaluation should be revisited by the Steering Committee.

Abbreviations: IRP-UDP = NIH Undiagnosed Diseases Program clinical site; ECS (external clinical site)

		Performance Metrics and Milestones
1	NIH	By Oct 1, 2014 NIH UDP will identify at least 5 candidate genes by working jointly with the Network by analyzing 400 SNPs and 400 WES or WGSs per year through FY2017
2	NIH	By Oct 1, 2016 Extramural Clinical Sites (ECSs) to see 25 patients per year per site to initiate phenotyping
3	NIH	UDN Sequencing Cores produce 200 exomes/genomes per year for analysis in collaboration with the UDN Clinical Sites to identify candidate genes by analyzing 200 exomes/genomes per year (year 1 clinical site participants and sequencing, continuing into year 2 with launch of the UDN), increasing to 300 exomes/genomes in FY2015 (year 2 clinical site participants and sequencing) and 450 exomes/genomes per year FY2016 to 2017 (year 3 and 4 clinical site participants and sequencing)
4	NIH	Define the mechanism of at least 1 candidate gene in the pathophysiology of a rare or yet-to-be described disease by FY2015 (year 2 of UDN Clinical Sites)
5	NIH	By Oct 1, 2017 (end of Year 2) all ECSs to see 50 patients per year per site
6	NIH	By Jan 2016–Identify 10 unidentified diseases; by Jan 2018, identify 20 unidentified diseases

XI. Biospecimens

Recommendations for research specimen collection on all UDN probands

A) Types of Specimens: The following specimens should be collected on all probands evaluated in person unless doing so would compromise participant safety or if they are refused by the proband (See Section B below regarding blood volume issues in pediatric probands).

1. 3 ml serum in 0.5 ml aliquots
2. 3 ml plasma in 0.5 ml aliquots
3. At least 20 micrograms of DNA (with goal of 50 micrograms) at a target concentration of 100-200 ng/ul (with a minimum of 50 ng/ul)
4. PBMCs stored in 1.0 ml aliquots containing $5x10^6$ cells each
5. 10 ml of urine in 1.0 ml aliquots

B) Pediatric Probands (and other probands with limited blood collections): For pediatric probands, the volume of blood drawn should be consistent with the allowable blood collection based on subject body weight. In cases where the blood volume that can be obtained is the limiting factor, samples should be obtained in the following order: EDTA tube (3 ml of blood) for DNA; EDTA tube (3 ml of blood) for plasma (if possible consider obtaining plasma and DNA from the same EDTA tube for pediatric patients); serum separator tube for serum (3 ml of blood); and 4 ml Citrate CPT tube for peripheral blood mononuclear cells (PBMCs).

C) Sample Collection:
   a. General Sample Collection Issues
      i. Blood samples for serum and plasma (optional for other sample types) should be obtained in the fasting state, defined as an overnight fast for adults and at least 3 hours of fasting for children. If a subject is unable to fast, samples should still be obtained. The clinical center should record whether blood samples were collected as fasting or non-fasting.
   b. Blood for PBMCs will be collected in CPT Vacutainer® tubes with citrate (one 8 ml tube for pediatric subjects and as total blood draw volume allows, and two 8 ml tubes for adult subjects; if sample volumes are limited by proband body weight use the smaller 4 ml CPT tube)
   c. Blood for DNA will be collected in one 10 ml purple top EDTA Vacutainer® tube and sent to a local CLIA laboratory for DNA extraction and quantification. If DNA from blood cannot be obtained, an alternative source of DNA such as skin fibroblasts should be considered.
   d. Blood for plasma will be collected in one 10 ml purple top EDTA Vacutainer® tube.
   e. Blood for serum will be collected in one 10 ml Red top Vacutainer® Serum Separator Tubes (SST) with clot activator.
   f. Urine samples should be the first morning void urine collected in a polypropylene container. A 24-hour urine sample is not required, but may be elected by the clinical center.

D) Blood Sample Processing:
    a. General Sample Processing Issues
        i. Processing of blood for plasma and serum, as well as urine, should be performed within two hours of sample collection. PBMCs should be processed within 24 hours of sample collection.
        ii. Serum, plasma, urine, DNA, and PBMCs should be stored in screw-cap cryovials appropriate for ultra-cold storage (Example: Nalgene NUNC 1.8 ml Cryovials, Fisher Scientific Catalog #: 12-565-170N).
    b. Serum Sample Processing
        i. After obtaining the SST sample, allow sample to clot 30 minutes in a vertical position
        ii. Follow manual instructions for use of local centrifuge, insuring balance of tubes within the centrifuge
        iii. Centrifuge at 2500 RPM or 1000 to 1300 g for 10 minutes either at ambient temperature or with refrigeration to 4^{o}C.
        iv. Remove Rubber Stopper and remove caps from Cryovial Tubes
        v. Aliquot 0.5 ml of serum into 6 cryovial storage tubes, and store samples in a -80^{o}C or liquid nitrogen freezer with appropriate labels (UDN ID number (7 digists), sample type, and collection date)
    c. Plasma Sample Processing
        i. Spin EDTA Vacutainer® tubes at 2500 RPM or 1000 to 1300 g for 10 minutes either at ambient temperature or at 4^{o}C.
        ii. Transfer plasma to storage tubes, with six 0.5 ml aliquots.
        iii. Store all samples in a -80^{o}C or liquid nitrogen freezer with appropriate labels (UDN ID number, sample type, and collection date)
E) DNA Extraction at CLIA Laboratory
    a. Multiple acceptable DNA extraction protocols for the EDTA Vacutainer tube blood samples can be used (Examples of suitable Extraction kits: Qiagen Gentra Puregene Blood Kit Catalog # 158445, or Qiagen DNeasy Blood and Tissue Kit Catalog# 69504). DNA should be stored in TE (Tris-EDTA) buffer). Tubes should be stored with appropriate labels (UDN ID number, date of birth, sample type, and collection date).
        i. DNA aliquots sent to the UDN sequencing cores must be labeled with patient name, date of birth, and UDN.
    b. DNA quantification should be performed with PicoGreen (not NanoDrop), and DNA concentration should be between 100 to 200 ng/ul (with a minimum of 50 ng/ul).
F) PBMC Isolation and Cyropreservation
    a. Isolation of PBMC – CPT tubes are the recommended cell separating device (refer to specific CPT tube manufacturer instructions for complete details for steps 1-4 and those below are offered as suggestions). Other cell separating devices may be utilized at individual sites for local biorepository.
        i. Centrifuge blood collected in CPT tubes at room temperature at 1500 to 1800 x g in a swing bucket rotor for 20-30 minutes with no brake following specific instructions from the CPT tube documentation. Visually inspect the CPT gel plug in addition to other guidance in the manufacturer's instructions.
        ii. Use an aspirating pipet to remove the PBMC layer located at the gel interface in the CPT tube.
        iii. Place the PBMCs in a new 50 ml conical tube

iv. Wash cells by gently resuspending the cell pellet in 10 ml sterile 4°C or ambient temperature PBS (or other physiologic buffer) followed by centrifugation at 250-400 x g. Repeat once.
v. Count the PBMCs on a hemocytometer, cellometer, or other standardized cell counting device
vi. Separate into aliquots of $5x10^6$ cells each in a separate polypropylene tubes and centrifuge at 250-400 x g to create the final cell pellet. The maximum number of aliquots should be made to allow for as many separate PBMC samples as possible to be saved from any one donor.

b. Preparation of PBMCs for storage in a cryorepository
i. To prevent contamination, all processing shall be completed in a sterile biological safety cabinet by wiping all inside surfaces with 70% alcohol and performing UV light treatment for at least 5 minutes.
ii. Resuspend the washed PBMC pellet containing 5 million cells in 0.5 ml of room temperature 0.2µm filter-sterilized 100% heat inactivated Fetal Bovine Serum. Ideally, all CSs will use a freezing media generated from the same FBS lot.
iii. Add dropwise 0.5ml of 0.2µm filter-sterilized 80% heat inactivated Fetal Bovine Serum with 20% dimethylsulfoxide.
iv. Mix cells by gently tapping the tube; do not use a pipette.
v. Pipette gently to minimize shear force and transfer into a labeled cryopreservation vial.
vi. For cryopreservation, transfer vials to a Controlled Rate Freezer to decrease the temperature in a controlled fashion (or if that is unavailable into an isopropanol containing cryopreservation system followed by transfer into a -80°C freezer for a minimum of 12 hours).
vii. Once cryopreservation vials have been appropriately cooled and contents frozen, transfer to designated receptacle within the liquid nitrogen storage unit for long-term storage with appropriate labels (UDN ID number, sample type, and collection date).

G) Urine Sample Processing
a. Urine samples will be collected from all probands as the first morning void sample and used for clinical laboratory studies and research purposes including metabolomics, lipidomics, and glycomics testing.
b. Urine samples will be preserved in 10x 1 ml aliquots of supernatant in 2.0 ml cryovial tubes at -80^{o}C or colder with appropriate labels (UDN ID number, sample type, and collection date).

H) Sample Tracking and Storage
a. Sample Labeling: Barcode labels of samples to be stored at a local or central biorepository should include the UDN ID number, sample type, and date of sample collection.
b. Sample Storage:
i. Serum, Plasma, and Urine aliquots will be stored at -80^{o}C or colder (e.g., liquid nitrogen)
ii. DNA samples will be stored at -20^{o}C or colder.
iii. PBMC will be cryopreserved in liquid nitrogen.

Recommendations for optional research biospecimen collection

A) Cerebrospinal fluid Collection and Processing

Cerebrospinal fluid (CSF) will be collected from neurological cases for clinical laboratory studies and for research use, including metabolomics, lipidomics, glycomics testing and luminex inflammome studies. All samples will be collected by lumbar puncture in the L3/L4 or the L4/L5 inter-space. If neurotransmitters are to be analyzed, they are to be collected in the following manner:

Tubes

1. Microtubes 1-5 (3.5 ml total): for shipment to Medical Neurogenetics

2. Microtubes 6-7 (1 ml each): for metabolomics, energetics, glycomics, and lipidomics

3. Polystyrene tubes 1-3 (0.5 ml each minimum): for measurement of glucose, cell count, protein, sterile fluid culture

4. Polystyrene tube 4: for IgG index, Oligoclonal bands, etc.

Collection

1. After CSF is collected by lumbar puncture, place the CSF on wet ice immediately and transport to the laboratory

2. If CSF is bloody, excessive blood may interfere with metabolomics testing

3. Store all samples in the -80^{o}C freezer with appropriate labels (UDN ID number, sample type, and collection date)

A) Optional Blood Collection and Processing
   a. Optional Blood Sample Collection
      1. As noted above, blood will be collected from all probands for DNA, plasma, serum, and PBMCs. Various other optional samples may be considered as well, including: PaxRNA, buffy coat, platelets, and blood spot cards.
      2. Blood for RNA will be collected in PAXgene blood RNA Vacutainer® tubes (VWR 77776-026)
      3. Blood for additional PBMC or other buffy coat collection will be collected in additional CPT (citrate) tubes or other site-specific collection tubes.
      4. If platelets are to be collected, draw 31.5 ml blood in 7 tubes of light blue top sodium citrate tubes (BLU).
      5. Blood spot cards may also be obtained and stored at room temperature.
   b. RNA Processing
      1. Use PAXgene Blood RNA Kit (Qiagen 762164)
      2. Aliquot in 80ul aliquots and store in 500ul sterile, RNase- DNase-free tubes at -20^{o}C until needed
   c. RBC isolation
      1. After removal of plasma (see procedures above), discard the remaining supernatant above the porous barrier using a plastic Pasteur pipette (wide orifice)
      2. Using a glass Pasteur pipette (narrow orifice), transfer the erythrocyte (RBC) pellet to a 50 ml conical tube

3. Fill 50 ml conical tube to 40 ml with Phosphate Buffered Saline (PBS), pH 7.4 and invert several times to mix
4. Centrifuge for 5min at 1811 x g at 4^0C
5. Remove saline layer and discard
6. Repeat wash with PBS pH 7.4 until PBS is clear (minimum of 3 times)
7. Aliquot 1 ml of the erythrocyte (RBC) pellet to clean cryovials. Store in -80°C freezer with appropriate labels (UDN ID number, sample type, and collection date). *Note: this fraction also contains granulocytes*

d. Platelet Isolation

8. Add 1 volume HEP buffer + PGE1
9. Mix very gently by inverting the tube slowly
10. Spin at 100 x *g* for 15-20 min at room temperature (with no brake applied) to pellet contaminating red and white blood cells
11. Transfer the supernatant into new plastic tube using a transfer pipet (wide orifice)
12. Pellet platelets by centrifugation at 800 x *g* for 15-20 min at room temperature (with no brake applied). Discard the supernatant.
13. Rinse the platelet pellet two times with platelet wash buffer by gently adding wash buffer and removing it slowly with a pipette. (DO NOT RESUSPEND! to avoid platelet activation)
14. Store the dry platelet pellet at -80^{o}C freezer with appropriate labels (UDN ID number, sample type, and collection date). *Note: Freezing the pellet will disrupt the platelet granules. This pellet is only to be used for determination of glycomics, lipidomics and proteomics that does not include the platelet granules.*

B) Skin Biopsy Collection and Processing

Skin biopsies will be used for culturing fibroblasts. These cell lines will then be used for various research purposes as well as glycomics testing. For subjects who are unable to provide PBMCs, skin fibroblasts provide an alternate source of living cells for future research.

Items needed: DMEM High Glucose (Invitrogen #11965-118); Fetal Bovine Serum (FBS), Certified, Heat-Inactivated, US Origin (Invitrogen #10082-147); 100X Antibiotic-Antimycotic (Invitrogen #15240-062); 0.25% Trypsin-EDTA (Invitrogen #25200-056); 1 x PBS pH 7.4 w/o Calcium or Magnesium (Invitrogen #10010-023); Dimethyl sulfoxide (Sigma #D8418-100ML)

Collection

1. 3-5mm punch full thickness skin biopsy obtained according to standard medical procedure

2. Place biopsy in sterile tissue culture medium (DMEM, 10% FBS, 1% antimycotic, antibiotic) contained in a 15 ml conical tube

3. Store and transport the biopsy at ambient temperature

4. Deliver the biopsy to the laboratory within 24 hours (Up to 96 hours is acceptable if shipped)

Processing- initiation of skin fibroblast culture

1. Spray hood and scalpels with Ethanol and wipe with Kimwipe
2. Clean the biopsy tube by spraying well with Ethanol before placing in the hood
3. Label 6-well tissue culture (TC) plate and place in TC hood
4. Aspirate medium from biopsy sample using a 2 ml aspirating pipet
5. Remove biopsy sample and place in one well of the 6-well dish
6. Using the scalpels, cut the biopsy sample into 6 pieces. (Try to attach the biopsy to the plate with the scratches made by the scalpel).
7. Add 1 ml of pre-warmed culture medium (DMEM, 10% FBS, 1% antimycotic, antibiotic) to each well, being careful not to dislodge the biopsy
8. Gently swirl the 6-well dish to coat the wells with culture medium
9. Place in the 37^{o}C, 5% CO_2 TC incubator for 4-5 days to allow the biopsy to attach to the well
10. Gently add 2 ml of fresh, pre-warmed culture medium to each well being careful to not dislodge biopsy sample
11. Allow the sample to remain in the 37^{o}C incubator until a monolayer of cells is present in the wells, feeding cells with fresh culture medium every 3-4 days
12. Once adequate cells have grown out of the biopsy fragment, remove the cells from each of the 6-wells by washing with 2 ml PBS, and following removal of the PBS, by adding 800ul trypsin (0.25% Trypsin EDTA, Invitrogen). Incubate at 37^{o}C until cells are released from substrate. Then add 2 ml of tissue culture medium and transfer cells to one T75 culture flask.
13. After 2 days, aspirate all but 1 ml of medium from flask.
14. Scrape the bottom of the flask using a cell scraper, and wash cells with the 1 ml of medium remaining in the flask. Remove the 1 ml of culture, and place in a 1.5 ml eppendorf tube. Perform mycoplasma testing using ATCC Universal Mycoplasma Testing Kit (see *Mycoplasma testing* below) on this 1 ml aliquot and record results.
15. Add 10 ml of fresh DMEM to flask and return to 37^{o}C tissue culture incubator to allow the remaining cells to proliferate.
16. Allow samples to reach confluency
17. Remove medium and wash cells with 10 ml PBS

18. Detach cells as described below (Detaching and Passaging Cells). Add 8 ml of tissue culture medium to collect cell suspension, use 1 ml of the culture to start another T75 culture flask. As described below (Freezing Cells), count the cells remaining in suspension, centrifuge at 1000 RPM for 10 minutes, add 3 ml of freezing medium, and freeze the remaining culture into 3x 1 ml cryovials at -80^{o}C in a cool cell for 3 days with appropriate labels (UDN ID number, sample type, collection date, and passage number). Transfer to -150^{o}C for permanent storage (Passage 1 cells).

19. Allow the second T75 flask to reach confluency, feeding with fresh DMEM every 2-3 days. Once confluent, trypsinize the flask, add 9 ml of DMEM count the cells, centrifuge at 1000 RPM for 10 minutes, aspirate supernatant, add 3 ml of freezing media, and freeze at -80^{o}C in a cool cell for 3 days with appropriate labels (UDN ID number, sample type, collection date, and passage number). Transfer to -150^{o}C for permanent storage (Passage 2 cells).

Processing- freezing fibroblast cells

1. Prepare 3-4 ml of fresh freezing medium (10% DMSO, 90% FBS) per T75 flask and warm to 37^{o}C.

2. Place the 9 ml fibroblast culture into a sterile 15 ml conical tube.

3. Count cells using the cell counter: Add 10ul of culture to BioRad cell counting slide and insert into the BioRad TC20 Automated Cell Counter. Multiply this number of cells by 9 (volume of total culture) and divide by the number of frozen culture aliquots you are making to determine the amount of cells frozen per tube.

4. Centrifuge culture at 1000 RPM for 5 minutes.

5. Aspirate off supernatant.

6. Add prepared freezing medium and mix by pipetting up and down.

7. Aliquot 1 ml into labeled cryovial.

8. Place cryovial into Cool-Cell that is labeled with your name and date.

9. Place Cool-Cell into -80^{o}C freezer for 3 days (36 hours) with appropriate labels (UDN ID number, sample type, collection date, amount of cells, and passage number) and then transfer into -150^{o}C storage

Processing- tissue culture for frozen skin fibroblasts

1. Once the tissue culture medium has warmed, remove cell vial from -150^{o}C and immediately place at 37^{o}C.

2. Prepare a T75 tissue culture flask or 10cm petri dish by adding 10 ml of pre-warmed tissue culture medium.

3. Remove cells from vial using a sterile 1 ml pipet tip or 1 ml pipet.

4. Add cells to flask and gently mix.

5. Place inoculated flask in a 37°C, 5% CO_2 tissue culture incubator for 24 hrs to allow cells to attach to the dish.

6. After 24 hrs, remove medium using a 2 ml aspirating pipet and replace with 10 ml of fresh tissue culture medium. Place at 37°C, 5% CO_2 in tissue culture incubator.

7. Feed cells every 2-3 days by aspirating off old tissue culture medium and replacing with fresh, pre-warmed, tissue culture medium.

8. Once cells have reached confluency, cells must be passaged and split into new T75 culture dishes.

Processing- Detaching and Passaging Cells

1. Warm 0.25% Trypsin EDTA to 37°C.

2. Warm DMEM, 10% FBS, and 1% Anti-Anti to 37°C.

3. Once reagents have warmed, aspirate medium from flask containing cells using a 2 ml aspirating pipet.

4. Rinse cells with 10 ml of 1X PBS pH 7.4 without Calcium or Magnesium.

5. Aspirate 1X PBS using 2 ml aspirating pipet.

6. Add 1 ml of pre-warmed 0.25% Trypsin EDTA to TC flask and spread across the attachment area by swirling the flask

7. Incubate flask at 37°C for ~ 5 minutes (or until cells are rounding), then gently tap the flask to release the cells.

8. Add 8 ml of pre-warmed tissue culture medium and wash cells to the bottom of the flask.

9. Add 3 ml of culture to fresh T75 flask containing 7 ml of tissue culture medium. Place at 37°C, 5% CO_2 in tissue culture incubator.

Mycoplasma testing

1. After 2-3 days of cell growth, aspirate all but ~1 ml of medium from flask.

2. Using a cell scraper, scrape the cells from the bottom of the flask only.

3. Using a 1 ml serological pipet, wash cells using the 1 ml of media in the flask.

4. Remove the 1 ml aliquot and place in a sterile 1.5 ml eppendorf tube.

5. Add fresh pre-warmed DMEM to flask and place in 37°C, 5% CO_2 tissue culture incubator for future use.

6. Perform mycoplasma testing using the Universal ATCC Mycoplasma Testing Kit (ATCC #30-1012K) according to the protocol on the 1 ml aliquot.

7. Record mycoplasma results and upload gel image into LIMS. ****If mycoplasma free, you can continue to passage. If mycoplasma positive, cells must be treated with Plasmocin (InvivoGen #ant-mpt) according to the protocol for 2 weeks and retested

XII. Central Biorepository

A. CS Web Access

The UDN Central Biorepository (UDNCB) website will be accessed through the Gateway. Here a CS will be able to submit samples, view samples available, request samples, and obtain information to contact the UDNCB lab for assistance.

I. Submitting Samples

1. CS follows specimen collection, processing, and storage guidelines described in Biospecimen section.
2. CS selects half of the processed samples for shipping to the UDNCB. Shipping can be in batches.
3. CS enters the sample information into the Sample Submission form (must be included with shipment).
4. CS prepares the samples for shipment on dry ice, FedEx priority overnight (Mon-Weds only), include printed copy of sample submission form (see 3).
5. UDNCB will notify CS when samples have safely arrived.
6. Website is still in development. More details on use of website and sample submission will be available.

II. Viewing Samples Available

1. Website is still in development but inventory will be viewable/searchable.

III. Requesting Samples

Prior Approval: Samples stored by the UDNCB are available to UDN investigators and their collaborators. However, quantities are limited. All sample requests will require prior approval from the UDN.

1. CS obtains prior approval from UDN.

2. CS enters sample information into the Sample Request Form (available on website).

3. UDNCB contact investigator to arrange a time to ship samples.

4. CS emails UDNCB that samples have arrived.

B. UDNCB Procedures

I. Storing and archiving of biological specimens

Mailed samples will be opened in a clean “no amplified DNA” laboratory. The frozen samples, pre-aliquoted in screw cap cryotubes and labeled by the sender, will be placed on dry ice while the labels are checked against the Sample Submission Form accompanying the package for confirmation. This form will be available for download on the Gateway and it

will be required to be included with all samples shipped to the biorepository. The Sample Submission Form will contain the participant UDN ID #s (also printed on the sample tubes). All tubes will have participant UDN ID #, sample type, and date of collection typed on the label. Receipt of the samples is recorded in the laboratory sample intake book and their condition noted (if dry ice is gone, samples are partially thawed, any tubes are cracked, etc). Samples are not required to have barcodes but the UDNCB has the ability to read 1D and 2D barcodes. If barcodes are included on the labels by the CS, the information outlined in the Biospecimen Section of the Manual of Operations must still be typed legibly on the sample labels.

Storage of Samples: The biological samples will be placed into liquid nitrogen cryotanks (PBMCs and possibly fibroblasts) and -80° freezers (DNA, serum, plasma, urine) for long term storage in the locations assigned by the Progeny Laboratory Information Management System (LIMS) database. Samples with multiple tubes will be divided into 2 separate freezers/cryotanks. All UDNCB equipment is on the Vanderbilt Delta alarm system with temperature and nitrogen fluctuation notification automatically going to the Director's and the senior Research Assistant's cell phone/pager.

Documentation: The Sender will be notified by email of shipment arrival and any problems that may have occurred with the shipment (late arrival, partially thawed tubes, broken tubes, etc). Any problems with the shipment will also be recorded into the Progeny LIMS database. The Sample Submission Forms will be completed by the CS submitting the samples and included with the samples when they are shipped. No patient names or identifying information is recorded in the Progeny LIMS, the sample intake notebook, or requested on the Sample Submission Forms.

II. Retrieval and Shipping of Biological Specimens

The UDNCB will retrieve biological samples from liquid nitrogen &/or -80° freezers, package samples in dry ice, and ship to UDN investigators and collaborators. Sample information in Progeny LIMS will be used to track quantities and distribution of biological samples.

Locating Biological Specimens in Storage: The Progeny LIMS database will contain participant UDN ID #, date of birth, sample type, date of collection, and sample location. In addition to sample and location information, the Progeny LIMS database will keep track of original and current quantities of the biological samples and record the distribution of samples to investigators.

Sample Retrieval and Transfer: The UDN must approve Sample Requests prior to application to the UDNCB. After UDN approval and receipt of a Sample Request Form the UDNCB will contact the CS or CS designated investigator requesting the samples by email to pre-arrange a date for shipment. The samples will then be located using the Progeny LIMS database, retrieved and placed on dry ice to prepare for transfer to the investigator requesting the sample.

Documentation of Retrieval: The type of sample, amount transferred, date of retrieval, and the CS designated investigator receiving the sample will be recorded in the Progeny LIMS

database. The Biorepository Sample Inventory on the Gateway will also be updated so that all UDN investigators can log in and see which samples and the amount(s) of each remain in the system.

Packaging and Shipping: The biological samples, already labeled and in screw cap tubes, will be packaged and shipped per International Air Transportation Association (IATA) requirements that apply to all dangerous goods (such as dry ice) by air. Samples must be triple packed which includes a leak proof bag with absorbent material. We will ship frozen samples in EPS foam containers (1.5 inch minimum thickness) with corrugated cartons, 10 lbs dry ice by FedEx priority overnight (Monday –Weds). Average dry ice sublimation in a 1-1/2 inch thick wall EPS container with corrugated container is 5 pounds over 24 hours (< 10 lbs in 48 hrs).

Delays can arise with FedEx and the extra dry ice is a safeguard to protect the samples in case of delays in delivery. A list of sample content will be included with shipment and the CS and CS designated investigator(s) will be notified by email that the sample has shipped, given the FedEx tracking number, and an electronic copy of the sample sheet. The email will request that the UDNCB be notified upon receipt of the shipment and that we be notified of any problems with the samples (tubes thawed or damaged, etc).

Packaging and Shipping Budget: Shipping samples to the UDNCB is at the CS expense and can be in batches to reduce costs. Shipping samples out from the UDNCB to investigators is at the expense of the UDNCB. We have budgeted for a total of 150 shipments over 3 years, as directed by the NIH, with 36 shipments in the first year and 57 in each subsequent year. If sample approvals by the UDN exceed the number of shipments allotted per year then additional funds will need to be made available for the UDNCB or, alternatively, the CS designated investigator(s) receiving samples will be required to pay the shipping costs (estimated to be $175/request).

Quality Control: The UDNCB will keep records on the number of samples received, their condition, date shipped/date arrived, etc. We will also keep records on the number of samples we ship out, date shipped/date received, condition upon arrival, any problems reported by the recipient, etc. This information will be compiled into a quality control report, along with total samples received and shipped, and presented to the Steering Committee 3 times per year.

III. Updating UDNCB Website Inventory

Samples collected by the CS and entered into the UDNCB Sample Submission Form prior to shipping are automatically entered into the Sample Inventory and will be able to be viewed in the Gateway. When samples are requested and shipped out through the UDNCB, the repository will edit/update the Sample Inventory on the Gateway.

XIII. Institutional Review Board (IRB) Communications

1. **Protocol Development**
   a. **Consent forms**
      i. **Templates**
      - The UDN PI, Central IRB (CIRB), and CC CIRB Liaison will develop model informed consent and assent form (ICF) templates for the UDN, noting sections of the template that must be customized by each CS.
      - The CC CIRB Liaison will make the template ICFs available to the CS Site Coordinators.
      - The CS Site Coordinators will customize only the areas of the ICFs specified in the template, including:
        - Placing the consent form on the institutional letterhead
        - Adding standardized language as required by the CS (due to local policy requirements)
        - Incorporating HIPAA authorization for use and disclosure of PII if HIPAA is not available as a separate document, as per with the CS institution's standard approach. If HIPAA is provided as a separate document, it does not need to be submitted.
      - The CS Site Coordinators will send the completed site specific ICFs to the CC CIRB Liaison.
      - The CC CIRB Liaison will review the ICFs and send them to the UDN PI to submit to the CIRB.
      - The CIRB will review the site specific ICFs with all of the other submitted site materials provided for site approval.
      - The CIRB will communicate the results of the review to the UDN PI, the CC CIRB Liaison, the local PIs, and the Institutional Designees.
      - The CC CIRB Liaison will communicate the results of the review to the CS Site Coordinators.
      - The CIRB will provide to the CC CIRB Liaison the approved ICFs for each CS. ICFs will have an expiration date as indicated on the last page of each form.
      - The CC CIRB Liaison will make the CIRB-approved ICFs available to the CS Site Coordinators and will store centrally for all CSs to access.

   b. **Investigator documentation**
      - Investigator documentation includes:
        - A roster of investigators who will be included on the protocol, a roster of non-investigator research staff, the site name, a description of the site, its location and Federalwide Assurance (FWA) number, documentation that UDN Site Human Research Protections Program (HRPP) training requirements have been met, and the name and contact information of responsible institutional officials.
        - Documentation of the local conflict of interest (COI) review for all investigators on the protocol indicating whether there are any unmitigated or existing conflicts.
        - Information about the UDN site's local research context as relevant to the site's role in the protocol.
      - The CC CIRB Liaison will send requests for documentation to the CS and Core Site Coordinators.

- The CS and Core Site Coordinators will send completed documentation to the CC CIRB Liaison.
- The CC CIRB Liaison will review and send the documentation to the UDN PI to submit to the CIRB.

2. Reportable Events

- Unanticipated problems involving risks to subjects or others (including adverse events and protocol violations) and/or serious or continuing noncompliance will be reported by the CS and Core PIs directly to the UDN PI, who will report them to the CIRB.
- The CC CIRB Liaison will make a form, generated by the CIRB, available to the CSs and Cores to use for reporting the unanticipated problems to the UDN PI.
- The CS and Core PIs will report all unanticipated problems or serious and/or continuing noncompliance within 7 or 14 days (depending on the nature of the unanticipated problem) to the UDN PI and copy the CC CIRB Liaison on all correspondences.
- The UDN PI will report the unanticipated problem or serious and/or continuing noncompliance to the CIRB within a timeframe that does not exceed the timing allowed for the PI.
- The CIRB will communicate the results of the review to the UDN PI, the CC CIRB Liaison, the local PI, the Institutional Designee, and in some cases, the Institutional Official related to unanticipated problems or serious and/or continuing noncompliance.
- The CC CIRB Liaison will be copied on all correspondences between the CC, UDN PI, and the CSs and Cores.

3. Continuing Review

- Three months prior to the continuing renewal deadline, the CIRB will notify the CC CIRB Liaison regarding information required for Continuing Review (CR) and provide the forms that all CSs and Cores, including the intramural site, must complete.
- The CC CIRB Liaison will notify each CS and Core Site Coordinator regarding information required for CR, including the CR Local Context Worksheet and other forms provided by the CIRB.
- The CS and Core Site Coordinators will submit their responsive information for CR to the CC CIRB Liaison within 2 months of the CR deadline.
- The CC CIRB Liaison will review the UDN site forms for accuracy and completeness.
- The CC CIRB Liaison will provide the individual site-specific CR forms as well as submit a single CR Application to the UDN PI.
- The UDN PI will review the applications and submit all documents to the CIRB.
- The CIRB will conduct CR of all submitted materials.
- The CIRB will communicate the results of the review to the UDN PI, the CC CIRB Liaison, the local PIs, and the Institutional Designees.
- The CC CIRB Liaison will communicate the results of the review to the CS and Core Site Coordinators.
- The CIRB will provide to the CC CIRB Liaison the approved ICFs for each CS, which will include a new expiration date.
- The CC CIRB Liaison will make the CIRB-approved ICFs available to the CS Site Coordinators and will store centrally for all CSs to access.
- **NOTE: All NHGRI protocols undergo review by the Scientific Review Committee (SRC) every three years. The SRC provides the UDN PI with a written review and a summary of outstanding comments and concerns. The UDN PI will provide the required materials to the SRC at least two months prior to submission to the CIRB for CR to permit sufficient time for SRC review. The same submission process**

used for CR, as referenced above, will be used for the triennial review.

4. **Amendments**

a. Study-wide amendments

- Study-wide amendments will be approved by the UDN Steering Committee before submission to the UDN CIRB.
- A completed amendment form with all supporting documentation, including tracked and clean copies of any modified documents, will be submitted to the CC CIRB Liaison. Study-wide amendments will be submitted to the CC CIRB Liaison the first Monday of the month.
- The UDN CC CIRB Liaison will finalize study-wide amendments and submit to the UDN PI.
- The UDN PI will submit study-wide amendments to the CIRB, including tracked and clean copies of all modified documents with updates to the version control of each document.
- If the study-wide protocol requires changes to the ICF:
    - The CC CIRB Liaison will modify the ICF template.
    - The CC CIRB will send tracked and clean copies of the modified ICF template to the UDN PI to submit for CIRB review.
    - Once the model template ICFs are approved by the CIRB, the CC CIRB Liaison will modify each site-specific document and provide them to the CIRB to update the version of the site specific ICFs.
- The CIRB will communicate the results of the review to the UDN PI, CC IRB Liaison, the local PIs, and the Institutional Designees.
- The CC CIRB Liaison will communicate the results of the review to the CS and Core Site Coordinators.
- The CC CIRB Liaison will make available to the CSs and Cores the approved amendment documents.
- If there are updated ICFs the CC CIRB Liaison will make the CIRB-approved ICFs available to the CS Site Coordinators and will store centrally for all CSs to access.

b. Site-specific amendments (e.g. study staff changes)

- The CC CIRB Liaison will provide a template for the CSs and Cores to complete for the amendment that fulfills the CIRB requirements.
- The CS and Core Site Coordinators will submit completed site-specific amendment forms with all supporting documentation, with tracked and clean copies of any modified documents, to the CC CIRB Liaison. Site-specific amendments will be submitted to the CC CIRB Liaison on the first and third Mondays of the month.
    - For study staff changes, the local PI must sign the completed amendment template indicating conflict of interest review and completed training requirements.
- The CC CIRB Liaison will submit the amendments to the UDN PI.
- The UDN PI will submit the amendments to the CIRB.
- The CIRB will communicate the results of the review to the UDN PI, CC CIRB Liaison, the local PI, and Institutional Designee.
- The CC CIRB Liaison will communicate the results of the review to the CS and Core Site Coordinators.
- The CC IRB Liaison will make available to the CS or Core the approved amendment documents.

5. Affiliated studies

- CSs and Cores may propose studies affiliated with the UDN that are not network-wide.
- These studies will require permission from the UDN Steering Committee and a separate IRB protocol through the site proposing the study.
- The CC CIRB Liaison will keep track of these studies.
- The UDN site carrying out an affiliated study that has an IRB-approved protocol will provide the CC CIRB Liaison with information about the study, which may include a brief summary of the study, sites involved, nature and characteristics of proband involvement and consent required, and lead UDN investigator.
- The CC CIRB Liaison will inform the UDN PI and the CIRB of affiliated studies.

DUE DATES

Site-specific amendments are due to CC CIRB Liaison on the 1st and 3rd Monday of the month.

Study-wide amendments are due to CC CIRB Liaison on the 1st Monday of the month.

TIMELINE- IRB PROTOCOL YEAR 1

APRIL 2015

UDN Protocol Approved: April 10th

MAY 2015

Site-specific amendments due to CC CIRB Liaison: May 18th

JUNE 2015

Site-specific amendments due to CC CIRB Liaison: June 1st, June 15th

Study-wide amendments due to CC CIRB Liaison: June 1st

JULY 2015

Site-specific amendments due to CC CIRB Liaison: July 6th, July 20th

Study-wide amendments due to CC CIRB Liaison: July 6th

AUGUST 2015

Site-specific amendments due to CC CIRB Liaison: August 3rd, August 17th

Study-wide amendments due to CC CIRB Liaison: August 3rd

SEPTEMBER 2015

Site-specific amendments due to CC CIRB Liaison: September 7th, September 21st

Study-wide amendments due to CC CIRB Liaison: September 7th

OCTOBER 2015

Site-specific amendments due to CC CIRB Liaison: October 5th, October 19th

Study-wide amendments due to CC CIRB Liaison: October 5th

NOVEMBER 2015

Site-specific amendments due to CC CIRB Liaison: November 2nd, November 16th

Study-wide amendments due to CC CIRB Liaison: November 2nd

DECEMBER 2015

Site-specific amendments due to CC CIRB Liaison: December 7th, December 21st

Study-wide amendments due to CC CIRB Liaison: December 7th

JANUARY 2016

Site-specific amendments due to CC CIRB Liaison: January 4th, January 18th

Study-wide amendments due to CC CIRB Liaison: January 4th

Continuing renewal forms sent to sites by CC CIRB Liaison: January 8th

FEBRUARY 2016

Site-specific amendments due to CC CIRB Liaison: February 1st, February 15th

Study-wide amendments due to CC CIRB Liaison: February 1st

Continuing renewal forms due to CC CIRB Liaison: February 8th

MARCH 2016

Site-specific amendments due to CC CIRB Liaison: March 7th, March 21st

Study-wide amendments due to CC CIRB Liaison: March 7th

April 2016

Continuing renewal deadline- April 9th

XIV. Billing Procedures

The UDN RFA stated that the CSs could bill subjects' health insurance for clinically indicated evaluations, procedures and tests, and use grant funds for underinsured or uninsured subjects. The CSs were also required to provide subject transportation and lodging/meals during the one-week stay at the CS. These practices would ensure that subjects did not incur out of pocket expenses and enable all subjects to have access to the UDN, irrespective of their health insurance status. This would also allow all subjects the same experience as at the NIH-UDP with no out of pocket expenses. However, while establishing billing procedures at the six CSs (outside of the NIH-UDP), it became evident that there were several challenges to implementing these practices. All the CSs were told by institutional representatives that insurance co-pays and deductibles could not be waived or reimbursed by the grant or institutional funds, due to a federal anti- inducement law that is framed for Medicare and Medicaid but is often applied to other insurance policies {42 U.S.C. § 1320a-7a(i)(6)}. Two sites were told by institutional representatives that they could not both bill insurance and pay for subject travel/lodging due to a federal anti-kickback law {42 U.S.C. § 1320a-7b}.

This led to the formation of a Billing Working Group to resolve the issues so as to not place the CSs and the subjects at an undue disadvantage. After considering the legalities and the available choices, two billing options were created. The first option utilizes grant funds solely to cover all the subject evaluations, made feasible by institutional discounts (~80%) for subject care performed as part of NIH-funded studies. The second option would bill the subjects' insurance companies for the clinical evaluations and cover underinsured/uninsured subjects or tests not reimbursed by insurance with grant or institutional funds. Each CS can choose which option is best based on their institutional policies. In addition, each CS can re-evaluate and change to the other option based on their institutional policies.

To enable payment of co-pays and deductibles at sites that would bill insurance, the UDN is collaborating with the National Organization for Rare Disorders (NORD, https://rarediseases.org/). NORD has established a UDN subject assistance fund with contributions from the Running for Rare Diseases team and a total of $212,000 has been allocated to the UDN for year 2. The Steering Committee will be allocated ~10% of these funds to spend at its discretion. All the CSs, including the NIH-UDP would receive ~20% of the NORD fund (total $35,000 in year 2: $5000 per site) to pay for tests needed for subjects before being accepted into the UDN. This amount will be utilized at the discretion of the CSs. The remaining ~70% of the NORD fund ($138,600 in year 2) will be utilized by the three CSs that will bill insurance to reimburse co-pays and deductibles for financially stressed patients (defined as those with an income below 300% of federal poverty guidelines). This plan would allow for seven patients at each of the three CSs to be reimbursed $6600 each, the maximum out of pocket expenses limit for an individual health insurance plan, as outlined by the Affordable Care Act (www.healthcare.gov)

The NORD funds ($212,000) for Y2 will be distributed, as detailed below:

	Discretionary	Pre-Clinical Evaluation	Clinical Care
Amount Allocated	$38,400	$35,000	$138,600
Sites Eligible	ALL	ALL	Baylor, Harvard, Stanford
Maximum – Per Site	n/a	$5,000	$46,200
Maximum – Per Patient	n/a	$5,000	$6,600

Similarly, a collaboration with Mercy Medical Angels (http://mercymedical.org/) would allow for provision of commercial air travel expenses for subjects who are financially stressed. A memo of understanding has been signed between the UDN and Mercy Medical Angels. Each CS will decide if and when they want to use Mercy Medical Angels to arrange travel for the subject and one care taker meeting the financial criteria (defined as having an income below 300% of the federal poverty guidelines). The CS will provide documentation of financial need and notify MMA at least two weeks prior to the date of travel to allow sufficient time for them to make the travel arrangements. The CSs will pay Mercy Medical Angels a $200 per ticket administrative fee from their grant funds and Mercy Medical Angels will arrange the travel for these subjects and their family member. Thus, the network is still able to offer evaluations to patients irrespective of their health insurance status.

The Billing Working Group will continue to review issues (see Appendix 18: Billing Surveys) that arise during the beginning of the patient enrollment period, and significant changes to the billing structures and/or NORD fund distributions will be presented before the Steering Committee for consideration.

XV. APPENDICES

APPENDIX 1: The NIH UDP Protocol

A) Screening (30 inquiries each week)

A Patient Care Coordinator (PCC), selected for having pleasing but firm interpersonal skills, provides a central point for all inquiries that range in specificity from direct physician-to-physician referrals to cold calls to NIH Call Center (866-444-8806) from patients or family members seeking to learn more about the UDP. The NIH Call Center refers these calls to the PCC (301-496-1465). Whatever the source of inquiries, the PCC mails the potential participant (or family, in the case of pediatric patients) an invitation package that includes a cover letter and an attached frequently asked questions document. A second letter is sent that the patient can share with his/her physician with an attached form for listing contact information of the current attending physician, a list of prior hospitalizations and specialists that have been involved in the patient's care. This is often followed by phone exchanges with the PCC to clarify goals and structure of the program and the information required for further evaluation. See Appendix 2: NIH UDP Patient Flow and Appendix 3: NIH-UDP Pre-CRC Admission for a detailed flow of patients prior to CRC admission.

Substantial delays are often encountered at this phase of patient recruitment as families often request medical records from multiple institutions, reflective of the long diagnostic odyssey. The UDP believes it is essential to obtain a physician referral letter in order to provide a clear, current picture of the patient's illness and to ensure follow-up care after completion of the UDP evaluation.

As detailed later, initial UDP medical review requires complete records of previous care and evaluations. Patients and physicians may encounter problems with collection of results of prior blood work, imaging, and special tests as they negotiate retrieval of these materials from various health care facilities. A series of form letters are used to remind potential participants of documentation required, but not yet received, including prior phenotyping and a physician's referral letter.

Clarification of the goals of the UDP sometimes results in withdrawal of applicants who have been interested only in a 'second opinion' process. Potential patients who fail to provide the necessary phenotyping data, or for whom there is no physician referral letter, will not be further considered. Approximately two-thirds of patients who were initially interested in learning more about the UDP or in participating fail to complete the information gathering process trimming the 30 per week who express an interest in the program to 10 who remain interested and whose records can be gathered and reviewed. For pediatric patients only approximately one-third of families fail to complete the process or are found ineligible, usually because they already have a diagnosis (e.g., they have an unbalanced chromosome translocation with multiple malformation syndrome but the family does not think this is the answer).

B) Creation and Careful Review of a CRC Medical Record (10 patients each week)

The next step in the recruitment process is to carry out a detailed review of each candidate's medical record including the referral letter from the current personal physician or physician-extender summarizing the salient features of the person's disorder, with reports detailing already collected phenotyping. These reports might include personal and family health history, physical examination, blood work and urine analysis/chemistries, imaging, and special studies such as cerebrospinal fluid findings, EMG, photos of skin lesions, and videos that display abnormalities of balance, gait, and strength. If biopsy or surgical procedures have been performed, biopsy slides may be reviewed by CRC pathologists if this appears to be essential for a decision. Prior imaging, especially CT and MR imaging, is extremely important in the

review process, and every effort is made to obtain the images themselves, and not simply reports. The clinical records available vary across patients, since some have had extensive prior evaluations by skilled physicians and others have had only a limited approach to finding a diagnosis.

The completed file is assigned to UDP team members and/or consultants to evaluate the likelihood that a rare or yet-to-be described disease is present and that the focused, systematic UDP approach might lead to a diagnosis. Useful indicators include other affected family members, objective physical findings, abnormalities found in blood work and/or imaging or other clues pointing to the presence of significant disease. A further consideration is whether, depending on family size and the availability of blood specimens on additional affected family members, the UDP's diagnostic armamentarium, especially SNP arrays or whole exome sequencing, could be useful in providing an answer. The review is physician intensive, and because records are often very extensive, the review process may be lengthy. Moreover, it may prove necessary to request additional information, or the advice of other UDP consultants. While the principal goal of this review process is to select patients for UDP evaluation, there are other potential results. Some patients may be more suitable for referral to other open NIH research protocols. If, in the judgment of the review panel, there has been incomplete patient evaluation, the panel may choose to return the patient to the referring physician with suggestions for further diagnostic approaches or recommend referral to an appropriate academic medical center.

The decision to invite applicants to travel, expense-free, to Bethesda, MD for a 5-day admission to the NIH CRC I made by Program Directors (Dr. Gahl and Dr. TIfft) after detailed discussion with consultants and other members of the UDP team. The goal of this review process is to insure, to the extent possible, that the problems posed by invited patients will be appropriate and match the resources of the UDP. A criterion for acceptance to the UDP is that the patient is safe to travel. The pediatric patients in particular are often medically fragile, medical clearance for commercial travel must be documented by the referring physician before patients can be accepted and scheduled for evaluation. Pediatric patients must have clearance from their physician one week prior to making the trip. In some cases visits need to be rescheduled if the patient is too sick to travel. UDP does not have the ability to pay for hospital-to-hospital transports, nor can they carry out these transports.

C) Preparation for the 5 Day Evaluation

Fitting the required diagnostic efforts into a 5-day evaluation requires careful planning to complete thorough phenotyping and place the findings in context for anxious patients and their families. This planning is complex and involves scheduling heavily used imaging resources and other diagnostic tests and insuring that initial evaluations by sub-specialists can be performed in a timely fashion. Patient- specific time constraints must also be considered.

D) Overall approach to phenotyping and specific data gathering in common subgroups

More than half of the patients accepted into the UDP have a neurological phenotype and in children particularly this leads to a common phenotyping framework that includes intracranial imaging (MRI and MRS), neurologic consultation, EEG, EMG/NCV, lumbar puncture for CSF neurotransmitters and other special testing, skin biopsy both for fibroblast culture for functional verification of new candidate genes and for immunohistochemistry and electron microscopy, ophthalmologic exam under anesthesia, physiatry consultation, and neurocognitive testing. In adult patients CSF is also obtained for immunologic studies.

E) UDN Metrics of success

The UDP has had much success, as can be seen from the following metrics:

a) Metrics in the 4 years following the establishment of the UDP:

- 6,300 inquiries evaluated
- 2,300 physician referral letters with patient medical records reviewed
- 450 patients admitted to the NIH-CRC (Clinical Research Center)

b) Weekly metrics:
- 30 new inquiries
- 10 ten patients with completed referral letters and results from prior diagnostic efforts evaluated
- 3 patients/families admitted for work-up at the CRC.

c) Diagnostic metrics:
- Approximately 100 patients (20-25%) were diagnosed with rare to extremely rare diseases
- Two patients were found with diseases unknown to medicine.
- 15 genes not previously associated with human disease were discovered and tentatively related to disease phenotypes.

F) Summary

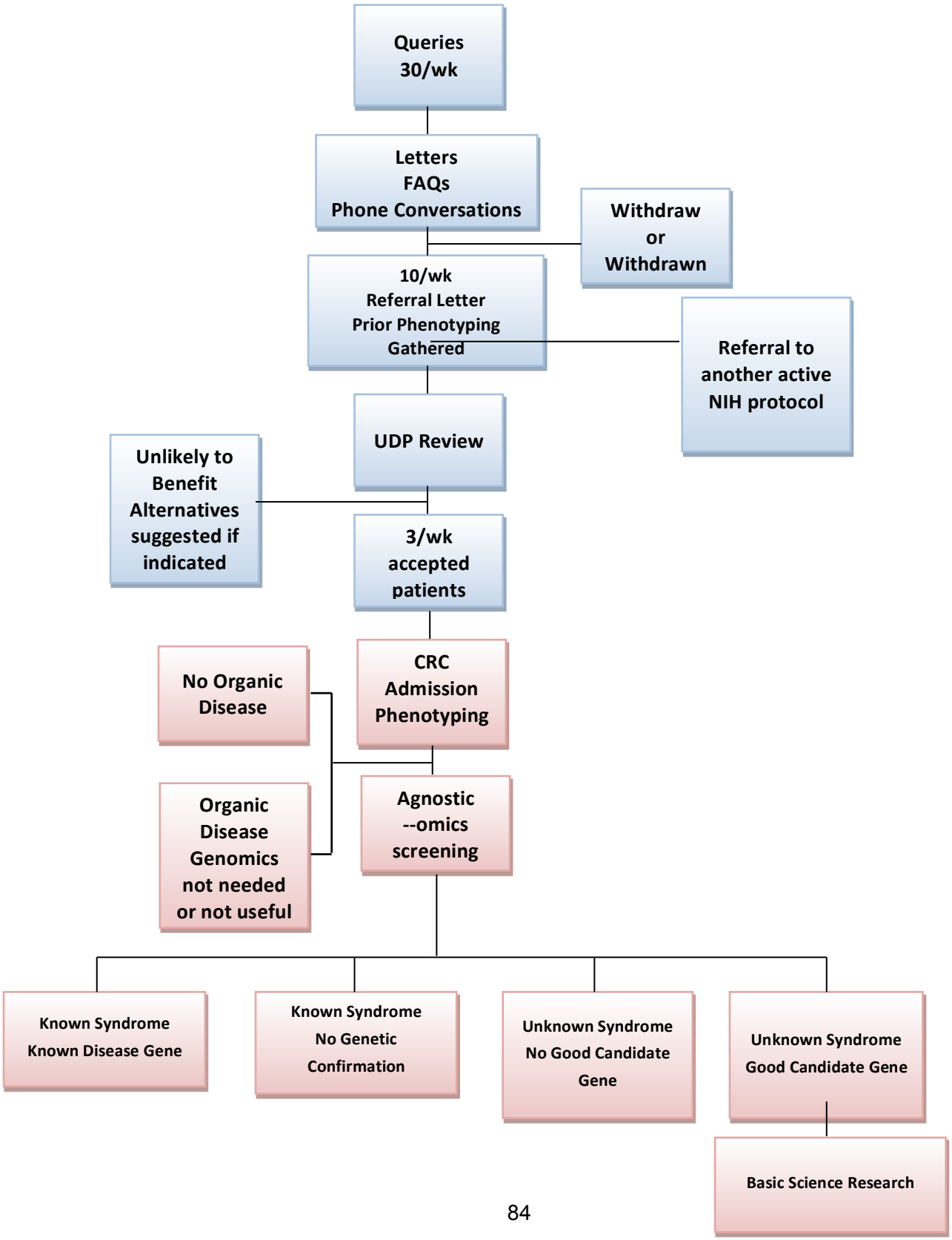
The current NIH-UDP initial approach to identifying and evaluating patients with undiagnosed diseases has been refined and focused over nearly five years. It seeks to identify participants who are most likely to have a rare or unknown undiagnosed disease. The 5-day admission to the NIH is designed to define the underlying pathophysiology by careful phenotyping and to identify settings in which genomics may prove useful.

The task for Network investigators including the NIH-UDP is to devise a UDN protocol that insures effective and efficient new site performance and retains a common approach to patient recruitment. Most importantly, uniform data collection with submission to the UDN coordinating center is critical to the success of the Network. Another important goal for the Network is the creation of a cooperative and collaborative team of Network investigators that recognizes and celebrates the diversity in new site talent and strengths, particularly in subspecialties. Additional work will be required to determine whether sites, in addition to evaluating all patients referred for evaluation, have a particular sub-specialty that might serve as a Network resource. Regularly scheduled discussions of difficult diagnostic cases among Network experts in the disorder suspected is likely to be yet another advantage of Network operations.

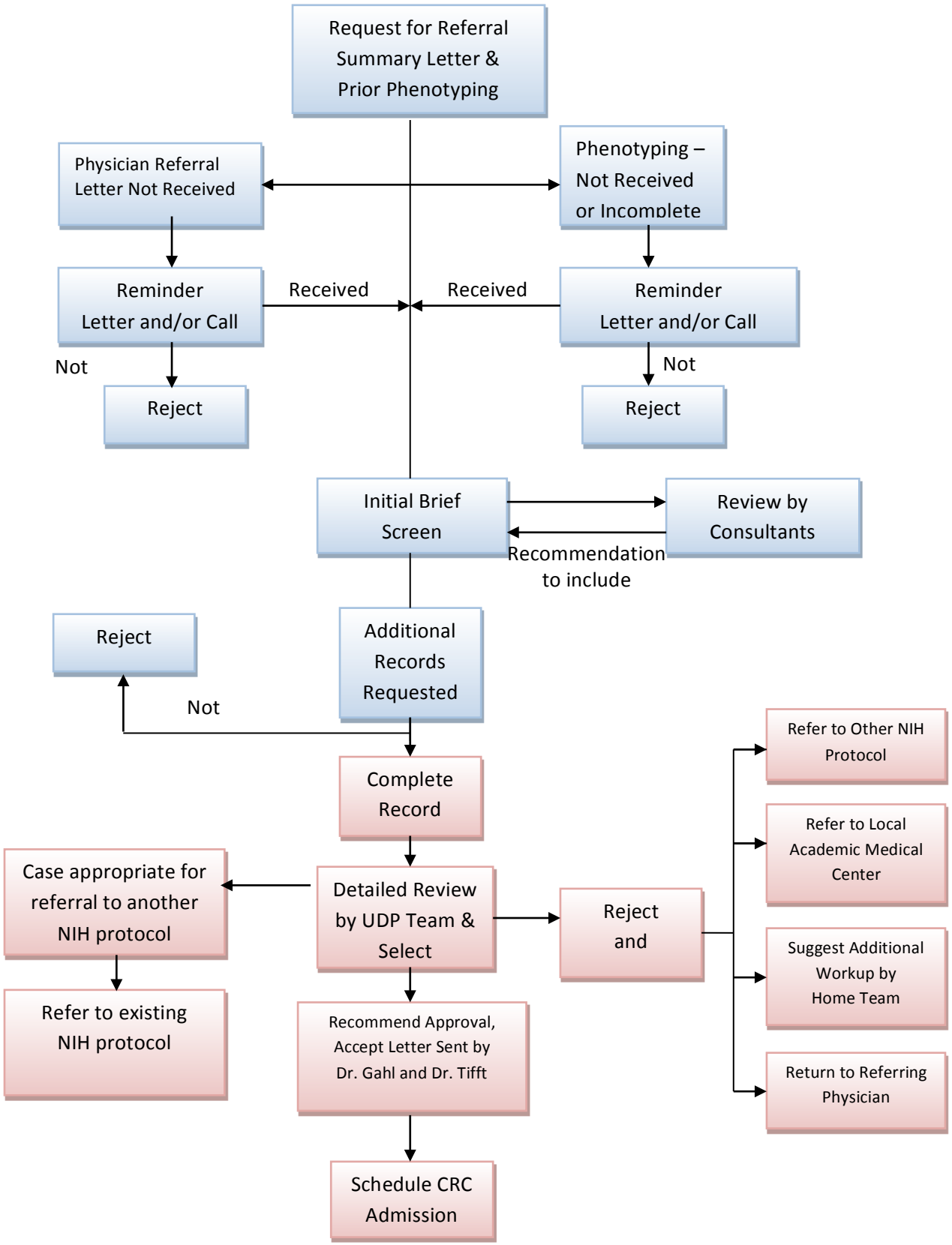
References (UDP background)

1. Gahl WA, Tifft CJ. The NIH Undiagnosed Diseases Program: Lessons learned. **JAMA** 305:1904-5, 2011.
2. Gahl WA, Markello TC, Toro C, Fuentes Fajardo K, Sincan M, Gill F, Carlson-Donohoe H, Gropman A, Pierson TM, Golas G, Wolfe L, Groden C, Godfrey R, Nehrebecky M, Wahl C, Yang S, Madeo A, Boerkoel CF, Tifft CJ, Adams D. The NIH Undiagnosed Diseases Program: Insights into rare diseases. **Genet Medicine** 14:51-9, 2012.
3. Gahl WA, Boerkoel CF, Boehm M. The NIH Undiagnosed Diseases Program: Bonding scientists to clinicians. **Dis Model Mech** 5:3-5, 2012.
4. St. Hilaire C, Ziegler SG, Markello T, Brusco A, Groden C, Gill F, Carlson-Donohoe H, Lederman RJ, Chen MY, Yang D, Siegenthaler MP, Arduino C, Mancini C, Freudenthal B, Stanescu HC, Zdebik A, Chaganti K, Nussbaum R, Kleta R, Gahl WA, Boehm M. *NT5E* mutations and arterial calcifications. **N Engl J Med** 364:432-42, 2011.

APPENDIX 2: NIH UDP Patient Flow



APPENDIX 3: NIH-UDP Pre-CRC Admission



APPENDIX 4: Case Review Committee of the UDN

Purpose: The Case Review Committee meeting is a forum for the CSs to concisely present patients to the UDN clinicians for review and input. The patients will fall into two general categories, 1) those that the CS has vetted and intends to invite for evaluation and 2) those for whom the CS is uncertain, has questions about, or thinks may be better served at another CS.

Format: The format of the meetings will be the presentation of a one-page summary of the case, and any imaging or pictorials that aid in the decision to accept for evaluation. Presentation and discussion of each case should last no more than five minutes. If two CSs need prolonged discussion about a patient, this can be taken off line after the meeting. Cases will be rotated among the seven CSs in each meeting. The meetings will terminate after an hour. A timer will move the meeting forward or take discussions off line if necessary.

Decisions: The decision to invite a patient for evaluation will be made by individual CSs. The meeting will exist to add value to the intended evaluation and to allow the UDN to be informed of the composition of the patient study population. It is expected that about half of cases will be pediatric, half adults. No more than half should be patients known to a CS, half completely new to any CS. “Patients known to a CS” are defined as any patient that is recommended by a healthcare provider from any of the institutions that are on that CS’s award. A patient recommended by an outside provider will not be considered “known to a CS” even if s/he has been seen previously at an institution on that CS’s award.

Structure: Each CS and SC should designate two Case Review Committee members and at least one alternate. Each CS should have at least one clinician present at any Case Review Committee meeting. Ordinarily, a pediatrician and an adult internist from each CS will be on each conference call.

The Chair and Co-Chair of the Committee will rotate among the CSs each 6 months. One of them should attend at each meeting. If both are conflicted for a meeting, then a member of the Committee will be asked to Chair.

APPENDIX 5: ClinicalTrials.gov Record

The UDN protocol "Clinical and Genetic Evaluation of Individuals With Undiagnosed Disorders Through the Undiagnosed Diseases Network" is shown below and **updates are** listed on ClinicalTrials.gov at the following URL: https://clinicaltrials.gov/ct2/show/NCT02450851

ClinicalTrials.gov

A service of the U.S. National Institutes of Health

Clinical and Genetic Evaluation of Individuals With Undiagnosed Disorders Through the Undiagnosed Diseases Network

This study is not yet open for participant recruitment. (see Contacts and Locations)
Verified April 2015 by National Institutes of Health Clinical Center (CC)
Sponsor:
National Human Genome Research Institute (NHGRI)
Information provided by (Responsible Party):
National Institutes of Health Clinical Center (CC) (National Human Genome Research Institute (NHGRI))
ClinicalTrials.gov Identifier:
NCT02450851
First received: May 19, 2015
Last updated: June 26, 2015
Last verified: April 2015

Purpose

Background:

- Without an explanation for severe and sometimes life-threatening symptoms, patients and their families are left in a state of unknown. The NIH helped create a network of medical research centers, called the Undiagnosed Diseases Network (UDN), to provide care and answers for these individuals.

Objectives:

- To improve diagnosis and care for people with undiagnosed diseases.

Eligibility:

- People with undiagnosed diseases, and their relatives.

Design:

- Participants will travel to one of the UDN medical centers for a 5-day clinical and research visit.
    - As part of the visit, UDN healthcare providers may ask participants to have:
        - Clinically indicated tests and procedures performed including:

        - A physical exam
        - Blood and urine tests
        - A review of health and family history
        - X-rays and body scans
        - Surveys
        - Photographs of the face and body
        - A special diet to see if the body can handle the food without having a reaction, like vomiting
        - Video or voice recordings
        - Other tests and procedures to help reach a diagnosis
    - Research tests and procedures performed including:
        - A skin biopsy. For this, a small piece of skin will be taken.
        - Surveys
        - Other tests and procedures for research that may not be related to a diagnosis or treatment.
- Most participants will be asked to give samples for genetic testing.
- Participants may be contacted after their visit to discuss test results. They may also be contacted in the future for interviews and surveys.
- Relatives of participants may be asked to give samples for genetic testing. They may be asked to have parts of their visit recorded and to have additional tests. They may also be contacted in the future for interviews and surveys.
- Clinical and research information collected will be stored in a database.
- Information and samples collected will be shared with others for research purposes.

Condition
Undiagnosed Disease

Study Type: Observational

Study Design: Time Perspective: Cross-Sectional

Official Title: Clinical and Genetic Evaluation of Patients With Undiagnosed Disorders Through the Undiagnosed Diseases Network

Further study details as provided by National Institutes of Health Clinical Center (CC):

Primary Outcome Measures:

- Making a diagnosis [Time Frame: Admission and ad hoc after that]
  [Designated as safety issue: No]

Estimated Enrollment:	8000
Study Start Date:	May 2015
Estimated Study Completion Date:	January 2021
Estimated Primary Completion Date:	January 2021 (Final data collection date for primary outcome measure)

Detailed Description:
Without an explanation for severe and sometimes life-threatening symptoms, patients and their families are left in a state of unknown. Many individuals find themselves being passed from physician to physician, undergoing countless and often repetitive tests in the hopes of finding answers and insight about what the future may hold. This long and arduous journey to find a diagnosis does not end for many patients- the Office of Rare Diseases Research (ORDR) notes that 6% of individuals seeking their assistance have an undiagnosed disorder. In 2008, the National Institutes of Health (NIH) Undiagnosed Diseases Program (UDP) was established with the goal of providing care and answers for these individuals with mysterious conditions who have long eluded diagnosis. The NIH UDP is a joint venture of the NIH ORDR, the National Human Genome Research Institute Intramural Research Program (NHGRI-IRP), and the NIH Clinical Research Center (CRC). The goals of the NIH UDP are to: (1) provide answers for patients with undiagnosed diseases; (2) generate new knowledge about disease mechanisms; (3) assess the application of new approaches to phenotyping and the use of genomic technologies; and (4) identify potential therapeutic targets, if possible. To date, the UDP has evaluated 3300 medical records and admitted 750 individuals with rare and undiagnosed conditions to the NIH Clinical Center. The NIH UDP has identified more than

70 rare disease diagnoses and several new conditions. The success of the NIH UDP prompted the NIH Common Fund to support the establishment of a network of medical research centers, the Undiagnosed Diseases Network (UDN), for fiscal years 2013-2020. The clinical sites will perform extensive phenotyping, genetic analyses, and functional studies of potential disease-causing variants. The testing performed on patients involves medically indicated studies intended to help reach a diagnosis, as well as research investigations that include a skin biopsy, blood draws, and DNA analysis. In addition, the UDN will further the goals of the UDP by permitting the sharing of personally identifiable phenotypic and genotypic information within the network. By sharing participant information and encouraging collaboration, the UDN hopes to improve the understanding of rare conditions and advance the diagnostic process and care for individuals with undiagnosed diseases.

 Eligibility

Ages Eligible for Study: 1 Month and older
Genders Eligible for Study: Both
Accepts Healthy Volunteers: No
Criteria

- INCLUSION CRITERIA:
- The applicant does not have a diagnosis that explains the objective findings.
- The applicant (or legal guardian) agrees to the storage and sharing of information and biomaterials in an identified fashion amongst the UDN centers, and in a de-identified fashion to research sites beyond the network.

EXCLUSION CRITERIA:

- The applicant has a diagnosis that explains the objective findings.
- Review of the records suggests a diagnosis and further evaluation is deemed unnecessary.
- The applicant is too seriously ill to travel safely to the UDN site.

 Contacts and Locations

Choosing to participate in a study is an important personal decision. Talk with your doctor and family members or friends about deciding to join a study. To learn more about this study, you or your doctor may contact the study research staff using the Contacts provided below. For general information, see Learn About Clinical Studies.

Please refer to this study by its ClinicalTrials.gov identifier: NCT02450851

Contacts

Contact: Paul Mazur (844) 746-4836 udn@hms.harvard.edu

Locations

United States, Maryland

National Institutes of Health Clinical Center, 9000 Rockville Pike — Not yet recruiting
Bethesda, Maryland, United States, 20892
Contact: Paul Mazur 844-746-4836 udn@hms.harvard.edu

United States, Massachusetts

Boston Children s Hospital — Not yet recruiting
Boston, Massachusetts, United States

United States, New York

Columbia University — Not yet recruiting
New York, New York, United States, 10032-3784

Sponsors and Collaborators

National Human Genome Research Institute (NHGRI)
Investigators

Principal Investigator:	William A Gahl, M.D.	National Human Genome Research Institute (NHGRI)

More Information

Additional Information:
NIH Clinical Center Detailed Web Page
https://undiagnosed.hms.harvard.edu

Publications:
Gahl WA, Boerkoel CF, Boehm M. The NIH Undiagnosed Diseases Program: bonding scientists and clinicians. Dis Model Mech. 2012 Jan;5(1):3-5. doi: 10.1242/dmm.009258.
Gahl WA, Markello TC, Toro C, Fajardo KF, Sincan M, Gill F, Carlson-Donohoe H, Gropman A, Pierson TM, Golas G, Wolfe L, Groden C, Godfrey R, Nehrebecky M, Wahl C, Landis DM, Yang S, Madeo A, Mullikin JC, Boerkoel CF, Tifft CJ, Adams D. The National Institutes of Health Undiagnosed Diseases Program: insights into rare diseases. Genet Med. 2012 Jan;14(1):51-9. doi: 10.1038/gim.0b013e318232a005. Epub 2011 Sep 26.
Gahl WA, Tifft CJ. The NIH Undiagnosed Diseases Program: lessons learned. JAMA. 2011 May 11;305(18):1904-5. doi: 10.1001/jama.2011.613.

Responsible Party:	National Institutes of Health Clinical Center (CC) (National Human Genome Research Institute (NHGRI))
ClinicalTrials.gov Identifier:	NCT02450851 History of Changes
Other Study ID Numbers:	150130, 15-HG-0130
Study First Received:	May 19, 2015
Last Updated:	June 26, 2015
Health Authority:	United States: Federal Government

Keywords provided by National Institutes of Health Clinical Center (CC):
Rare Diseases
Undiagnosed Disease

ClinicalTrials.gov processed this record on July 14, 2015

APPENDIX 6: Example Referral Letters

PEDIATRIC REFERRAL LETTERS

Example Letter #1:

To Whom it May Concern:

We are writing to you to request consideration of siblings, [patient names], for enrollment in the Undiagnosed Diseases Network (UDN). [Patient names] are followed by multiple specialists at [hospital name]. They are also followed by local pediatrician [physician name] for routine pediatric care.

[Patient name] is now a [age] year old [gender] with a history of dysmorphic features, failure to thrive, and hepatomegaly of unknown origin. Due to cryptogenic cirrhosis, liver transplant was performed at [age] months of age. Pathology results of [patient name]'s previous liver biopsy was suspicious for a [condition], specifically [specific condition], however, molecular testing for the [gene name] was negative. [Condition] enzyme screening and [condition] screen for the explanted liver sample came back in the low ranges, but not in the deficiency range usually seen. Additional extensive work-up was unrevealing.

[Patient name] is now a [age] month old [gender] noted prenatally to have holoprosencephaly via fetal MRI at [time] weeks gestation. Brain MRI performed on DOL [time] was consistent with [description of MRI]. [He/she] was admitted at [age] months of life for evaluation of liver steatosis, microcephaly, and failure to thrive. At [age] months of age, [patient name] was identified to have new onset hepatomegaly in [month] with vomiting. A liver biopsy from [date] identified [results of liver biopsy]. Due to persistent FTT, G tube was placed in [month] with subsequent fungal peritonitis, now post-[time] day course of [medication]. [Patient name] continues to have daily emesis. [Patient name] is currently evaluated for liver transplantation (persistently elevated transaminases and synthetic dysfunction).

Of note, both siblings have a history of IUGR with failure to thrive, improved for [patient name] following liver transplantation. [Patient name] has a history of developmental delays, making significant progress with therapies, and now within normal limits. [Patient name] continues to have developmental delays and facial features similar to [his/her] [brother/sister] during infancy.

Given the similarities in the presentation of these two siblings with an unremarkable family history (parents are not consanguineous), whole exome sequencing was obtained for [patient name] and identified a heterozygous mutation in the [gene name], which in the homozygous state is associated with [condition]. Subsequent deletion/duplication testing via the MitoMet oligonucleotide array returned as normal. Mitochondrial genome testing via massively parallel sequencing was obtained for [patient name] and was unrevealing.

At this time, we are unable to identify a specific genetic etiology that would explain the findings seen in [patient name] and [patient name]. The presentation of two siblings with similar features, however, is suggestive of a possible autosomal recessive condition, which remains undiagnosed at this time. Parents are interested in identifying a diagnosis, and are also interested in having a third child. We would like to refer these siblings to the Undiagnosed Diseases Network for further evaluation to try and identify a diagnosis. Thank you for your review and consideration for acceptance into the program. Please do not hesitate to contact our office at [phone number] if you have any questions or require any additional materials.

Sincerely,

[Referring provider]

Example Letter #2:

To Whom it May Concern:

I have followed [patient name] since [age] months of age. [He/she] has a history of significant global developmental delay, [he/she] is nonverbal, has hyperoral behavior, macrocephaly, small stature, [further description]. [He/she] has a great disposition and visually interacts with [his/her] environment. [He/she] has continued to make very slow but steady motor development but has never developed speech. [He/she] has never had seizures or developmental regression. Significant genetic, metabolic, and neurodiagnostic evaluation (as listed below) has yet to yield an underlying diagnosis. I am referring [him/her] to the Undiagnosed Diseases Network in attempts to find a unifying diagnosis for [his/her] multitude of symptoms. I truly feel that there is an underlying metabolic or genetic cause for [his/her symptoms that our testing thus far has not uncovered. [He/she] has been seen by numerous other specialists across the country.

[His/her] evaluation to date includes:

Normal or negative metabolic studies:

Urine organic acids

Serum amino acids

Creatinine guanidinoacetate

Etc.

Normal or negative genetic studies:

Routine chromosomes

Chromosome microarray [years]

mtDNA point mutations and deletions

GeneDx 101 mitochondrial nuclear gene panel

Etc.

Neuroimaging/neurodiagnostics:

[year]- MRI showed [results]

[year] CT showed [results]

Etc.

Normal or negative CSF studies:

Neurotransmitters

Biopterin

I truly appreciate your consideration for evaluation for [patient name]. This family has been on a very long quest to find a diagnosis and would be grateful for the opportunity to have [him/her] evaluated through the UDN.

Sincerely,

[Referring provider]

Example Letter #3:

Dear Undiagnosed Diseases Network,

I wholeheartedly recommend [patient name] to be evaluated by the Undiagnosed Diseases Network. [He/she] is a [age] year old with persistent myalgias, dyspnea, [description of condition] of unknown etiology. There are several other family members who are less severely affected with similar symptoms, suggesting a genetic etiology.

I recently met [patient name] to evaluate him for endocrinologic involvement of [his/her] presentation. While I did not find any endocrine pathology, I wanted to take the opportunity to refer him to the UDN. The notes from [his/her] neurologist Dr. [name] will have more details on his history, but I will describe the summary of what I learned.

[patient name] currently presents with [symptoms]. [Patient name]'s family reports that [patient name]'s symptoms initially began at [age] years of age when he began complaining of leg pain out of proportion to those expected for his age. He was evaluated at [age] years of age by a rheumatologist at [hospital], and then by Neurology where a deltoid biopsy was performed and reportedly normal. Additionally, other genetic testing for different forms of [condition] was negative. [He/she] was then referred to Dr. [name] at [hospital]. An EMG was normal, but a quadriceps biopsy showed a predominance of [finding] of unclear significance.

[Patient name]'s symptoms have all progressed over time. [He/she] complains of significant exercise intolerance and weakness in all muscles that have been slowly worsening over time. [His/her] weakness is particularly extreme after activity. Additionally, [patient name] has pain in [his/her] legs, around [his/her] neck, and lower back that is present all the time, although also worsened with activity. [He/she] has seen some improvement in the pain, especially in [area], after starting [medication]. [He/she] occasionally tries [medication] without much relief. The pain is particularly bad [time of day] whereas [his/her] other symptoms seem to be more extreme [time of day]. [His/her] [parent] notes that [he/she] also has some ptosis and [symptom] on several mornings when [he/she] wakes up that sometimes persists later in the day. Initially, this was one-sided, predominantly on the [side], but now appears to be bilateral. [Patient name] walks when [he/she] is at home but uses a wheelchair for transportation of further distances. [He/she] also appears to have [symptoms]. [His/her] motor strength and reflexes, however, are typically normal when [he/she] is evaluated in the neuromuscular clinic, suggesting that [patient name] has more trouble with fatigue than baseline muscle weakness. [He/she] also has a normal serum CK level. [He/she] has had evaluations for [syndromes] that were negative. [He/she] also had an empiric trial of [medication] that did not improve [his/her] symptoms. Dr. [name] most recently requested a [test] given the [symptoms].

[Patient name] recently developed [symptom] on [his/her] back, which particularly precipitated the referral to my clinic. I did not feel any sign of excess [hormone]. [He/she] was also evaluated by dermatology who felt these to be [condition]. Additionally, [he/she] has a [birthmark].

[Patient name] has been seen by several other specialists. [He/she] follows with Dr. [name] at [hospital] for pulmonary and has been noted to have [symptom]. [He/she] also was briefly followed by Dr. [name] in [state] at [hospital] for some time, but no further diagnoses were noted. [He/she] has been evaluated by Cardiology with a normal echo and EKG. [He/she] has also been evaluated by Physical Therapy, who did not think that [he/she] would benefit from their intervention due to [his/her] exercise intolerance.

[Patient name]'s family history is of particular interest. [His/her] [Parent] is healthy other than migraines and is of [ethnicity] background. [His/her] [Parent] is healthy and of [ethnicity] descent. There is no consanguinity in the family. [Patient name] has [number] siblings. [His/her] oldest sibling is [age] years old with some slight degree of muscle weakness as well. [He/she] has [number] healthy child and is currently pregnant with no complications. [Patient name]'s oldest brother is [age] years old, and his next sibling is [age] years old. Both of them are healthy except for some asthma and allergies. [Patient name] has an [age]-year-old sibling who has joint and muscle problems that are not as severe as [patient's]. Etc.

Thank you for your consideration of [patient's] application.

Sincerely,

[referring provider]

ADULT REFERRAL LETTER

Example Letter

Dear Undiagnosed Diseases Network Team:

I propose my patient [name] for your special protocol in the Undiagnosed Diseases Network. When I learned of your protocol, I immediately thought of [him/her]. [He/she] seems an ideal participant in your program.

Symptoms & History: [Name] suffers from an excruciating and bizarre illness that has devastated [his/her] life and gone undiagnosed for [number] years despite exhaustive workups at [institution] and here at [institution]. [He/she] has consulted over 100 medical specialists of whom many are at the pinnacle of their fields. [name] is a pleasant, intelligent [man/woman] and a motivated, cooperative patient.

- [Name] is a fair-skinned [age]-year old [man/woman] who has been disabled for the last [number] of years by burning facial pain and flushing of elusive etiology. [His/her] entire face and ears are involved; they are inflamed, red, and hot to the touch.
- Onset was rapid and for no apparent reason. Prior to the illness, [he/she] was in excellent health, a parent with a healthy child and successful businessman who worked full-time.
- The facial pain requires [name] to remain nearly all the time in a cold room with a fan blowing directly on [his/her] face. [more explanation]
- While [name]'s face and ears are chronically hot, the rest of [his/her] body [description].
- [Name] has anhidrosis over 90% of [his/her] body. However, sweating that cannot be elicited by heat can sometimes be elicited with [system] stimulation.
- [He/she] developed [eye condition] in [his/her] [age], since remedied surgically.
- Other major symptoms include:

Diagnostics & Etiology: [name]'s case is a medical mystery cutting across many organ systems/braches of medicine. One might describe it functionally as a putative sympathetic neurologic disorder of the thermoregulatory system that especially affects the vasculature and skin of the head. The origin of the proposed neuropathy could be genetic, autoimmune, infectious, toxicological, or some combination.

There are a number of tantalizing but unexplained clues including:

1. [He/she] is a carrier of one copy of the gene for the rare recessive genetic disease [condition], of which [his/her] relative died. But the [condition] experts have never seen symptoms manifested in a [condition] carrier.
2. [Protease] levels are chronically high, but not high enough for [condition]/
3. [He/she] tests relatively normal on most blood and urine diagnostics, but with some curious exceptions: high on [tests]. Low on [tests].
4. [Medication] has a minor positive effect on [his/her] symptoms and [he/she] takes it on an ongoing basis. This is the most helpful of the 100 or so medications that have been tried.
5. [He/she] has idiosyncratic negative reactions to many medications, often responding to "subclinical" doses.
6. [Name] was on a course of the medication [medication name] when [his/her] illness started, but there are no other documented cases of such a reaction to this medication.
7. A number of surgical sympathetic blocks have been implemented on a temporary basis, sometimes with great beneficial effect and sometimes the opposite.
8. Her/His illness bears some similarity to [condition], itself a rare and largely unexplained disease. However, [condition] affects the feet and sometimes the hands, and there is little or no reference in the literature to a similar disease affecting only the face.

Records: [Name] has carefully retained and organized the voluminous diagnostics and reports on [his/her] condition over [time] years seeking a diagnosis and treatment. This should be helpful to your efforts. I enclose the information your program requires including case summaries, laboratory reports, and reports from consults.

My role: While I am a [specialist] in private practice, I have served as [his/her] primary physician since very early in the illness. I would be pleased to support your efforts and provide follow-up. I understand that several other physicians that regularly see [name] are also in support of [his/her] application and would be available to communicate with you if requested.

Patient's perspective: [name] has been exhaustive and courageous in seeking an explanation for this illness. [He/she] read about your program in [magazine]. [He/she] fully understands that your program is primarily for research purposes and that the chances of significant benefits from participating are rather small. Please consider [him/her] for your program. My contact information and [his/hers] appears below.

Sincerely,

[Referring provider]

APPENDIX 7: Suggested Triage Methods

1. Once the applicant has been assigned to a clinical site, the site will contact the applicant and request that he/she send all information they have related to the reason for their application to the UDN These may include: medical records, reports, laboratory studies, radiographic studies, photographs or videos, and pathology slides and reports.
2. Records will be reviewed by the CS for completeness. The CS will request any missing components (e.g., images, biopsy slides). With appropriate release of information from the applicant, the site may request medical records directly from any medical centers where the patient has been seen.
3. Once the assigned site receives the information, the site will collate the information collected into folders and will scan the files to facilitate distribution for review.
4. The site director or his/her designee will assign the records for review to consultants based upon the specialty involved.
5. If during the review it becomes clear that more information is needed, the staff at the CS will contact the applicant to request more information.

APPENDIX 8: Applicant Review Form (completed by Clinical Sites)

Applicant name: _____

UDN identifier: _____

Date of birth: _____

Date application submitted: _____

**Auto-populates from Gateway application*

UDN site: _____ *******Auto-populates from site assignment*

Name of primary reviewer(s): _____

Category of primary condition (*drop down list)***:**

- Allergy/immunology
- Cardiology and vascular conditions
- Dentistry and craniofacial
- Dermatology
- Endocrinology
- Fibromyalgia/chronic fatigue syndrome
- Gastroenterology
- Gynecology and reproductive
- Hematology
- Infectious disease
- Musculoskeletal and orthopedics
- Nephrology
- Neurology
- Oncology
- Psychiatry
- Pulmonary
- Rheumatology
- Multiple pediatric (multiple congenital anomalies)
- Other
- None of the above

Please provide a narrative summary (150-200 words) of the applicant's condition. If applicable, please include:

- History of present illness
- Date symptoms first noted
- Past medical history
- Previous diagnoses/comorbidities (using ICD terms if possible)

- Prior procedures and surgeries.

Please indicate the applicant's pertinent prior evaluations. If applicable, please include:

- Prior positive or negative test results
- Prior genetic testing (especially whole exome sequencing)

Provisional diagnosis/working plan:

Other family members affected: Yes/No

- If yes:
  - How many affected? _____
  - How many available for analysis? Unknown/Some/All/None

Patient images: *Attach files*

Other files: *Attach files*

Category 1: Inclusion/Exclusion Criteria

Inclusion Criteria

- □ Does Not Have Diagnosis Explaining Objective Findings
- □ Agrees to Storage and Sharing of Information & Biomaterials

Exclusion Criteria

- □ Has Diagnosis Explaining Objective Findings
- □ Diagnosis Suggested Based on Record Review; Further Evaluation Unnecessary
- □ Too Ill to Travel Safely to UDN site

Category 2: Strengths (≥3 Recommended)

- □ Objective Abnormal Finding(s)
- □ Unique Clinical Presentation

- ☐ Multiple Systems Affected
- ☐ Family History of Condition
- ☐ Relevant Family Members Available for Testing
- ☐ High Likelihood of Genetic Diagnosis
- ☐ Local Patient
- ☐ Relevant to Other UDN Patients
- ☐ Can Offer Sequencing
- ☐ Can Offer Additional Clinical Workup
- ☐ Other

Category 3: Limitations (<1 Recommended)

- ☐ No Relevant Family Members Available for Genetic Testing
- ☐ UDN Resources Not Appropriate for Case
- ☐ High Likelihood of Not Solving Case at Present
- ☐ Proband Likely to Refuse Certain Tests/Procedures
- ☐ No Objective Clinical Findings
- ☐ Other

Recommend for Acceptance- At Clinical Site	**Questionable Case-** Send to Case Review	**Not Accepted with Recommendations-** Seek expert care	**Not Accepted-** Diagnosis Identified
Recommend for Acceptance- At Different Site	**Not accepted/Reconsider**	**Not Accepted with Recommendations-** Specific testing	**Not Accepted-** UDN Would Likely Not be Able to Help Find a Diagnosis
			Not Accepted- Insufficient records made available to UDN site

APPENDIX 9: UDN Generic Letters (for patients and health care providers)

Description: *Information for patients*

Date:
Address:

Dear [patient]:

Thank you for your interest in the Undiagnosed Diseases Network (UDN). Participants accepted into this program will be part of a clinical research study aimed at answering questions about medical conditions that have eluded diagnosis. We hope to advance medical knowledge in ways that can help improve health care for everyone. The study will be conducted at Baylor College of Medicine, Boston Children's Hospital/Brigham and Women's Hospital/Massachusetts General Hospital, Duke University, the National Institutes of Health, Stanford University, University of California-Los Angeles, and Vanderbilt University Medical Center.

Please discuss your participation in this program with your primary healthcare provider. Important considerations include:

- This is a pilot program with strict eligibility requirements.
- Many cases accepted will NOT result in a diagnosis.
- A referral by a healthcare provider is required.
- The provider who refers you will be asked to provide your medical information.
- The UDN will communicate the decision on accepting your case for evaluation in writing to you and your referring healthcare provider.
- If your case is accepted for UDN evaluation, the UDN will provide information from the evaluation to you and to your healthcare provider.
- Your healthcare provider will be responsible for your medical care after you have been evaluated in the UDN.

Details about the information needed from your referring provider are on the attached letter. Please insert information where requested and give the *Information Sheet for Referring Healthcare Providers* to your provider.

A UDN staff member will notify you when the information from your provider has been received. Once all materials are received, UDN review is expected to take about six to eight weeks.

Again, thank you for your interest in the UDN. Medical advances depend on individuals like you who volunteer as partners in medical discovery. More information about this new program is on line at https://undiagnosed.hms.harvard.edu.

The Undiagnosed Diseases Network Team

Description: *Information for healthcare providers*

Information for healthcare providers

Your patient has contacted the Undiagnosed Diseases Network (UDN) about participating in this program. Patient participants will be evaluated using the unique combination of scientific and medical expertise and resources. Participants must have a condition that has not been diagnosed following a thorough medical evaluation.

There is a stringent referral and review process. If your patient's case is accepted for UDN evaluation, the UDN will provide information from the evaluation to you and to your patient. You will be responsible for your patient's follow up medical care.

The following information is needed to determine your patient's eligibility:

1. Verification that this patient has a primary healthcare provider who will provide ongoing consultation to the UDN team and appropriate follow-up care for the patient if accepted into the UDN clinical research protocol.

2. A summary letter from you describing your patient's pertinent medical information, including

- When the undiagnosed condition was first noted;
- How it presented;
- The patient's current medical status;
- Treatments/medications tried and their effects.

3. Copies of reports and results of pertinent diagnostic tests, along with X-rays, MRI results, and other imaging records/studies.

4. Your office address, phone numbers and email address.

5. Your patient's mailing address.

Once all materials have been received, notification of receipt will be sent to the patient. UDN review is expected to take about six to eight weeks from the time the requested materials have been received. Once a decision has been made you will be notified within a week.

More information about this program is available online at https://undiagnosed.hms.harvard.edu.

Thank you for considering this opportunity to consult with the UDN on your patient. We appreciate your commitment to providing the best possible care for your patients in ways that help advance medical knowledge and discovery.

The Undiagnosed Diseases Network Team

Undiagnosed Diseases Network

Your institutional logo here

Description: *Records received*

Date:

Address:

Dear [patient]:

Thank you for your interest in the Undiagnosed Diseases Network (UDN). We are writing to inform you that your records have been received. Please allow 60 days from this time for us to review your records. Our team reviews all records, however, not all cases are accepted into the UDN.

When the team has made a decision about your case you will be notified.

Thank you again for your interest in our program.

Sincerely,

[name]
Principal Investigator at [Institution]

If you have any questions, please contact [Site Coordinator] at [phone number and email address].

Undiagnosed Diseases Network

Your institutional logo here

Description: *Incomplete records*

Date:

Address:

Dear [patient]:

Thank you for your interest in the Undiagnosed Diseases Network (UDN). We are writing to inform you that some of your records have been received, however, they are not complete. When you send records: (1) please make sure there are not multiple copies of the same report, (2) organize the reports by subspecialty and date seen (i.e. genetics, gastroenterology, neurology etc.), and (3) Do NOT send double-sided copies. This will greatly speed processing and timely review of your case. Specifically, we are requesting:

_____ Summary letter from healthcare provider
_____ Medical records
_____ Labs
_____ Biopsy reports and slides
_____ All imaging on CD, including brain
_____ Anesthesiology records
_____ Seizure medication levels within 30 days of the admission
_____ Birth/Neonatal records **for pediatric patients**
_____ Growth curves **for pediatric patients**
_____ Photos **for pediatric patients**
_____ Other:

Please allow 60 days from the time of our receipt of these materials for us to review your records. Our team reviews all records, however, not all cases are accepted into the UDN.

When the team has made a decision about your case you will be notified.

Thank you again for your interest in our program.

Sincerely,

[name]
Principal Investigator at [Institution]

If you have any questions or have difficulty requesting your medical records, please contact [Site Coordinator] at [phone number and email address].

Undiagnosed Diseases Network

Your institutional logo here

Description: *Partial application*

Date:

Address:

Dear [patient]:

You have expressed interest in the Undiagnosed Diseases Network (UDN) and have submitted a partial application. We requested medical records on [date], however, these records have not been received. Since three months has passed since our initial request, we assume that you are no longer interested in being evaluated in the network and will be removing your name from our active rolls.

Sincerely,

[name]
Principal Investigator at [Institution]

If you have any questions or have difficulty requesting your medical records, please contact [Site Coordinator] at [phone number and email address].

Cc: [referring provider]

Undiagnosed Diseases Network

Your institutional logo here

Description: *Acceptance Letter (Pediatric) to Healthcare provider*

Date:

Address:

Dear [healthcare provider]:

Your patient _____ [DOB] has applied to the Undiagnosed Diseases Network (UDN). After a stringent review process, your patient's case has been accepted for evaluation at [your institutional name; city, state]. Participants in the Network will be examined using the unique combination of scientific and medical expertise and resources at [your institution]. This evaluation will require a 2-5 day visit for inpatient and/or outpatient care. There is no need to order additional tests or procedures for the purpose of preparing your patient for this evaluation.

Travel, meals and lodging expenses will be covered for research participants according to our policies, to the extent allowed by law. A representative of the Network will contact the participant within the next few weeks. If the parents of your patient are separated or divorced we will need to receive the court paperwork specifying who is legally able to consent for the child to participate in medical research. We may also request blood specimens for DNA isolation from one or both parents prior to scheduling the child's visit to the [your institutional name].

Not all admissions will result in a diagnosis, but the evaluations should yield valuable information that medical researchers will use to: (1) help identify previously unrecognized rare diseases; (2) suggest new ways to treat and prevent common illnesses; and (3) determine promising options for continued medical research.

[Your institution] will provide information from the evaluation to you and to your patient as a part of this Network. You will be responsible for your patient's follow up medical care. Selected patients may be eligible for other ongoing research studies.

Thank you for consulting with the Undiagnosed Diseases Network on your patient. We appreciate your commitment to providing the best possible care for your patients in ways that help advance medical knowledge and discovery.

Sincerely,

[name]
Principal Investigator at [Institution]

If you have any questions, please contact [Site Coordinator] at [phone number and email address].

Cc: [participant]

Undiagnosed Diseases Network

Your institutional logo here

Description: *Acceptance Letter (Adult) to Healthcare provider*

Date:

Address:

Dear [healthcare provider]:

Your patient _____ [DOB] has applied to the Undiagnosed Diseases Network (UDN). After a stringent review process, your patient's case has been accepted for evaluation at [your institutional name; city, state]. Participants in the program will be examined using the unique combination of medical and scientific expertise and resources at [your institution]. This evaluation will require a 2-5 day visit for inpatient and/or outpatient care. There is no need to order additional tests or procedures for the purpose of preparing your patient for this evaluation.

Travel, meals and lodging expenses will be covered for research participants according to our policies, to the extent allowed by law. A representative of the Network will contact the participant within the next few weeks.

Not all admissions will result in a diagnosis. In addition to contributing to the diagnosis of individual participants, UDN evaluations should yield valuable information that medical researchers will use to: (1) help identify previously unrecognized rare diseases; (2) suggest new ways to treat and prevent common illnesses; and (3) determine promising options for continued medical research.

[Your institution] will provide information from the evaluation to you and to your patient as a part of this Network. You will be responsible for your patient's follow-up medical care. Selected patients may be eligible for other ongoing research studies.

Thank you for consulting with the Undiagnosed Diseases Network on your patient. We appreciate your commitment to providing the best possible care for your patients in ways that help advance medical knowledge and discovery.

Sincerely,

[name]
Principal Investigator at [Institution]

If you have any questions, please contact [Site Coordinator] at [phone number and email address].

Cc: [participant]

Undiagnosed Diseases Network

Your institutional logo here

Description: *Not Accept Letter to Healthcare provider*

Date:

Address:

Dear [healthcare provider]:

Your patient _____ [DOB] has applied to the Undiagnosed Diseases Network (UDN). After a stringent review process, your patient's case has not been accepted for evaluation. The Network's goals are to provide answers to patients with mysterious conditions that have long eluded diagnosis and to advance medical knowledge about rare and common diseases. The medical team bases its judgments on whether or not there is a reasonable chance to achieve these goals.

[Insert one of these or another scenario below depending on whether you have additional suggestions for work up and whether or not you would be willing to re-consider the patient if the suggested work up comes back normal]

Upon extensively reviewing [name of patient] records, we do not believe we can improve on the comprehensive work-up [he/she] already received.

MITO AND METABOLIC RECOMMENDATIONS
Members of the Board had a few thoughts for your consideration. Specifically, they suggested possible pursuit of a mitochondrial/metabolic evaluation including: plasma and urine amino acids, urine organic acids, plasma lactate, pyruvate, carnitine levels (free and total), leukocyte CoQ, and acylcarnitine profile.

For a complete metabolic and mitochondrial disease work up, you may consider contacting the Medical Genetics Laboratories at the Baylor College of Medicine: www.bcm.edu/geneticlabs/.

In addition, the Board recommended testing for Pitt-Hopkins Syndrome by looking at mutations or even a deletion of the *TCF4* gene. For testing center sites, visit GeneTests at: **http://www.ncbi.nlm.nih.gov/sites/GeneTests/**.

You may want to consider congenital disorders of glycosylation (CDG) testing at the Mayo Medical Laboratories. Results are reported as the mono-oligosaccharide/di-oligosaccharide transferrin ratio, the a-oligosaccharide/di-oligosaccharide transferrin ratio, the tri-sialo/di-oligosaccharide transferrin ratio, the apolipoprotein CIII-1/apolipoprotein CIII-2 ratio and the apolipoprotein CIII-0/apolipoprotein CIII-2 ratio. For more information on how to send this test, visit: http://www.mayomedicallaboratories.com/test-catalog/Overview/89891

In addition to the CDG testing, the Case Review Committee would like to suggest *PGM1* Full Gene Sequencing which is offered at multiple laboratories that can be identified at: **http://www.ncbi.nlm.nih.gov/sites/GeneTests/?db=GeneTests**.

In addition to the CDG panel, the Case Review Committee would like to recommend a Lysosomal Enzyme Screening. Information about this screening can also be found at the Emory Genetics Laboratory. Their website is: http://genetics.emory.edu/egl/.

If a diagnosis remains elusive after further work up we would be willing to reconsider Pt's name for admission to the UDN.

Again, thank you for consulting with the Undiagnosed Diseases Network on your patient. We appreciate your commitment to providing the best possible care for your patients in ways that help advance medical knowledge and discovery.

Sincerely,

[name]
Principal Investigator at [Institution]

If you have any questions, please contact [Site Coordinator] at [phone number and email address].

Cc: [participant]

Undiagnosed Diseases Network

Your institutional logo here

Description: *Welcome packet to patient*

Welcome to the Undiagnosed Diseases Network

We want your visit to the UDN clinical site at [institution] to be as comfortable and productive as possible. We have created a package of information for you to review prior to your admission so you will know what to expect. The following are included in the package:

1. **[institution] map and interior layout map of [institution]**
2. **Cafeterias [information]**
3. **Shuttle Service [information]**
4. **Airport transportation [information]**
5. **Security Procedures [information]**
6. **Patient Library [information]**
7. **Chapel [information]**
8. **Hospitality Services [information]**
9. **Gift Shops [information]**

For further information, please visit the following websites:
http://www

Please be sure to bring the following with you:

- **List of medications and dosages in their original containers**- Please discuss any questions you may have regarding medications and/or equipment with your admitting Nurse Practitioner or Physician Assistant.
- **Any assistive devices that you use daily**, e.g., wheelchair, cane, walker, braces
- **Complete list of current physicians and their contact information**, i.e., addresses and phone numbers

A UDN staff member will contact your shortly to make travel arrangements and book local hotel accommodations. An additional member of the UDN team will contact you to review the admission process and ask additional questions about your medical history.

In the event you are unable to keep your scheduled visit, please contact your admitting Clinical Site Coordinator. You may be asked to re-schedule your visit.

Please feel free to contact our team if you have any questions or concerns. Generally, phone messages and e-mails are responded to promptly. We are looking forward to meeting you at [your institution].

[team contact information- names, emails, phone numbers]

Undiagnosed Diseases Network

Your institutional logo here

Description: *Directions for remote blood draw*

DIRECTIONS: Remote Blood Draw

Date:

Address:

Dear [patient],

In order for your blood to be processed appropriately, please follow the directions below carefully. If possible, please arrange the blood draw through your healthcare provider.

Directions:

1) Draw [number] tubes of blood in the lavender topped tubes enclosed.
2) While talking to a UDN team member, review, sign, and date the consent form(s) enclosed and send the signed copy of the consent form(s) with your blood sample. **If we do not receive your signed consent form(s), we cannot process your blood.**
3) Ship the blood overnight (Monday-Thursday) to the UDN clinical site at [institution].
4) Call [number] or email [email] on the day that you ship the blood and provide the FedEx Tracking number.

Please contact us with any questions or concerns.

[name]
Principal Investigator at [Institution]

If you have any questions, please contact [Site Coordinator] at [phone number and email address].

Cc: [referring provider]

Undiagnosed Diseases Network

Your institutional logo here

Description: *No Diagnosis Following Evaluation*

Date:

Address:

Dear [patient]:

Thank you for your participation in the Undiagnosed Diseases Network (UDN) evaluation at [institution]. At this time, the clinical testing and evaluation phase is complete and has not yielded a definitive diagnosis. We will continue to pursue leads as they arise based upon ongoing research and new ideas that are generated among our expert consultants. We will keep the valuable information and biological samples collected during your visit in the hope that future research studies will be able to shed light on the medical problems that brought you to the UDN. If new prospects for investigation appear, we will contact you.

We very much appreciate your involvement in the UDN and your commitment to our joint goal of helping to advance medical knowledge, scientific discovery, and optimal care.

Sincerely,

[name]
Principal Investigator at [Institution]

If you have any questions, please contact [Site Coordinator] at [phone number and email address].

Cc: [referring provider]

APPENDIX 10: Suggested Sites for Testing

Condition/Test	Laboratory	Information	Website
Congenital disorders of glycosylation	Emory Genetics Lab and Mayo Clinic	-Analyze both N-glycosylation and O-glycosylation	http://genetics.emory.edu/egl/tests/?testid=1022 http://genetics.emory.edu/egl/tests/?testid=1341
Mucopolysachharidoses and oligosaccharidoses	University of Alabama Metabolic Disease Laboratory	-Urine screen	https://www.uab.edu/medicine/genetics/clinical-laboratories/metabolic-dise
Lysosomal storage diseases	Emory Genetics Lab	-Blood -New comprehensive enzyme panel (soon to be released)	
Sanfilippo syndrome	Greenwood Genetic Lab	-Blood -Most comprehensive enzyme panel for Sanfilippo syndrome	http://www.ggc.org/diagnostic/tests-costs/test-finder/test-finder.html?id=
Peroxisomal disorders	Kennedy Krieger Lab		http://www.kennedykrieger.org/patient-care/patient-care-laboratories/genet
Cerebrospinal fluid neurotransmitters	Medical Neurogenetics	-Customer service is modest and website is challenging to navigate	https://www.medicalneurogenetics.com/
Urine purines and pyrimidines	Baylor College of Medicine Medical Genetics Lab		https://www.bcm.edu/cancergeneticslab/test_detail.cfm?testcode=4220&show=

APPENDIX 11: Wrap-up Template

Name:
DOB:
Dates of Visit:
Primary Clinician:
Attending Physician:
Presenting Symptoms/short summary of case:
Testing/Recommendations by System (as applicable):

GASTROENTEROLOGY

Consultant: **Testing/Results:** **Recommendations:**

NUTRITION

Consultant: **Testing/Results:** **Recommendations:**

NEUROLOGY

Consultant: **Testing/Results:** **Recommendations:**

PULMOLOLOGY

Consultant: **Testing/Results:** **Recommendations:**

CARDIOLOGY

Consultant: **Testing/Results (EKG/ECHO):** **Recommendations:**

IMMUNOLOGY/INFECTIOUS DISEASE

Consultant: **Testing/Results:** **Recommendations:**

HEMATOLOGY

Consultant: **Testing/Results:** **Recommendations:**

ENDOCRINOLOGY

Consultant:
Testing/Results:
Recommendations:

DENTAL

Consultant:
Testing/Results:
Recommendations:

DERMATOLOGY

Consultant:
Testing/Results:
Recommendations:

OPHTHALMOLOGY

Consultant:
Testing/Results:
Recommendations:

AUDIOLOGY

Consultant:
Testing/Results:
Recommendations:

PHYSICAL MEDICINE & REHABILITATION

Consultant:
PT:
OT:
Speech:
Recommendations:

NEPHROLOGY

Consultant:
Testing/Results:
Recommendations:

RADIOLOGY

Test:
Results:
Test:
Results:

SPECIAL STUDIES

EMG/NCV: **Sleep Study:** **Metabolic Cart:** **Other:**

OTHER

Consultant: **Testing/Results:** **Recommendations:**

SEND OUT AND PENDING TEST RESULTS

Test Name	Date Sent	Date Resulted Rev'd	Testing Lab	Result/Interp	Normal Range	Copies sent (date and person)
CSF studies						
CDT/N-glycan screen						
Urine Oligosaccharides						
WBC CoQ level						
Lysosomal Screen						
WBC Buffy Coat						
Urine Sulfocysteine						
MitoGEN						
POLG						
Muscle mtDNA Content						
Muscle ETC Enzymology & CoQ content						

You will be contacted by the UDN for surveys to tell us about your experience. The first contact will be a week or so after you get home.

APPENDIX 12: Patient Follow-up Surveys

1-7 days post visit survey. In addition to the answers below, there will also be "refused to answer" and "not applicable" checkboxes.

INTERPERSONAL PROCESSES OF CARE SURVEY: SHORT FORM (IPC-18)

The next questions are about your experiences talking with your doctor(s) at ____[*clinic name*] over the past 12 months. If you see more than one doctor at that clinic, please tell us, on average, how often they did the following:

	Never	Rarely	Sometimes	Usually	Always
1. How often did doctors speak too fast?	1	2	3	4	5
2. How often did doctors use words that were hard to understand?	1	2	3	4	5
6. How often did doctors really find out what your concerns were?	1	2	3	4	5
7. How often did doctors let you say what you thought was important?	1	2	3	4	5
8. How often did doctors take your health concerns very seriously?	1	2	3	4	5
9. How often did doctors explain your test results such as blood tests, x-rays, or cancer screening tests?	1	2	3	4	5
10. How often did doctors clearly explain the results of your physical exam?	1	2	3	4	5
15. How often did you and your doctors work out a treatment plan together?	1	2	3	4	5
16. If there were treatment choices, how often did doctors ask if you would like to help decide your treatment?	1	2	3	4	5
19. How often were doctors concerned about your feelings?	1	2	3	4	5
20. How often did doctors really respect you as a person?	1	2	3	4	5
21. How often did doctors treat you as an equal?	1	2	3	4	5
24. How often did doctors pay less attention to you because of your race or ethnicity?	1	2	3	4	5

Q25. How often did you feel discriminated against by doctors because of your race or ethnicity?	1	2	3	4	5

Open ended questions:

Q26 – What part/s of the UDN experience worked particularly well for you?

Q27 – What part/s of the UDN experience did not work well?

Q28 – What could we do to make the UDN experience better?

APPENDIX 13: Research Inventory Form

Sample ID:

Candidate discovery progress:

1. Is analysis of next generation sequencing (NGS) data complete or ongoing?
2. Names of collaborators or groups
3. Candidate gene(s) or causative element(s) found for phenotype? (enter name of gene or indicate “no”)
    a. Was result Sanger confirmed?
    b. Was result confirmed in a CLIA-compliant clinical facility?
        i. If so, which one?
    c. Was result returned to proband/physician?
    d. Has the result been published?
        i. Citation (if any)
        ii. If not published yet, do you expect to publish these data?

Omics and Models:

1. Please indicate any work done on the following:
    a. Glycomics
    b. Lipidomics
    c. Metabolomics
    d. Energetics
    e. Drophilia model
    f. Mouse model
    g. Yeast model
    h. Zebrafish model
    i. Additional collaboration

Financial data:

1. Has this data been used in any grant applications?
    a. If grants have been awarded, please name the grant and the amount.
2. How else do you plan to use this data? (eg. In-house database)

APPENDIX 14: Feature Request Form

Feature Requester Name(s):

Feature Requester Contact Information:

Name:

Institution:

Email:

Detailed Description of Feature:

Please provide a description of the feature and types of users that will interact with the feature and how they will access and use the feature. Describe the workflow from the perspective of each of these users. Screen shots of the Gateway where the new feature will exist are also preferable.

Importance to the UDN and Justification:

□ Critical

□ Major

□ Minor

Justification:

APPENDIX 15: UDN Data Sharing and Use Agreement

UDN DATA SHARING AND USE AGREEMENT

This Undiagnosed Diseases Network ("UDN") Data Sharing and Use Agreement (the "**Agreement**"), effective on _____ (the "**Effective Date**") is entered into by and among the UDN Participating Institutions listed on Exhibit 1 to this Agreement, each of which will be known as a "**Party**" and collectively as "**Parties**" to this Agreement.

RECITALS

WHEREAS, each non-NHGRI Party has received an award from the NIH (or is the recipient of a subaward) to participate in the UDN and to assist in the conduct of the UDN Study;

WHEREAS, the Parties recognize that to achieve the objectives of the UDN, the Parties will need to share Study Data with one another and to maintain the Study Data in a centralized data repository; and

WHEREAS, the Parties recognize that there must be mutual agreement as to the permitted uses and disclosures of the Study Data.

Now therefore, the Parties agree as follows:

I. Definitions

A. Capitalized Terms. Capitalized terms shall have the meaning as set forth herein or in this Section I.A.

1. Applicable Law shall mean the applicable legal and regulatory requirements, principles, and standards set forth and espoused in: (i) HIPAA; (ii) the Federal Policy for the Protection of Human Subjects, also known as the Common Rule, as applicable; (iii) the U.S. Food and Drug Administration's research laws, as applicable, including without limitation the regulations contained in 21 C.F.R. Parts 50, 54 and 56, 312, 314, 601, 812 and 814 as amended or augmented from time to time; (iv) Department of Health and Human Services Guidance on "Financial Relationships and Interests in Research Involving Human Subjects: Guidance for Human Subject Protection," Federal Register, Vol. 69, No. 92, p. 26393 (May 12, 2004); (v) state and federal fraud and abuse laws, including but not limited to Stark and the Anti-kickback Act; (vi) the Federal Privacy Act of 1974; (vii) rules and regulations governing the application for, receipt of, and use of federal research Grants and contracts; and (viii) all other federal, state, or local laws, regulations, guidances, or other requirements governing the conduct of the UDN Study and/or the data sharing activities as contemplated herein.

2. Central Database shall have the meaning set forth in Section I.A.4.

3. Confidential Business Information shall have the meaning set forth in Section V.C.

4. Coordinating Center shall mean the UDN Participating Institution that has principal responsibility within the UDN for establishing and maintaining a central database for the Study Data required by the UDN Protocol to be forwarded to the Coordinating Center (the "**Central Database**") and coordinating the sharing of UDN Study Data as part of the UDN Study. The functions of the Coordinating Center will be performed by Harvard Medical School and those additional Parties listed on Exhibit 1 that are recipients of a subaward from Harvard Medical School. With respect to its activities as Coordinating Center for the UDN Study, Harvard Medical School is not a Covered Entity or a Business Associate under HIPAA.

5. De-Identified Study Data shall have the meaning set forth in Section III.B.3.

6. Evaluation Site shall have the meaning set forth in Section II.C.2.

7. Health Care Provider shall mean a health care provider that is not a UDN Participating Institution and that discloses Protected Health Information about a Participating Human Subject, as permitted by the UDN Tier 1 ICA Form or the UDN Tier 2 ICA Form, to the UDN.

8. HIPAA shall mean the Administrative Simplification section of the Health Insurance Portability and Accountability Act of 1996 (Public Law 104-191), the amendments thereto by Title XIII of the American Recovery and Reinvestment Act of 2009 also known as the Health Information Technology for Economic Clinical Health Act, and their respective implementing regulations as amended from time to time.

9. Intake Site shall have the meaning set forth in Section II.C.1.

10. Participating Human Subject shall mean each individual who is enrolled in the UDN Study. For the avoidance of doubt, individuals who only participate in the Tier 1 Phase of the UDN Study are Participating Human Subjects.

11. Principal Investigator shall mean each UDN Investigator listed on Exhibit 2 to this Agreement.

12. Protected Health Information ("**PHI**") shall have the meaning set forth in HIPAA.

13. Publication shall have the meaning set forth in Section VI.A.

14. Recipient shall mean a Party, other than the Coordinating Center, that receives UDN Study Data from another Party under this Agreement.

15. Required Data Sharing ICA Form Elements shall have the meaning set forth in Section III.B.

16. Required Tier 1 Data Sharing ICA Form Elements shall have the meaning set forth in Section III.A.

17. Sequencing Core shall mean each of the UDN Participating Institutions listed on Exhibit 1 that has principal responsibility for generating genetic sequence data for Participating Human Subjects as part of the UDN Study.

18. Study Data shall have the meaning set forth in Section II.C.2.

19. Tier 1 Phase shall have the meaning set forth in Section II.C.1.

20. Tier 1 Study Data shall have the meaning set forth in Section II.C.1.

21. Tier 2 Phase shall have the meaning set forth in Section II.C.2.

22. Tier 2 Study Data shall have the meaning set forth in Section II.C.2.

23. UDN Central IRB shall mean the IRB of the National Human Genome Research Institute ("**NHGRI**"), which is responsible for the review, approval and on-going oversight of the UDN Study. NHGRI is not a HIPAA Covered Entity.

24. UDN Clinical Site shall mean any clinical site where Participating Human Subjects are evaluated as part of the UDN Study.

25. UDN Tier 1 ICA Form shall mean the combined informed consent and HIPAA Authorization Form that each Participating Human Subject in the UDN Study must sign prior to participating in the Tier 1 Phase of the UDN Study. The UDN Tier 1 ICA Form is incorporated into the UDN Protocol.

26. UDN Tier 2 ICA Form shall mean the combined informed consent and HIPAA Authorization form that each Participating Human Subject in the UDN Study must sign prior to participating in the Tier 2 Phase of the UDN Study. The UDN Tier 2 ICA Form is incorporated into the UDN Protocol.

27. UDN Investigator shall mean a researcher at a UDN Participating Institution that assists in the conduct of the UDN Study.

28. UDN Participating Institution shall mean the Coordinating Center, each Sequencing Core, each of the UDN Clinical Sites, and any other institution in the United States engaged in the conduct of the UDN Study. Each UDN Participating Institution, other than NHGRI, has received an award from the NIH (or a subaward from another UDN Participating Institution) to participate in the UDN.

29. UDN Protocol shall mean the research protocol, attached as Exhibit 3 to this Agreement, for the UDN Study, and any amendments thereto that may be made and approved by the UDN Central IRB during the course of the UDN Study.

30. UDN Study shall mean the research study described in the UDN Protocol.

31. Variant Results shall have the meaning set forth in Section VI.B.1.

II. Background and Purpose

A. **Background**. The National Institutes of Health ("**NIH**") Common Fund's UDN is a program established by NIH to promote cross-disciplinary approaches to identifying, diagnosing, and treating rare non-diagnosed/differentiated diseases by academic centers located in the United States. The objectives of the UDN are to: (1) improve the level of diagnosis and care for patients with undiagnosed diseases through the development of common protocols designed by an enlarged community of investigators; (2) facilitate research into the etiology of undiagnosed diseases, by collecting and sharing standardized, high-quality clinical and laboratory data including genotyping, phenotyping, and documentation of environmental exposures; and (3) create an integrated and collaborative research community across multiple clinical sites and among laboratory and clinical investigators prepared to investigate the pathophysiology of these new and rare diseases and share this understanding to identify improved options for optimal patient management. To achieve these objectives, the Parties wish to share certain Study Data and work collaboratively to enable the identification of eligible individuals to participate in the Tier 2 Phase in an effort to diagnose their conditions and to conduct subsequent research on undiagnosed diseases.

B. **UDN Participating Institutions**. The UDN is comprised of the UDN Participating Institutions set forth on Exhibit 1, as may be amended from time to time in accordance with this Agreement. In order to be considered a UDN Participating Institution, an Institution must have (a) received an NIH award (or a subaward from another UDN Participating Institution) to participate in the UDN Study (except for NHGRI), and (b) caused an authorized representative to execute this Agreement and provide a copy of the signature page to each Party. The Coordinating Center is authorized to amend Exhibit 1 to reflect the addition of a new UDN Participating Institution upon the Coordinating Center's receipt of notice from the NIH of the addition of the institution to the UDN and the Coordinating Center's receipt of a copy of this Agreement signed by an authorized representative of the new UDN Participating Institution. The grant reference numbers for each non-NHGRI UDN Participating Institution are set forth in Exhibit 1.

Should there be any change in the designation of any Principal Investigator for a Party, including additions or replacements, Exhibit 2 shall be deemed amended upon receipt of a letter signed by the authorized representative of said Party when a copy has been sent to each other Party.

C. **UDN Study Tiers**. The UDN Study is differentiated into two tiers, as further described in the UDN Protocol.

1. During the first tier phase of the UDN Study (the "**Tier 1 Phase**") and only after the Participating Human Subject has completed a UDN Tier 1 ICA Form, (1) the Participating Human Subject will submit to the Coordinating Center demographic and medical information; (2) the Participating Human Subject's Health Care Providers will submit to the Coordinating Center additional medical information as well as a Health Care Provider referral letter summarizing medical history and other pertinent clinical information and (3) the Coordinating Center will assign such information to a UDN Clinical Site (the "**Intake Site**") that will be responsible for evaluating whether the Participating Human Subject is eligible to participate in the Tier 2 Phase (as defined below) of the UDN Study. In connection with assessing eligibility for enrollment in the Tier 2 Phase, the applicable Intake Site will evaluate the information submitted to the Coordinating Center and will also collect additional medical records, laboratory results, radiographic and pathology reports and any other information deemed pertinent by the Intake Site consistent with the UDN Protocol and the UDN Tier 1 ICA Form (collectively, the "**Tier 1 Study Data**").

2. During the second phase of the UDN Study (the "**Tier 2 Phase**"), and only after the Participating Human Subject has signed a UDN Tier 2 ICA Form, the applicable Intake Site or other UDN Clinical Site to which a Participating Human Subject is assigned ("**Evaluation Site**") will perform a clinical evaluation of the applicable Participating Human Subject, pursuant to the UDN Protocol, may collect data from the evaluation and from post-evaluation surveys of the Participating Human Subject, and may request other UDN Participating Institutions, including without limitation, a Sequencing Core, to perform additional tests or analysis pertaining to the Participating Human Subject (all resulting data, collectively, the "**Tier 2 Study Data**" and together with the Tier 1 Study Data, the "**Study Data**").

III. ICA Forms

A. **Required Content of the UDN Tier 1 ICA Form**. The UDN Tier 1 ICA Form, once approved by the UDN Central IRB, will be attached and incorporated into this Agreement as Exhibit 4. The UDN Tier 1 ICA Form may be amended, subject to UDN Central IRB approval, provided that the elements set forth in Sections III.A.1-3 are included (the "**Required Tier 1 Data Sharing ICA Form Elements**"). Specifically, the Required Tier 1 Data Sharing UDN ICA Form Elements inform Participating Human Subjects that:

1. Tier 1 Study Data pertaining to them, including Tier 1 Study Data containing PHI, may be disclosed to, and maintained by, the Coordinating Center.

2. Tier 1 Study Data pertaining to them, including Study Data containing PHI, will be disclosed to one or more UDN Clinical Sites in connection with efforts to determine whether such Participating Human Subject is eligible to participate in the Tier 2 Phase.

3. Tier 1 Study Data pertaining to them may be maintained by the Coordinating Center and by one or more UDN Clinical Sites.

B. **Required Content of the UDN Tier 2 ICA Form**. The UDN Tier 2 ICA Form, once approved by the UDN Central IRB, will be attached and incorporated into this Agreement as Exhibit 5. The UDN Tier 2 ICA Form may be amended, subject to UDN Central IRB approval, provided that the elements set forth in Sections III.B.1-3 are included (the "**Required Data Sharing ICA Form Elements**"). Specifically, the Required Data Sharing ICA Form Elements inform Participating Human Subjects that:

1. Study Data pertaining to them, including Study Data containing PHI, may be disclosed to, and maintained by, the Coordinating Center.

2. Study Data pertaining to them, including Study Data containing PHI, (a) will be disclosed by the Coordinating Center to UDN Participating Institutions and (b) may be disclosed by one UDN Participating Institution to another UDN Participating Institution, in connection with efforts to diagnose that Participating Human Subject or to identify commonalities with other Participating Human Subjects that might ultimately assist with the diagnosis or treatment of the Participating Human Subject or other Participating Human Subjects.

3. Study Data pertaining to them that has been de-identified in accordance with a methodology set forth in HIPAA ("**De-Identified Study Data**") may be shared with researchers at UDN Participating Institutions that are not specifically engaged on the UDN Study and with non-UDN third parties so that these recipients can conduct research, which may or may not be related to the objectives of the UDN Study.

C. **Revocation**. In the event that a Participating Human Subject submits a revocation of his/her UDN Tier 1 ICA Form and/or his/her UDN Tier 2 ICA Form to a UDN Participating Institution other than the Coordinating Center, such UDN Participating Institution will immediately notify the Coordinating Center so that the Coordinating Center can take all necessary steps to effectuate revocation. The Coordinating Center will notify all other UDN Participating Institutions of the Participating Human Subject's revocation. Each UDN Participating Institution shall comply with any required return or destruction of Study Data in its possession containing PHI per the instructions from the Coordinating Center and the Coordinating Center will take the necessary steps to delete and destroy Study Data containing PHI from the Central Database consistent with the Participating Human Subject's revocation.

IV. Permitted Uses and Disclosures of Study Data

A. **Uses and Disclosures of Study Data Within the UDN.** The Study Data may be used and disclosed by a UDN Participating Institution solely for the purposes permitted by, and in accordance with, the UDN Protocol and the UDN Tier 1 and Tier 2 ICA Forms.

1. The Intake Site will forward its summary of findings, along with any additional information relevant to the decision regarding a Participating Human Subject's eligibility for Tier 2 Phase participation, to the Coordinating Center following completion of the Tier 1 Phase for the applicable Participating Human Subject.

2. Each UDN Participating Institution will be responsible to provide all elements of Tier 2 Study Data that it collects in the course of the UDN Study to the Coordinating Center for inclusion in the Central Database. In general, it is expected that each Evaluation Site will make reasonable efforts to transfer Tier 2 Study Data pertaining to a Participating Human Subject to the Coordinating Center within thirty (30) days following completion of the Participating Human Subject's clinical evaluation during the Tier 2 Phase, and to update such information promptly upon receipt of related test results or additional Tier 2 Study Data. Subject to the foregoing, a Participating Human Subject's Evaluation Site may transfer directly such portions of his/her Study Data to another UDN Participating Institution (such as a Sequencing Core) as are necessary for the performance of testing or analysis of such Study Data requested by the Evaluation Site during the Tier 2 Phase of the UDN Study. The results of such testing or analysis shall also constitute Study Data, and the UDN Participating Institution performing such testing or analysis shall transfer the resulting Study Data to the Evaluation Site, which will then transfer them to the Coordinating Center.

3. Subject to the terms of this Agreement, the Coordinating Center will provide the UDN Participating Institutions with access to the Tier 2 Study Data it maintains to enable them to (a) assist in the diagnosis of a Participating Human Subject; (b) identify distinguishing, unique or common clinical or biological themes across Participating Human Subjects that might ultimately assist with diagnosis or treatment of one or more Participating Human Subjects and (c) conduct research on the etiology of undiagnosed diseases, consistent with the procedures and processes set forth in the UDN Protocol and the terms of this Agreement. The Coordinating Center may also use Study Data to perform analyses in support of the UDN Study, including quality control measurement and providing NIH and other institutions with process and outcome measures of the functioning of the UDN Study.

4. UDN Participating Institutions other than the Coordinating Center may also generate and transfer to each other Study Data that, pursuant to the UDN Protocol, does not need to be submitted to the Coordinating Center; provided that each UDN Participating Institution transferring the Study Data or receiving the Study Data shall be obligated to treat such Study Data in a manner that complies with the terms of this Agreement. The Coordinating Center shall not be responsible for Study Data maintained by or transferred by or among other UDN Participating Institutions.

5. All transfers of Study Data, whether to or from the Coordinating Center, or among UDN Participating Institutions other than the Coordinating Center, will be done via encrypted and authenticated data transfer. Each UDN Participating Institution agrees to employ technical,

physical and other safeguards to maintain the Study Data in a secure and confidential manner that prevents uses or disclosures of the Study Data not permitted by this Agreement. For paper records, safeguards include, but are not limited to, locked file cabinets or continual physical presence in a room that locks. For electronic records, safeguards include authentication of each Study Data access, explicit authorization of each Study Data access, end-to-end encryption at-rest and in-transit, and an audit trail of such access. Without limiting the foregoing, each UDN Participating Institution agrees that it shall protect, store and secure all Study Data made available to it as part of the UDN Study, whether received directly or through the Central Database, using HIPAA-compliant systems and controls. No UDN Participating Institution shall be responsible for the security of Study Data held or maintained by another UDN Participating Institution. Access to the Central Database, and transfer of Study Data by one UDN Participating Institution to another Recipient, will be limited only to those authorized named individuals identified by each UDN Participating Institution in a written notice sent to the Coordinating Center by a designated official of the UDN Participating Institution. The Coordinating Center, as administrator of the Central Database, may take such actions as it believes necessary or appropriate for proper administration of the Central Database, at its sole reasonable determination, including without limitation, determining and administering processes for issuance of accounts and passwords for authorized access to the Central Database; determining and issuing standard operating procedures for security incident response within the UDN; and restricting or suspending any Recipient's or UDN Investigator's or other individual's access to Study Data in the Central Database. Failure of a UDN Participating Institution or authorized user of the Central Database to comply with the information security plan and the standard operating procedures regarding incident response as may be issued and modified from time to time by the Coordinating Center, and with the terms of this Agreement, may give rise to restriction or suspension of access to the Central Database.

B. **Uses and Disclosures of Study Data Outside of the UDN**. The Coordinating Center may provide De-Identified Study Data in the database of Genotypes and Phenotypes (dbGaP) of the National Center for Biotechnology Information or other research data repository for further sharing with researchers from UDN Participating Institutions who are not themselves personally engaged in the UDN Study and with non-UDN third parties, under specific rules established by the NIH and consistent with Applicable Law.

C. **Cloud Service or Third Party Provider.** The Coordinating Center may engage one or more cloud service or other third party providers to host the Central Database, and may provide third party service providers access to the Central Database, as may be necessary from time to time for maintenance, quality control, and other administration and management of the Central Database. The Coordinating Center may take such measures as it deems necessary from time to time for proper management and security of the Central Database: this may include downtime for servicing or maintenance purposes, and may include imposition of access restrictions.

D. **Not Expressly Permitted; Prohibited.** For the avoidance of doubt, notwithstanding anything to the contrary in this Article IV, a use or disclosure of Study Data is not permitted unless it is expressly permitted by the UDN Protocol and, as applicable, the UDN Tier 1 and/or Tier 2 ICA Forms.

E. **Principal Investigator Obligations**. Study Data provided to a UDN Participating Institution containing PHI will be used only by the Recipient's Principal Investigator and those individuals under his/her direct supervision in accordance with this Agreement, the UDN Protocol, and, as applicable, the UDN Tier 1 and/or Tier 2 ICA Forms. The Principal Investigator is responsible for informing the individuals under his/her supervision of the provisions and restrictions contained herein and to secure documentation of their agreement to abide by such provisions and restrictions before providing access to the Study Data.

F. **No Contact**. Each Recipient agrees that it will not use the Study Data to contact or attempt to contact Participating Human Subjects about whom the Study Data pertains, except as expressly permitted by the UDN Protocol and**,** as applicable, the UDN Tier 1 and/or Tier 2 ICA Forms. In the event that a Participating Human Subject must be contacted, such contact will be by the Coordinating Center or applicable Evaluation Site. Each Recipient further agrees that it will not seek to re-identify any individual whose information is included within De-identified Study Data.

G. **Study Data Retention**. Each UDN Participating Institution will maintain all Study Data it collects, at a minimum, for such time as is required by the UDN Protocol.

V. Compliance

A. **Generally**. Each Party agrees to comply with Applicable Law that is pertinent to such Party.

B. **IRB**. The Parties agree to comply with all directives from the UDN Central IRB.

C. **Confidentiality**. In connection with the UDN Study, a UDN Participating Institution may provide another UDN Participating Institution with certain of the disclosing Party's non-public business, technical, financial or strategic information, other than Study Data, that it marks as confidential or indicates is confidential by written notice given to the receiving Party within fifteen (15) days following disclosure ("**Confidential Business Information**"). Each UDN Participating Institution shall maintain the confidentiality of all Confidential Business Information provided to it by another UDN Participating Institution, and shall not disclose such information to any third party, for a period of five (5) years after its receipt of such Confidential Business Information. The receiving Party shall not be bound by confidentiality obligations hereunder with respect to Confidential Business Information if (i) such Confidential Business Information is or becomes part of the public domain, except through breach of this Agreement by the receiving Party; (ii) such Confidential Business Information was in the receiving Party's

possession prior to the time of disclosure by or on behalf of the disclosing UDN Participating Institution; (iii) such Confidential Business Information becomes available to the receiving Party from a third party who is not legally prohibited from disclosing such Confidential Business Information; (v) the receiving Party can demonstrate by clear and convincing written evidence such information was developed by or for the receiving Party independently of the disclosure of the Confidential Business Information to the receiving Party; or (vi) disclosure is required by Applicable Law and the receiving Party, to the extent practicable, provides prior written notice to the disclosing Party of such legal requirement so that the disclosing Party may seek a protective order or similar remedy.

VI. Intellectual Property

A. **Publication**. In all oral presentations or written publications (each, a "**Publication**") involving the analysis of Study Data, UDN Investigators will acknowledge the UDN in a form of acknowledgement specified in the UDN Publication Policy. The Parties will abide by the UDN Publication and Presentation Policy and Procedures as defined by the UDN Publications Working Group and ratified by the UDN Steering Committee, attached as Exhibit 6 to this Agreement. The UDN Publications Working Group may from time to time propose, and the UDN Steering Committee may from time to time ratify, modifications to UDN Publication and Presentation Policy and Procedures, with material changes to be emailed to each Party's email address as provided on the signature pages to this Agreement. No Publication will contain PHI.

B. **Commercial Purposes; Prohibited**. The Parties agree that Study Data will not be used for commercial purposes, including selling, advertising, commercial screening, or transferring the Study Data to a third party for commercial purposes.

1. Notwithstanding anything to the contrary in Section VI.B, De-identified Study Data involving the use of variant information, including allele frequency and other derived characteristics (collectively "**Variant Results**") generated during the term of this Agreement by a Sequencing Core for any previously diagnosed, subsequently diagnosed, and still undiagnosed diseases, may be used by any such Sequencing Core as part of commercial sequencing services it offers to third parties; provided, however that no Party providing said services shall secure, nor attempt to secure or apply, a patent or other intellectual property right, including any trade secret protection, to the use of the Variant Results. Each Sequencing Core agrees to the foregoing proviso for itself only.

C. **Intellectual Property**. Subject to the rights of the Parties to use and share Study Data pursuant to this Agreement and the UDN Protocol, each Party shall own the portion of original Study Data it collects under, and in accordance with the terms of, its NIH award (or subaward). Inventorship of any invention that is either (i) conceived or (ii) first actually reduced to practice in the performance of the UDN Study will be determined according to U.S. patent laws, and ownership shall follow inventorship. Joint inventions will be owned jointly. Other results

obtained from the uses permitted by this Agreement will be governed by the NIH Grants Policy Statement, section 8.2 "Availability of Research Results: Publications, Intellectual Property Rights, and Sharing Research Resources" (see http://grants.nih.gov/grants/policy/nihgps_2013/nihgps_ch8.htm).

VII. Certification

The Parties certify and affirm that the contents of any statements made herein are truthful and accurate and that they are authorized by their institution to agree to adhere to the principles and policies specified outlined in this document.

VIII. Term and Termination

A. **Term.** The term of this Agreement shall commence on the Effective Date and continue in full force and effect until terminated in accordance with Section VIII.B.

B. **Termination of the Agreement.**

1. This Agreement may be terminated upon the mutual written agreement of all Parties.

2. This Agreement shall terminate automatically upon completion of the UDN Study.

3. In the event that the UDN Central IRB requires that the UDN Study terminate early for any reason, this Agreement will terminate immediately; provided that the Parties will follow any UDN Central IRB requirements regarding the orderly termination for the protection of Participating Human Subject safety.

C. **Termination of a Party.**

1. In the event of the termination or expiration of any Party's UDN award from NIH (or subaward, if applicable), such Party shall be automatically terminated from this Agreement and Exhibit 1 shall be deemed modified to remove such Party's name from the list of UDN Participating Institutions.

2. Upon notice of termination of any Party to this Agreement, the UDN Principal Investigator of the terminating Party shall transfer Study Data for inclusion in the Central Database to the Coordinating Center in accordance with Section IV.A.2.

IX. Liabilities

A. **Liabilities**. Each Party shall be responsible for its negligent acts or omissions and the negligent acts or omissions of its employees, officers, or directors, to the extent allowed by law; provided, however, that any Party that is an agency of the United States Government, may be liable only to the extent as provided under the Federal Tort Claims Act (28 U.S.C. Chapter 171).

No indemnification for any loss, claim, damage, or liability is intended or provided by any Party under this Agreement.

B. **Limitation**. The Study Data are provided as a service to the research community. THEY ARE BEING SUPPLIED TO RECIPIENT WITH NO WARRANTIES, EXPRESS OR IMPLIED, INCLUDING WITHOUT LIMITATION ANY WARRANTY OF MERCHANTABILITY OR FITNESS FOR A PARTICULAR PURPOSE. NO WARRANTY WITH RESPECT TO THE CENTRAL DATABASE IS PROVIDED, INCLUDING WITHOUT LIMITATION, ANY UPTIME WARRANTY. The Parties make no representations that the use of the Study Data will not infringe any patent or proprietary rights of third parties.

X. Miscellaneous

A. **Amendments and Modification.** This Agreement may only be amended, modified or supplemented by an agreement in writing signed by each Party, except as specified in Sections II.B., VI.A. and VIII.C. above. Amendments to the UDN Protocol will be deemed incorporated as modifications to Exhibit 3 (and Exhibits 4 and 5, as applicable) upon approval by the UDN Central IRB.

B. **Assignment**. No Party will assign or transfer any rights or obligations under this Agreement without the prior written consent of each of the other Parties, and only if permitted under the UDN Protocol, the UDN Tier 1 and Tier 2 ICA Forms, the applicable NIH award (or subaward), and Applicable Law.

C. **Entire Agreement.** This Agreement and any Exhibits attached hereto constitute the entire agreement among the Parties and supersede all prior communications, representations, or agreements, either verbal or written among the Parties with respect to the subject matter hereof. Notwithstanding the foregoing, this Agreement shall not supersede the terms of any NIH notice of grant award. Each Party confirms that it is not relying on any representations or warranties of any other Party except as specifically set forth herein.

D. **Independent Contractors; Relationship of the Parties.** This Agreement shall not be deemed to create any partnership, joint venture, or agency relationship between or among the Parties. Each Party shall act hereunder as an independent contractor and its agents and employees shall have no right or authority under this Agreement to assume or create any obligation on behalf of or in the name of, the other Parties. All persons employed by a Party shall be employees of such Party and not of the other Parties, and all costs and obligations incurred by reason of any such employment shall be for the account and expense of such Party.

E. **Notice.** All notices and other communications required or permitted to be given pursuant to this Agreement shall be in writing and shall be deemed sufficiently given if personally delivered or sent by mail, recognized delivery service, electronic mail, postage prepaid, or by

facsimile transmission with mail confirmation. Such communications shall be given to each Party at the addresses listed on each signature page.

F. **Severability.** All agreements and covenants contained herein are severable, and in the event any of them shall be held to be invalid by any competent court, this Agreement shall be interpreted as if such invalid agreements or covenants were not contained herein.

G. **Survivability.** All causes of action accruing to any Party under this Agreement shall survive termination. Each provision of this Agreement that would by its nature or terms survive any termination of the Agreement shall survive, including without limitation Articles I, IV.D.-G., V, VI.B., VI.C, VIII.B.3, VIII.C.2, IX and X.

H. **Waiver.** Failure by any Party to insist upon strict performance of any provision herein by any Party shall not be deemed a waiver by such Party of its rights or remedies, or a waiver by it of any subsequent default by such other Party, and no waiver shall be effective unless it is in writing and duly executed by the Party entitled to enforce the provision being waived.

I. **Signatures**. The Parties individually and collectively have caused this Agreement to be executed by their duly authorized representatives as of the dates below on their respective signature page. This Agreement may be executed in one or more counterparts, all of which shall be considered one and the same. The Parties agree that execution of this Agreement by exchanging facsimile, PDF, or e-Signature signatures shall have the same legal force and effect as the exchange of original signatures.

E-signature, for purposes of this Section XI.I. shall mean signature that consists of one or more letters, characters, numbers or other symbols in digital form incorporated in, attached to or associated with the electronic document, that (i) is unique to the person making the signature; (ii) the technology or process used to make the signature is under the sole control of the person making the signature; (iii) the technology or process can be used to identify the person using the technology or process; and (iv) the electronic signature can be linked with an electronic document in such a way that it can be used to determine whether the electronic document has been changed since the electronic signature was incorporated in, attached to or associated with the electronic document.

APPENDIX 16: Publications and Research Reference Sheets

TITLE: Grant Applications (which utilize UDN data and resources)

1. RESPONSIBLE PERSONNEL:

1.1. Investigators: Investigators at the UDN Clinical Sites, Cores, Coordinating Center, and NIH.
1.2. Coordinating Center Project Manager: Individual at the Coordinating Center who communicates with the Clinical Sites, Cores, Coordinating Center, and NIH team members to complete project-related activities.
1.3. Coordinating Center Executive Director: Individual at the Coordinating Center who directs project-related activities of the UDN.
1.4. Steering Committee: Committee made up of UDN Investigators that decides on the priorities and order of business of the UDN.

2. PROCEDURE:

2.1. Investigators submit a concept form (link: https://hms.az1.qualtrics.com/SE/?SID=SV_3CyZOKOuiEvnCx7) for a grant application that will rely on UDN data to the Coordinating Center Project Manager.
2.2. The Coordinating Center Project Manager updates the grant application log in Box.com.
2.3. The Coordinating Center Project Manager submits the grant application request to the Coordinating Center Executive Director.
2.4. The Coordinating Center Executive Director notifies the Coordinating Center Project Manager of the grant application Steering Committee presentation date.
2.5. The Coordinating Center Project Manager notifies the Investigator of the grant application presentation date.
2.6. The Investigators present at the Steering Committee meeting.
2.7. The Steering Committee votes on the application.
2.8. The Coordinating Center Project Manager updates the grant application log with the decision.
    2.8.1. If the grant application is accepted, the Investigators notify the Coordinating Center Project Manager and the Coordinating Center Project manager updates the grant application log.

Version: 1
Effective Date: May 1, 2015
Last Reviewed Date: June 9, 2015

TITLE: Internal UDN Research Concept Sheets

1. RESPONSIBLE PERSONNEL:

1.1. Investigators: Investigators at the UDN Clinical Sites, Cores, Coordinating Center, and NIH.
1.2. Coordinating Center Project Manager: Individual at the Coordinating Center who communicates with the Clinical Sites, Cores, Coordinating Center, and NIH team members to complete project-related activities.
1.3. Publication and Research Committee Co-Chairs: Individuals who lead the Publications and Research Committee.
1.4. Publication and Research Committee Members: Individuals who participate in the Publications and Research Committee.
1.5. Coordinating Center Administrator: Individual at the Coordinating Center who handles the administrative tasks for the Coordinating Center.

2. PROCEDURE:

2.1. Investigators submit a research concept sheet (link: https://hms.az1.qualtrics.com/SE/?SID=SV_3CyZOKOuiEvnCx7) to the Coordinating Center Project Manager.
2.2. The Coordinating Center Project Manager updates the research concept sheet log in Box.com.
2.3. The Coordinating Center Project Manager sends the research concept sheet to the Publication and Research Committee Co-Chairs.
  2.3.1. The co-chairs identify any duplication of effort or other concerns before circulation to the full committee.
2.4. The Publication and Research Committee Co-Chairs send the research concept sheet to the Publication and Research Committee Members for comments.
2.5. The Publication and Research Committee Members vote on the research concept sheet by email. For example, one basis of rejection would include if there is already another study/s underway with overlapping aims.
  2.5.1. If a Publication and Research Committee Member wants discussion, the Publications and Research Committee co-chairs contact the Coordinating Center Administrator to schedule a meeting.
2.6. The Publication and Research Committee Co-Chairs notify the Investigator and Coordinating Center Project Manager with the decision.
2.7. The Coordinating Center Project Manager updates the research concept sheet log with the decision.
  2.7.1. If the research concept sheet is accepted, the Coordinating Center Project Manager uploads the research concept sheet to Box.com.

Version: 1

Effective Date: May 1, 2015

Last Reviewed Date: June 09, 2015

TITLE: Research Projects Led by External Investigators

1. RESPONSIBLE PERSONNEL:

1.1. Investigators: Investigators outside of the UDN.
1.2. Coordinating Center Project Manager: Individual at the Coordinating Center who communicates with the Clinical Sites, Cores, Coordinating Center, and NIH team members to complete project-related activities.
1.3. Coordinating Center Executive Director: Individual at the Coordinating Center who directs project-related activities of the UDN.
1.4. Steering Committee: Committee made up of UDN Investigators that decides on the priorities and order of business of the UDN.

2. PROCEDURE:

2.1. Investigators submit a concept form (link: https://hms.az1.qualtrics.com/SE/?SID=SV_3CyZOKOuiEvnCx7) for an external project that will rely on UDN data to the Coordinating Center Project Manager.
2.2. The Coordinating Center Project Manager updates the external project log in Box.com.
2.3. The Coordinating Center Project Manager submits the external project request to the Coordinating Center Executive Director.
2.4. The Executive Director notifies the Coordinating Center Project Manager of the external project Steering Committee presentation date.
  2.4.1. The Executive Director or the Steering Committee may form an ad hoc review committee to vet the initial proposal.
2.5. The Coordinating Center Project Manager notifies the Investigator of the presentation date.
2.6. The Investigator presents at the Steering Committee meeting.
2.7. The ad hoc review committee gives its recommendations to the Steering Committee.
2.8. The Steering Committee votes on the external project.
2.9. The Coordinating Center Project Manager updates the external project log with the decision.

Version: 1

Effective Date: May 1, 2015

Last Reviewed Date: June 9, 2015

TITLE: Manuscripts

1. RESPONSIBLE PERSONNEL:

1.1. Investigators: Investigators at the UDN Clinical Sites, Cores, Coordinating Center, and NIH.
1.2. Coordinating Center Project Manager: Individual at the Coordinating Center who communicates with the Clinical Sites, Cores, Coordinating Center, and NIH team members to complete project-related activities.
1.3. Publication and Research Committee Co-Chairs: Individuals who lead the Publications and Research Committee.
1.4. Publication and Research Committee Members: Individuals who participate in the Publications and Research Committee.
1.5. Coordinating Center Administrator: Individual at the Coordinating Center who handles the administrative tasks for the Coordinating Center.

2. PROCEDURE:

2.1. Investigators submit a manuscript to the Coordinating Center Project Manager.
2.2. The Coordinating Center Project Manager updates the manuscript log in Box.com.
2.3. The Coordinating Center Project Manager sends the manuscript to the Publication and Research Committee Co-Chairs.
2.4. The Publication and Research Committee Co-Chairs send the manuscript to the Publication and Research Committee Members for comments (initial review of appropriate authorship, acknowledgements and broad science).
2.5. The Publication and Research Committee Members vote on the manuscript by email.
    2.5.1. If a Publication and Research Committee Member wants discussion, the group contacts the Coordinating Center Administrator to schedule a meeting.
2.6. The Publication and Research Committee Co-Chairs notify the Investigator and Coordinating Center Project Manager with the decision (ideally within 2 weeks).
2.7. The Coordinating Center Project Manager updates the manuscript log with the decision.
    2.7.1. If the manuscript is accepted for publication, the Coordinating Center Project Manager uploads the manuscript to Box.com.

Version: 1

Effective Date: May 1, 2015

Last Reviewed Date: June 9, 2015

APPENDIX 17: Proposed UDN Metrics

Note: NIH indicates NIH Program metrics, which will be calculated over the course of the network. UDN indicates UDN-nominated metrics, which may be calculated over the course of the network.

	Source	Performance Metrics and Milestones
1	NIH	By Oct 1, 2014 IRP-UDP will identify at least 5 candidate genes
2	NIH	IRP-UDP will analyze 400 SNPs and 400 WES or WGSs per year through FY2017
3	NIH	By Oct 1, 2015 Extramural Clinical Sites (ECSs) to see 25 patients per year per site to initiate phenotyping
4	NIH	By Oct 1, 2015 ECSs to identify candidate genes by analyzing 200 SNPs and 200 exomes/genomes per year, increasing to 800 SNPS and exomes/genomes per year in years 3 and 4
5	NIH	Define the mechanism of at least 1 candidate gene in the pathophysiology of a rare or yet-to-be described disease
6	NIH	By Oct 1, 2016 all ECSs to see 50 patients per year per site
7	NIH	By Jan 2016–Identify 10 unidentified diseases; by Jan 2018, identify 20 unidentified diseases
8	UDN	Number of inquiries from potential patients and doctors through the UDN portal (coordinating center)
9	UDN	Number and percentage of inquiries that result in admission reviews performed (each site)
10	UDN	Number and percentage of admission reviews that result in acceptance (each site)
11	UDN	Number and percentage of people accepted for admission who are actually admitted (each site).
12	UDN	Number and percentage of people admitted who get a provisional diagnosis (each site).
13	UDN	Number and percentage of people admitted who get a validated diagnosis (each site).

	Source	Performance Metrics and Milestones
14	UDN	Number and percentage of people admitted who have a novel diagnosis (overall)
15	UDN	Number and percentage of people admitted who die or become ineligible before admission (each site).
16	UDN	Number and types of adverse events and deaths while traveling to/from site or during hospital stay (each site).
17	UDN	Number and percentage of people admitted who experience an adverse event while traveling to site or during hospital stay (each site).
18	UDN	Time from inquiry to chart completion (each site).
19	UDN	Time from chart completion to admission review/decision. (each site)
20	UDN	Time from decision to admit to admission (each site).
21	UDN	Time from completion of admission stay to data upload to UDN data repository (each site)
22	UDN	Time from completion of admission stay to first provisional or validated diagnosis communicated to patient (each site).
23	UDN	Number and percentage of patients with complete data in the UDN data repository (each site)
24	UDN	Time from when sample is gathered to when extracted DNA is received by the sequencing center (each site)
25	UDN	Time from receipt of extracted DNA to providing raw sequencing data (sequencing center)
26	UDN	Time from completion of sequencing to network sequencing analysis completion (each site)
27	UDN	Number and percentage of isolated individual sequences that result in identification of a candidate gene (network-wide)
28	UDN	Number and percentage of trio (or larger) sequences that result in identification of a candidate gene (network-wide)
29	UDN	Number and percentage of sequences that result in the identification of a candidate gene (network-wide)
30	UDN	Number of people who receive a non-genetic diagnosis.

	Source	Performance Metrics and Milestones
31	UDN	Time from sequence generation and completion of phenotyping to upload to dbGaP (clinical site + coordinating center?)
32	UDN	Number and percentage of candidate variants that go on to be matched with a lab for functional testing (network-wide)
33	UDN	Quality of Human Phenotype Ontology phenotyping by site (each site)
34	UDN	Patient and family satisfaction (each site and network-level)
35	UDN	Ability to integrate or link data with Phenome Central (coordinating center)
36	UDN	Geographic distribution (zip code) of patient
37	UDN	Geographic distribution (zip code) of referring doctor
38	UDN	Number and percentage of admissions referred by site (e.g., Site A refers patient who is seen at Site A) (each site)
39	UDN	Number and percentage of admission reviews, admissions, and diagnoses by race, ethnicity, and age-range. (network-wide)
40	UDN	Predominant phenotype class/subspecialty admitted (each site)
41	UDN	Proportion of patients for whom insurance was billed (each site)
42	UDN	Average, minimum, and maximum direct cost of evaluation tests per patient.
43	UDN	Publications and presentations (each site and coordinating center)
44	UDN	Time to complete phenotype from date of admission (each site)
45	UDN	Number and proportion of patients with an HPO phenotype (by site)

APPENDIX 18: Billing Surveys

Billing Survey- Institutions using Clinical Billing (to be completed for an individual patient)

1. List the primary, secondary, and tertiary insurers that were billed for the patient (**if patient is self-pay and if all expenses were thus paid for out of the grant, please fill out the individual patient grant billing survey rather than this one**).
2. List all clinical consultations/procedures/tests/imaging ordered for this patient

   Please list those that were:

   a) Rejected:
   b) Reimbursed:
   c) Appealed (outcome of the appeal):
   d) Paid for out of grant funds:
   e) Paid for out of Supplemental Funds (please specify the fund e.g CTSA):
   f) Written-off by medical center:

   If available, please list the total co-pay/deductible payment that this patient incurred: $

   List total reimbursed costs for this patient $_____

   List total unreimbursed costs for this patient $_____

3. Please list services (if any) that were directly charged to the grant (never submitted for reimbursement) and why.
4. Was pre-authorization or a billing estimate performed for the patient? Yes/No

   Was it for the entire UDN evaluation? Yes/No

   If it was for a specific test/s, please list:

   Did study personnel perform this task, or did you have assistance from the medical center?

5. Was the billing processed "specially" for this UDN patient at your medical center (for example manually handled rather than automated)? Yes/No

   Please provide details regarding the special handling.

   Who was responsible for the special handling (study personnel, medical center staff, other)?

6. Were there any unanticipated costs for the patient? Please list the reasons and the costs (These would not include co-pays/deductibles, but would be expenses such as needing acute care for adverse events related to the UDN procedures and/or unrelated acute illnesses that needed hospitalization etc).

How were such costs paid for? Insurance billed_____ Grant funds _____
Self-pay____ Other (specify)_____

7. Did your patient incur any other out-of pocket expenses (do not include co-pays/deductibles)? If so state the reasons and the amount.
8. What was the total cost of travel for this patient and family member and what did this include?

   How were the travel costs reimbursed? Grant_____ NORD central fund _____

9. Please describe your or your patient's experience with being reimbursed for travel by the NORD fund.
10. Any lessons learned from this patient's billing experience? Please elaborate.

Billing Survey- Institutions using Grant Billing (to be completed for an individual patient)

1. List all clinical consultations/procedures/tests/imaging ordered for this patient

   Please list those that were:

   a) Paid for out of grant funds:
   b) Paid for out of Supplemental funds. Please specify type/source of funds:
   c) Written-off by medical center:

   Total costs paid for out of the grant $_____

2. List the primary, secondary and tertiary insurers (if available) for the patient that could have been billed for the UDN evaluations at your site. Please specify if the patient is self-pay.
3. Was a billing estimate performed for the patient prior to or at the UDN visit? Yes/No
   If so, did study personnel perform this task, or did you have assistance from the medical center?
4. Was the billing processed "specially" for this UDN patient at your medical center (for example manually handled rather than automated)? Yes/No

   Please provide details regarding the special handling.

5. Were there any unanticipated costs for the patient? Please list the reasons and the costs (These would not include co-pays/deductibles, but would be expenses such as needing acute care for adverse events related to the UDN procedures and/or unrelated acute illnesses that needed hospitalization etc).

   How were such costs paid for? Insurance billed_____ Grant funds _____
   Other (specify)_____

6. Did your patient incur any other out-of pocket expenses? If so state the reasons and the amount.
7. What was the total cost of travel for the patient and family member and what did this include?

   How were the travel costs reimbursed? Grant_____ NORD central fund _____

8. Any lessons learned from this patient's billing experience?

Billing Survey- Institutions using Clinical Billing (summary of first 5 patients)

1. What is the average reimbursed expense for the first five patients (sum of all reimbursed expenses/total sum of all expenses submitted for reimbursement across the 5 patients)?
   $_____
   (Please include all costs that were submitted to insurers: clinical consultations, tests, procedures etc. Do not include co-pays/deductibles and travel costs or expenses that were paid for out of grant funds)
2. What is the average unreimbursed expense for the first five patients (sum of unreimbursed expenses/total sum of all expenses submitted for reimbursement across the 5 patients)?
   $_____
3. Please detail the insurance carriers that you have billed so far (n= number of patients)
   a. Private Carriers (n=)
   b. Medicaid (n=)
   c. Medicare (n=)
   d. Other (institutional funds etc.) (n=)
4. How many of your patients have been uninsured thus far and thus expenses were paid for out of the grant?
5. Any lessons learned from your clinical billing experience thus far?

APPENDIX 19: Collaborative Clinical Site Application

Collaborative Clinical Site Application

Date of Application:

_____

Organization:

_____

Main Contact Name:

_____

Main Contact e-mail:

_____

The UDN is open to Collaborative Clinical Sites that agree to the criteria for participation described below.

Criteria for Participation in the UDN are:

- Each participant will inform the UDN NIH PO and the UDN Steering Committee about his/her group's plans for an affiliate UDN site.
- Each participant will specify the sequencing, laboratory, and clinical evaluation plans for his/her proposed affiliate site.
- Each participant is expected to contribute significantly to the project, bringing his/her particular expertise to bear on accomplishing the goals of the UDN in a timely manner. Participation in the UDN should include substantial intellectual contributions to the Network.
- Each participant will adhere to UDN data sharing and publications policies, guidelines and agreements.
- Each participant will take part in group activities, including attending UDN Steering Committee meetings and working group calls and contributing to the products of these groups.
- Each participant will agree that s/he will not disclose confidential information obtained from other members of the UDN.
- Additional criteria may be added upon recommendations of the UDN Steering Committee, External Scientific Panel, and the NIH UDN Working Group.

Applications will be reviewed by the UDN Steering Committee, UDN program staff, and the UDN External Scientific Panel to determine whether a Collaborative Clinical Site will be accepted. **A limited number of Collaborative Clinical Sites may be approved and acceptance may be limited to one-year after which an assessment will be conducted for continuation.**

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1. Please provide a concise description of your DNA sequencing, other laboratory, and clinical evaluation plans and a rationale for how your proposed Collaborative Clinical Site addresses the goals of the UDN. *(maximum length 3 pages, font 11, single spacing)*

2. Please provide evidence that the proposed Collaborative Clinical Site's research has received appropriate IRB approvals and is consistent with participants' informed consent.

3. Please provide evidence of funding to conduct the proposed research.

4. _____ (organization name) agrees to abide by the UDN Data Sharing and Use Agreement and data submission policies by providing a signed UDN Data Sharing and Use Agreement to the UDN Coordinating Center.

5. _____ (organization name) agrees to participate fully in UDN activities, including attending UDN Steering Committee meetings and working group calls and contributing to the products of these groups. Initials of main organization contact: _____

6. _____ (organization name) agrees to abide by the UDN publications policies.

7. _____ (organization name) agrees to not disclose confidential information obtained from other members of the UDN.