CureGN: Cure Glomerulonephropathy Network

Core Study Protocol

Version 1.2

Version 1.1: July 28, 2014
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Sponsor:
NIH-NIDDK
NephCure Kidney International

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Protocol Approval

Protocol: CureGN Protocol 1.2: Cure Glomerulonephropathy Network	Version/Date: 1.1/July 28, 2014 1.2/ September 8, 2014
IND: N/A	**CureGN DCC Principal Investigator :** Matthias Kretzler, MD
Study Sponsors: The National Institute of Diabetes and Digestive and Kidney Diseases (NIDDK), NephCure Kidney International	
INSTRUCTIONS: The Principal Investigator must print, sign, and date below. The original signature page should be kept in the site's records. After signature, please scan the signature page and email or fax to the CureGN DCC at the address listed below: CureGN DCC CureGN-Admin@ArborResearch.org Fax: 734-665-2103	
I confirm that I have read the above protocol in the latest version. I understand it, and I will work according to the principles of Good Clinical Practice (GCP) as described in the United States Code of Federal Regulations (CFR) – 21 CFR Parts 45, 50, 56, and 312, and the International Conference on Harmonization (ICH) document "Guidance for Industry: E6 Good Clinical Practice: Consolidated Guidance" dated April 1996. Further, I will conduct the study in keeping with local, legal, and regulatory requirements. As the Principal Investigator, I agree to conduct and to carry out the study by the criteria written in the protocol and understand that no changes can be made to this protocol without written permission of the CureGN Steering Committee.	
Site Principal Investigator (Print)	
Site Principal Investigator (Signature)	
Date	

1. INTRODUCTION

Glomerular disease, including minimal change disease (MCD), focal segmental glomerulosclerosis (FSGS), membranous nephropathy (MN), and immunoglobulin A nephropathy (IgAN), often share a common clinical presentation. These chronic diseases, affecting both children and adults, produce proteinuria, hypoalbuminemia, hematuria, and/or edema, as the glomerulus is damaged by the underlying disease process. Progressive loss of kidney function often occurs over many months or years and results in substantial individual and societal burden.

There exist several major challenges to understanding the underlying biology of these conditions and to translating that understanding into effective therapies for patients. These include the fact that glomerular diseases are a relatively rare cause of chronic kidney disease (CKD) as compared with more common etiologies such as diabetes, hypertension, or congenital anomalies of the kidney and urinary tract. The slow progression in many patients may require follow-up periods of decades to measure effectiveness of an intervention, as alternative endpoints to death and end stage kidney disease (ESKD) have not been definitively validated in this population. As a result, it is difficult to recruit sufficient numbers of patients to study underlying mechanisms, identify disease targets and biomarkers, and evaluate new therapies.

Cure Glomerulonephropathy (CureGN) is a multi-center consortium that will work collaboratively to address these challenges by recruiting a large, ethnically diverse cohort of glomerular disease patients and follow them prospectively with a common protocol. This study will establish an infrastructure that enables the following questions to be addressed for glomerular disease patients:

- What is this disease?
- Why do I have this disease?
- What will happen to me?
- What effective treatments can you offer me?

2. BACKGROUND/SIGNIFICANCE

MCD, FSGS, MN, and IgAN are glomerular diseases which often result in devastating complications of nephrotic syndrome and progressive renal insufficiency. Although relatively rare compared with the most common causes of CKD, they present a significant individual and societal burden. The morbidity and mortality from these diseases are related both to complications of the disease itself (e.g., ESKD, venous thromboembolism[1,2], bacterial peritonitis[3], hypertension, symptom burden, and reduced quality of life[4,5]) as well as the immunomodulatory therapies (e.g., steroid toxicity[6], calcineurin nephrotoxicity[7], impaired fertility[8] and bladder toxicity of cyclophosphamide[9], and infectious complications). In 2010, glomerular diseases accounted for 13% of ESKD prevalence (84,521/640,023 patients) in the United States[10]. Among patients <20 years old, FSGS is the leading cause of acquired ESKD. Furthermore, IgAN is the leading cause of primary glomerular disease and an important contributor to kidney failure worldwide[11].

Although these glomerular diseases are currently categorized as four distinct histopathologic categories, they result from multiple biological mechanisms. At the same time, their clinical phenotypes cross these four histopathologic categories and are treated with common therapeutic strategies. In the current

treatment paradigm, diagnostic, prognostic, and therapeutic decisions are largely based on histological and crude clinical parameters that do not account for the heterogeneity of the biological antecedents and disease trajectories. As a result, available therapies are few, and individual response uncertain. Progress has been limited by the rarity of these diseases and long duration of observations required to evaluate clinically relevant outcomes such as ESKD. As a result, many current treatment recommendations are based on retrospective data, small numbers, and heterogeneous study populations[12]. Thus, we are challenged to provide specific, individualized treatments for people with glomerular disease.

Novel insights into pathophysiology of these disorders have been described recently. Anti-phospholipase A2 receptor (PLA2R) antibodies have been identified in approximately 70% of idiopathic MN cases in adults and may serve as an important marker for diagnosis and disease activity, as well as potentially a therapeutic target[13]. Bovine serum albumin targeted antibodies have been identified in childhood onset MN[14]. Antibody-antigen complex stimulated by aberrant IgA1 O-glycan has been identified as a pathogenic mechanism in IgAN. Genetic studies of primary glomerular diseases have identified specific genetic risk loci associated with disease, disease-specific phenotypes, and risk of both progression to ESKD and post-transplant recurrence[15,16,17,18,19]. In parallel to the disease-specific advances, we are engaged in a fundamental transition from research models focused on the functions of single molecules or pathways to an integrative biology analyzing biological systems as a unified whole. This systems biology approach integrates genome-scale data sets to define key drivers of diseases and allows the formation of novel hypotheses of organ function and failure[20].

A key underlying hypothesis of CureGN is that different glomerular disease mechanisms can result in similar histological and clinical phenotypes, but very different disease courses. A similar hypothesis has been extensively evaluated in oncology. Comprehensive molecular analysis of tumor tissue has allowed the definition of cancer-specific molecular fingerprints representing different disease mechanisms or states of classically indistinguishable neoplastic lesions[21,22], with some currently under prospective evaluation as prognostic and predictive biomarkers[23]. The application of a similar strategy to glomerular disease will allow a mechanistic disease definition and, we believe, will have far-reaching consequences for diagnostic classification, prediction of disease and risk of progression, definition of patient cohorts for clinical trials, and identification of personally tailored therapeutic regimes[20].

To accomplish these goals, the CureGN consortium will recruit and maintain a large cohort of patients with glomerular disease and follow them prospectively with standardized clinical data and biospecimen collection. The infrastructure and study design presented in this protocol will form the backbone for a broad range of scientific approaches and inquiries, essential to moving the field forward and improving the outcomes of patients affected by these diseases.

3. AIMS

CureGN Consortium Aims

The CureGN Consortium Aims are principally procedural. Aims 1-4 will be established via this protocol, while Aims 5 & 6 are outside the scope of this protocol.

1. Recruit a multiethnic cohort of at least 2,400 adult and pediatric patients with biopsy-documented IgAN, FSGS, MN, and MCD
2. Establish a longitudinal database of patients with these glomerular diseases
3. Perform standardized collection of biospecimens at scheduled visits
4. Facilitate the development of translational and clinical ancillary studies that will advance the diagnosis and care of patients with glomerular diseases
5. Recruit ethnicity-matched controls for these diseases (outside scope of protocol)
6. Engage and educate investigators in clinical and translational research in glomerular diseases (outside scope of protocol)

Scientific Aims

The following Scientific Aims describe four broad categories of research that CureGN will address. The Aims are not exhaustive, but establish over-arching goals that guide the study design, eligibility criteria, visit schedule, sample collection efforts, and eventual integration with ancillary studies. Each Aim will be addressed for each of the target CureGN diseases: IgAN, FSGS, MN, and MCD.

1. **Aim 1 (Epidemiology)**. To describe the disease trajectory under current clinical care; to estimate event rates for clinically meaningful outcomes; to identify patient characteristics (demographic, clinical, laboratory, environmental) associated with glomerular disease and non-renal complications of disease; to identify clinical predictors of short- and long-term outcomes, including therapeutic response; and to evaluate intermediate outcomes, such as proteinuria, as potential surrogates for longer-term outcomes.
2. **Aim 2 (Biomarkers)**. To identify and characterize clinical, histological, molecular, and genetic biomarkers that are linked to glomerular disease, disease outcomes, or that might be used to improve disease classification; to identify and characterize biomarkers that may be employed in clinical practice or clinical trials to predict disease trajectory, disease activity, or response to therapy.
3. **Aim 3 (Genetics)**. To understand the genetic architecture of the four glomerulopathies, including studies of germline sequence variation, somatic mutations, epigenetic changes, and transcriptomic profile, and their impact on disease presentation and clinical outcome; study gene-gene and gene-environment interactions that contribute to the development of the four glomerulopathies; and devise systems genetics approach to clarify pathogenesis.
4. **Aim 4 (PROs)**. To identify Patient Reported Outcomes (PROs, e.g., symptom burden, physical function, quality of life) associated with primary glomerular diseases; to validate disease-specific instrument(s) to assess the impact of disease and its therapy on patients; and to test the associations of PROs with disease progression.

4. INVESTIGATION PLAN

4.1. STUDY METHODS

4.1.1 Overview

The core CureGN study is a multi-center prospective cohort study of approximately 2,400 adult and pediatric patients with biopsy-documented IgAN, FSGS, MN, and MCD. For each disease category, approximately 600 participants will be recruited. Participants will be recruited concurrently from each of the four Participating Clinical Center (PCC) networks: Columbia University, Midwest Pediatric Nephrology Consortium, University of North Carolina, and University of Pennsylvania. Each PCC represents multiple clinical sites, with current representation in the United States, Canada, and Italy. Participants meeting the enrollment criteria below will be enrolled if they or their legally authorized representative(s) provide signed informed consent. Patients will be followed until death, withdrawal from the study, or end of study. For the initial funding period, total study duration is approximately 4 years. Participant recruitment may occur at any point during the study period, and at least until recruitment goals are met for each disease category.

4.2. PARTICIPANT SELECTION

4.2.1 Inclusion Criteria

- Diagnosis of MCD, FSGS, MN, or IgAN on first diagnostic kidney biopsy, as per specified pathology definitions
- First diagnostic kidney biopsy within 5 years of study enrollment
- Access to first kidney biopsy report and/or slides
- All ages
- Willingness to comply with study requirements, including intention to fully participate in protocol-specified follow-up at a clinical study site
- Informed consent and, where age appropriate, informed assent

4.2.2 Exclusion Criteria

- ESKD, defined as chronic dialysis or kidney transplant
- Institutionalized patient
- Solid organ or bone marrow transplant recipient at time of first kidney biopsy
- Diagnosis of any of the following at the time of first diagnostic kidney biopsy:
    - Diabetes mellitus
    - Systemic lupus erythematosus
    - HIV infection
    - Active malignancy, except for non-melanoma skin cancer
    - Active Hepatitis B or C infection, defined as positive viral load

**For participants in the NEPTUNE cohort study with one of the core diagnoses in CureGN (MCD, FSGS, MN, IgA nephropathy), CureGN exclusion criteria may be waived to allow for long term follow-up via CureGN study.*

4.2.3 Special Considerations

Racial/Ethnic Distribution

A recruitment goal is to have a multi-racial and ethnic cohort with representation from Caucasian, African-American, American Indian/Alaska Native, Asian/Pacific Island and Latino/Hispanic groups. Recruitment from these groups will be monitored, with targeted recruitment efforts implemented if goals are not met.

Pediatric Patients

Children (≤18 years old at the time of first diagnostic kidney biopsy) are eligible for the study.

4.3. SCHEDULE OF VISITS, TEST, AND ASSESSMENTS

4.3.1 Visit Schedule

Table A provides a schematic of the visit schedule. The enrollment visit is denoted as V0, and the follow-up visit schedule is based on 4-month visit windows starting the day after V0. As participants may be enrolled at any time during the 4-year study period, duration of follow-up will be up to 4 years.

For each 4-month follow-up visit window, the first clinic visit in any study window should be used as the study visit. All visit windows are contiguous. This schedule is intended to be flexible to align with clinical care to every extent possible, and to capture biospecimens and clinical data at times of disease activity and remission. It anticipates more frequent visits for participants with active disease and less frequent visits for participants with stable disease. For example, incident patients enrolled shortly after biopsy are more likely, but not required, to have more frequent visits in the first year of study follow-up.

Patients will have a visit every 4 months throughout the 4-year study. V0 and one annual visit (V1, yearly visit) are required in-person (shaded visits in Table A). The other visits (during the V2 and V3 windows, event visits) should occur as an in-person study visit if the participant is coming for routine clinical care; however if a regular clinical visit is not scheduled, a remote (phone or e-mail) visit should occur. See section 4.3.4.

Table A: Visit Schedule

Study Year	**1**				**2**			**3**			**4**		
Study Visit	Enroll V0	Y1 V1	Y1 V2	Y1 V3	Y2 V1	Y2 V2	Y2 V3	Y3 V1	Y3 V2	Y3 V3	Y4 V1	Y4 V2	Y4 V3
Study Month	0	1-4	5-8	9-12	13-16	17-20	21-24	25-28	29-32	33-36	37-40	41-44	45-48

4.3.2 Screening Procedure

Screening Log

Throughout the enrollment period, limited data will be collected on patients who are seen in clinic and had a first diagnostic kidney biopsy with one of the four diagnoses (IgAN, FSGS, MN, or MCD) within the last 5 years. The biopsy may have been performed at the enrollment site or elsewhere.

Data Elements

For the Screening Log, data elements will include:

- Non-identifying demographics (e.g., age, sex, race, and ethnicity)
- Inclusion/exclusion criteria

- For eligible patients, documentation of informed consent or reason for study non-participation

Eligibility Encounter

For eligible patients, this will serve as an introduction to the CureGN study. This encounter may occur by phone or in person. It may occur prior to, or at the same time as, V0. The study coordinator will confirm eligibility, review study requirements, and determine the participant's willingness to participate in the study. Informed consent may be obtained at this time or during V0.

4.3.3 Enrollment Visit

Consent

If previously obtained, a physician investigator and/or a study coordinator will review and explain necessary information with the potential participant in accordance with the requirements of the respective Institutional Review Board (IRB) and federal human subject research regulations. The IRB-approved written comprehensive consent and (if applicable) assent documents will be reviewed. Upon granting of consent, V0 will be conducted.

Data Collection

At V0, clinical data will be gathered by patient interview, patient questionnaire, and chart extraction as outlined in Table B (Data Elements). This includes medical history, prior disease course, medication exposures, and local laboratory results. Additional information will include contact information, next of kin, information regarding participant's health providers, and documentation for permission to obtain medical records with appropriate signatures. Personal identifiers will be accessible to the local study team only.

A brief, focused physical exam will be performed, and PRO measures will be assessed. Biospecimen collection will occur, as per Table B (Data Elements) and Table C (Biospecimen Volumes). A spot urine collection will be obtained. Attempts should also be made to collect a 24-hour urine sample, especially if the eligibility encounter was performed prior to V0.

Pathology Review for Enrollment

Patients may proceed with V0 prior to review of the kidney biopsy by the CureGN study pathologists, if the physician investigator and/or the study coordinator believe that the subject meets eligibility criteria. Participants may be withdrawn from the study if biopsy review by the CureGN study pathologists reveals that the patient does not meet pathology inclusion criteria. Confirmation of diagnosis and assignment into diagnosis category (MCD, FSGS, MN, and IgAN) is accomplished, at each PCC, by a PCC pathologist by: (1) review of report if diagnosis is clear and glass slides are not readily available to core PCC pathologist; (2) review of report and glass slides, electron microscopy (EM), and immunofluorescence (IF) images, if pathology materials are readily available and at PCC pathologist's discretion; or (3) review of report and glass slides (+ EM and IF images if available) if pathology report is not clear.

Digital Pathology Repository

De-identified biopsy slides will be scanned into the CureGN Digital Pathology Repository for pathology scoring and future research for all consented patients for whom slides are available.

Pathology slide scanning may occur at the local pathology site or at the CureGN Image Coordinating Center. Patients for whom slides are not available may be enrolled in CureGN, with a target to enroll a minimum of 80% of patients with accessible slides.

4.3.4 Follow-Up Visits

In-Person Visits

To every extent possible, study visits will take place at the time of clinical visits to the study site. Thus, more frequent study visits/biospecimen collection will occur in patients who are being seen more frequently in clinic (e.g., with recent diagnosis or active disease) and less frequently in patients seen less frequently (e.g., with more stable disease). At a minimum, each participant will have two in-person visits during the first year of study and one visit in subsequent years (Table A).

Data collection, a brief focused physical exam, PRO measures, and biospecimen collection will occur, as in Table B (Data Elements) and Table C (Biospecimen Volumes). In addition to a spot urine collection, attempts should be made to collect a 24-hour urine sample or first morning void, as per the footnote to Table B.

Interim Remote Visits

Contact by telephone or secure electronic correspondence will occur at a minimum of once per 4-month interval (typically near the end of each 4-month visit window) if the participant has not been seen for a study visit during that window. These interim remote visits will maintain connection with the patient, ascertain major clinical events (e.g., ESKD, hospitalizations, and major medication changes), and assure that a study visit is scheduled at least annually. Chart abstraction of clinical data such as laboratory results, clinic visits and/or medication changes will typically occur during each 4-month interval (whether associated with an in-person visit or interim remote visit).

Data Collection

As in V0, in-person follow-visit data will be gathered by a combination of patient interview, patient questionnaire, and chart extraction. Reason for clinical visit (e.g., routine follow-up, medication adjustment, disease flare) will be recorded. A brief, focused physical exam will be performed, and PRO will be assessed. Data and biospecimen collection will occur, as in Table B (Data Elements) and Table C (Biospecimen Volumes). Attempts should be made to collect a 24-hour urine sample on an annual basis. For all visits, a morning void urine and spot collection during the visit should be obtained. If a morning void sample cannot be obtained, a spot collection is acceptable. The spot collection should include the collection time.

4.4. Data Elements

Table B provides an overview of categories of data elements by visit type.

Table B: Overview of Data Elements

Visit	Eligibility	Enrollment	In-Person Follow-Up	Remote Follow-Up
Screening Log Data				

Demographics	X			
Biopsy diagnosis	X			
Exclusion criteria	X			
Consent/assent	X	(X)[a]		
Medical Data				
Comorbidities		X	X	X
Family history		X	X	
Birth history		X	X[b]	
Pregnancy history		X	X	
Prior disease course		X		
Interim disease course			X	X
Medications		X	X	X
Hospitalizations		X	X	X
ESKD status			X	X
Vital status			X	X
Physical exam		X	X	
Vital signs		X	X	
PRO Data				
Symptoms		X	X	X
PRO questionnaires		X	X	
Local Laboratory Tests (if measured, based on abstraction from clinic record)				
Blood chemistries		X	X	X
Coagulation studies		X	X	X
Hematology studies		X	X	X
Rheumatologic serology		X	X	X
Infectious serology		X	X	X
Urine studies		X	X	X
Central Laboratory Tests (measured by CureGN laboratory)				
Serum creatinine		X	X	
24-hour , morning void, or spot urine (protein, creatinine)[c]		X	X	
Biospecimens				
Blood sample including DNA*		X	X	
Immortalized cell lines[d]		X		
24-hour, morning void, or spot urine [c]		X	X	

(a) If not previously performed
(b) If not previously collected
(c) Attempts should be made to collect a 24-hour urine sample on an annual basis. For all other visits, a morning void collection in a designated, pre-labeled container should be obtained. Additionally, a spot collection should be done during the visit, noting the time of collection. If a morning void collection cannot be obtained, a spot urine collection is acceptable.
(d) Pediatric patients only (for core CureGN protocol)
** Blood sample should note time of day at collection*

Biospecimens

Total blood and urine volumes at baseline and follow-up visits are listed in Table C below. Pediatric patients' blood volumes will be adjusted based on weight at the time of visit. Specimen volumes required for clinical testing will be subtracted from these volumes.

Table C: Biospecimen Volumes

	Total Blood Volume			Total Urine Volume		
Visit type	V0, Baseline (mL)	V1, Yearly (mL)	V2-3, Event(mL)	V0, Baseline (mL)	V1, Yearly (mL)	V2-3, Event (mL)
Pediatric participants						
<21 pounds	20	20	20	75	20	75
21-<52 pounds	45	20	45	75	20	80
>52 pounds	50	20	50	80	20	80
Adult participants	50	20	50	80	20	80

Follow-Up Data on ESKD and Vital Status

For enrolled patients who progress to ESKD during the follow-up period, additional data collection will include date of ESKD, renal replacement therapy (RRT) modality, and kidney transplant donor type. For patients who die during the follow-up period, data collection will include date of death, cause of death, and ESKD status at death. Patient consent will include permission to link data to external data sources, such as Centers for Medicaid and Medicare Services (CMS) End-Stage Renal Disease (ESRD) data and National Death Index, for ascertainment of ESKD and vital status.

4.5. Sample size and power calculations

The statistical power calculations are based on the following assumptions: (1) data will be obtained from more than 40 sites coordinated by the four PCCs; (2) 800 patients will be recruited per year over 3 years, resulting in an average follow-up of 2 years during the first 4-year data collection phase; and (3) assume a loss of 10% of the available follow-up due to patient loss of follow-up. Additional assumptions include: a power of 80%, an alpha of 0.05, an intra-cluster correlation of 0.05, and a between-facility normalized standard deviation of the sample size of 0.15. Power is computed for a range of sample sizes, representing subgroup comparisons such as within and between diagnosis groups (FSGS, MCD, MN, IgAN), among pediatric patients, and comparisons with control populations. The statistical power for time-to-event analyses are calculated as the minimum detectable hazard ratio (MDHR) associated with comparisons between equal numbers of patients, i.e., 50% exposure, out of the number shown in the column headings of Table D. Study durations of 4 and 20 years correspond to the first phase of the study and to potential future phases. Event rates are derived from results in published literature, including examples of ESKD/death rates for FSGS of 0.03[24] and 0.05[25] per patient-year, IgAN around 0.03[25,26], and MN of 0.01[27] and 0.02[25]; proteinuria reduction (remission) rates for FSGS of 0.18[24] and for MN of 0.23[27]; and rates of a 50% drop in estimated glomerular filtration rate (eGFR, progression) for MN of 0.04[28]. These rates are not outcome-specific; any analysis of an event with a similar rate on the data described would have the MDHR indicated in Table D.

Table D: Minimum Detectable Hazard Ratios by Observation Period, Expected Event Rate, and Cohort Size						
Outcome	Study Duration (avg. patient years)	Rates	300 (150/Group)	600 (300/Group)	1200 (600/Group)	2400 (1200/Group)
ESKD/death	4 year (2 yrs)	.01-.04	>10	>10-7.4	>10-4.5	>10-3.6
	20 year (17 yrs)	.01-.04	>10-2.6	5.9-2.0	4.0-1.8	3.3-1.6
50% eGFR	4 year	.04-.12	>10-3.2	7.3-3.4	4.5-2.0	3.6-1.9
(progression)	20 year	.04-.12	2.5-1.8	2.0-1.6	1.8-1.5	1.7-1.4
Proteinuria	4 year	.18-.23	2.4-2.2	2.0-1.8	1.8-1.7	1.7-1.6
(remission)	20 year	.18-.23	1.7-1.6	1.5-1.5	1.4-1.4	1.4-1.3

4.6. Statistical Analysis

Descriptive statistics will be used to characterize the overall cohort and subgroups of interest. Summary statistics, including mean (standard deviation), median (interquartile range [IQR]), and frequencies will be calculated. Graphical methods will be used to examine distributions, identify potential influential points and guide in data transformations as needed. Relationships between variables will be similarly assessed for linearity, symmetry, and homoscedasticity. To compare subgroups within the larger cohort or to other cohorts, both univariate testing (e.g., using t-tests, analysis of variance [ANOVA], Kruskal-Wallis, Mann-Whitney as appropriate) and model-based adjustment for other factors will be performed. Both logistic regression and a mixed model approach with center as a random effect (to account for between-center differences) will be used as they are suited for cross-sectional and retrospective case-control analyses of multiple exposures and/or biomarkers. Comparisons between the Screening Log and enrolled patients will allow assessment of the extent of recruitment and consent bias in the sample.

Outcome measures for the many possible research aims will include, for example, measures of disease activity (e.g., complete remission defined as proteinuria <300mg/day adjusted for body surface area, change in proteinuria, or change in urine protein/creatinine ratio); measures of eGFR (e.g., time to a fixed eGFR loss, time to 40% or 50% reduction in eGFR, and eGFR slope); time to ESKD or death; time to cause-specific events (e.g., infection, thrombosis, malignancy); and PRO measures. Changes in continuous outcomes, such as urine protein and eGFR, will be graphically depicted using restricted cubic splines. Semi-parametric models will be constructed to identify distinct subgroups within the population based on clusters of trajectories. Events such as proteinuria remission, GFR slope, and 50% decline in eGFR will also be described.

To identify predictors of renal and non-renal outcomes, including therapeutic response, time-to-event analyses will be performed. Potential predictors may include clinical characteristics, genetic markers, and/or novel biomarkers. Analyses of longitudinal factors, e.g., a slope or change of a factor over time, would involve non-intersecting measurement periods for the predictor factor and follow-up periods for the outcome. Mixed longitudinal regression models will be applied for disease progression measures such as eGFR. Patient-level identifiers will be included as a random effect in these models. Time-to-

event Cox regression models will be used when analyzing defined outcomes and will use left truncation to allow each patient's experience to contribute to the time period after biopsy that corresponds to their study follow-up period. For factors likely to be influenced by treatment-by-indication bias, we will evaluate whether techniques such as instrumental variables analysis are appropriate. Predictive power of the model will be evaluated using log-likelihood tests and, when evaluating analyses where absolute predictive accuracy is more important than relative predictive accuracy, the c-statistic. Other approaches, including the net reclassification index may also be used as appropriate.

Special consideration will be applied when analyzing molecular biomarkers. For example, to screen a large number of potential predictors without losing analytical power, analyses will be performed in a hypothesis-generating manner. Regression analysis will be performed to obtain p-values for the association of outcome with a biomarker, and a threshold value of false discovery rate would be set to determine a pool of potentially important biomarkers. Selected biomarkers will be analyzed and grouped according to the relevance of their biological functions. Panels will be further tested for the association with outcome via ridge regression.

5. HUMAN SUBJECTS

5.1. PROTECTION OF HUMAN SUBJECTS

5.1.1. Institutional Review Board

This study and analysis will be performed under IRB oversight. Prior to the initiation of the study, an IRB approval for study of human subjects will be obtained separately from the IRB of each of the PCCs and the Data Coordinating Center (DCC). Revisions to the study protocol and changes in the study design will also be submitted to the individual IRBs for approval prior to implementation.

Subjects will be enrolled in the protocol with full informed consent which will include the gathering of protected health information (PHI), the collection of blood and urine specimens beyond that normally performed for clinical care, sharing of archived kidney tissue specimens collected for routine clinical care, and the collection of medical and PRO information at defined intervals.

5.1.2. Patient Confidentiality

Special procedures for ensuring patient confidentiality will be implemented. Data transmission and the distributed data systems have multiple layers of security, as discussed below in Section 7, Study Management. Each study subject will be assigned an identification number. Only this number will be used to identify subjects in any individual tabulation. The PHI that is collected will represent the minimum necessary to successfully execute the study.

Personally identifying information, such as name and e-mail address entered into the database at the site level, will only be visible to study personnel accessed through a triple password regimen. The information is encrypted at the site level. Site personnel have the decryption key, and it is not available to the DCC. It is expected that only group data will be published. If individual subject data are to be published, no identifying information will be included. The study files will be maintained in a secure location as described below. Access to computerized data will be restricted to study personnel. Password authorization will be enforced.

Authorized representatives of the Sponsor, the National Institute of Diabetes and Digestive and Kidney Diseases (NIDDK), National Institutes of Health (NIH), participating clinical institutions, DCC monitoring

staff, as well as the IRB, have access to medical records and records from participation in this study. Such access is necessary to ensure the accuracy of the findings.

5.1.2.1. Certificate of Confidentiality

To help protect participant privacy, a Certificate of Confidentiality will be obtained from the NIH. With this Certificate, the researchers cannot be forced to disclose information that may identify a study participant, even by a court subpoena, in federal, state, or local civil, criminal administrative, legislative or other proceedings. The researchers will use the Certificate to resist any demands for information that would identify a subject, except as explained below.

The Certificate cannot be used to resist a demand for information from personnel of the United States Government that is used for auditing or evaluation of federally funded research projects or for information that must be disclosed in order to meet the requirements of the federal Food and Drug Administration (FDA).

Even with the Certificate of Confidentiality, the investigators continue to have ethical and legal obligations to report child abuse or neglect and to prevent an individual from carrying out threats to do serious harm to themselves or others. If keeping information private would immediately put the study participant or someone else in danger, the investigators will release information to protect the participant or another person.

5.1.3. **Informed Consent**

The consent process may differ somewhat by PCC, according to local IRB guidelines. Participants will be asked to complete all study procedures. However, each study participant is able, during any study visit, to decline one or more of the data collection procedures without withdrawing from the study.

Each PCC will prepare an informed consent document following the template provided by the DCC and modified to meet their local IRB requirements. The initial informed consent document will be signed and dated by the participant before initiation of any study-related activity.

Before obtaining a potential participant's signature on the informed consent document, the local study investigator or his/her designee will review the details of the consent form orally with the potential participants and answer any questions the participant has concerning involvement in the study. The original signed consent form will be stored at the PCC, and a copy of the signed consent form will be given to the participant.

5.1.4. **Risks to the Patient**

Patients enrolled in this study will experience more than the normal amount of testing that is customary for their clinical care. Additional time will be required for the gathering of medical and PRO information. Blood and urine will be collected and stored for special tests and archival storage which are not normally required for clinical care. Venipuncture carries risks of pain and bruising at the puncture site. There is also a risk of anxiety and a small risk of dizziness associated with blood draws.

5.1.5. **Unauthorized Data Release**

There is always the theoretical possibility of unauthorized release of Health Insurance Portability and Accountability Act (HIPAA) PHI about subjects. Such disclosure would be extremely unlikely to involve a threat to life, health, or safety but would be a serious invasion of the subject's privacy. It is conceivable that such disclosure could have psychological, social, or legal effects on the patient. The standard

security procedures will effectively minimize the risk of unauthorized disclosure of data. All study personnel who have access to patient data will be educated regarding the need to protect confidentiality and the procedures to be followed to ensure such protection. All relevant staff will also be required to sign a medical record confidentiality agreement. The computer systems on which data are maintained use password protection procedures to prevent access by unauthorized users. Data to be used for analysis will contain only the assigned identification numbers.

5.1.5.1 Adverse Event Monitoring

- ***Definition of Adverse Event:*** An adverse event (AE) is any untoward medical occurrence or unfavorable and unintended sign in a research subject that occurs during or as a result of a research procedure. For this study, the majority of the procedures are standard clinical care, and adverse effects of clinical care will be tracked as complications but will not be considered adverse study events. Each center will review the list of study procedures and identify the specific procedures that are not standard-of-care at their institution, and these will be considered research procedures. Complications that are a result of research procedures will be reported and tracked as AEs.

- ***Assessment of Event Severity and Relationship to Study Procedure/Treatment:*** The modified World Health Organization (WHO) grading system will be used for grading severity of AEs (See Manual of Procedures). For AEs not covered by the modified WHO grading system, the following definitions will be used:

Mild:	Awareness of sign, symptom, or event, but easily tolerated
Moderate:	Discomfort enough to cause interference with usual activity, and may warrant intervention
Severe:	Incapacitating with inability to do usual activities or significantly affects clinical status, and warrants intervention
Life-threatening:	Immediate risk of death

The investigator must also assess the relationship of any AE to the research procedure, based on available information, using the following guidelines:

Unlikely related:	No temporal association, or the cause of the event has been identified; or the procedure cannot be implicated
Possibly related:	Temporal association, but other etiologies are likely to be the cause; however, involvement of the procedure cannot be excluded
Probably related:	Temporal association; other etiologies are possible, but unlikely

- ***Definition of Serious Adverse Events:*** A serious AE (SAE) is any adverse experience that results in any of the following outcomes:
  - Death;

- Life-threatening AE (i.e., one that places the subject, in the view of the investigator, at immediate risk of death from the AE as it occurs);
- Persistent or significant disability/incapacity;
- Required in-patient hospitalization, or prolonged hospitalization;
- Congenital anomaly or birth defect.
- Additionally, important medical events that may not result in death, be life-threatening, or require hospitalization may be considered an SAE when, if based upon appropriate medical judgment, they may jeopardize the subject and may require medical or surgical intervention to prevent one of the outcomes listed in this definition.

- ***Reporting Responsibility:*** Only AEs possibly or probably related to this observational study must be recorded. Events related to the disease or therapy of the patient need not be reported as *Observational Study-Related* AEs. The onset and end dates, severity, and relationship to study procedure(s) will be recorded for each AE. Any action or outcome (e.g., hospitalization, additional therapy, etc.) will also be recorded for each AE. Subjects will be questioned and/or examined by the investigator or his/her designee for evidence of AEs.

  All AEs and SAEs reported by the investigator to the CureGN DCC will be reviewed. The DCC may request additional information from sites for analysis of these events. Sites will report SAEs related to the study according to the time frames outlined below.

  All events that are serious and related (possibly or probably) to the observational study must be reported to the DCC within 24 hours of the investigator being informed of the event. Follow-up information about a previously reported serious and related AE may be reported to the DCC within 7 working days of the investigator receiving the information; however, important follow-up information must be submitted within 24 hours. All deaths related to a study procedure must be reported to the DCC within 24 hours of the investigator being informed of the event.

5.2. Benefits to the Patients

There are no direct benefits to subjects for participation in the study. Potential benefits include the satisfaction of altruism and detection of new information that may improve the management of patients with glomerular diseases in the future.

5.2.1 Inclusion of Women

This is a multi-center study drawing on a clinical population from PCCs in the United States, Canada, and Europe. Women will be recruited into the study. It is envisioned that the representation of women will correspond to the fraction of women in the population diagnosed with biopsy confirmed primary glomerular diseases emanating from FSGS, MCD, IgAN, and MN. Special efforts will be incorporated into the recruitment process to facilitate the optimal inclusion of women with these diseases in the study.

5.2.2 Inclusion of Minorities

Racial and ethnic minority groups will be recruited into the study. It is envisioned that the representation of persons comprising racial and ethnic minority groups will correspond to the fraction of those groups in the population diagnosed with biopsy confirmed primary glomerular diseases emanating from FSGS, MCD, IgAN, and MN. Special efforts will be incorporated into the recruitment process to facilitate the optimal inclusion of persons of racial and ethnic minority groups. Recruitment will be monitored to ensure adequate representation of minority groups.

5.2.3 Inclusion of Children

Children will be recruited into the study. It is envisioned that the representation of children will correspond to the fraction of children in the population diagnosed with biopsy confirmed primary glomerular diseases emanating from FSGS, MCD, IgAN, and MN. Special efforts will be incorporated into the recruitment process to facilitate the optimal inclusion of pediatric cases in the study.

5.2.4 Observational Study Data Safety and Monitoring Plan

Accepted principles of data and safety monitoring will be observed throughout the conduct of the CureGN study. The NIH will appoint an independent External Expert Group (EEG) that will provide study oversight. The EEG will approve the study protocol prior to enrollment and will also approve all subsequent protocol revisions.

Each PCC principal investigator will be responsible for monitoring the enrollment of subjects and submission of data to the DCC. The DCC will be responsible for monitoring for effective conduct of the protocol and accurate and timely data submission.

Training of study coordinators and study monitoring activities for the PCCs will be conducted by the DCC to ensure patient confidentiality and privacy and to maximize the reliability, accuracy, and timeliness of study data. The lead PCC of each consortium will be responsible for the training of study coordinators and study monitoring activities for its sub-sites.

Data will be routinely exported from the system, examined for accuracy and completeness, and backed up to secure storage devices. Upon completion of data collection, final processing and cleaning of data will be conducted. A technical report detailing specific project methodology, response rates, and other details will be produced.

6. STUDY ORGANIZATION

6.1. PARTICIPATING CLINICAL CENTERS

The PCCs will have primary responsibility for participant enrollment, maintaining acceptably high rates of follow-up and data collection, obtaining data of high quality, and interpreting, presenting, and publishing findings from the study. Four PCCs serve as clinical center hubs, with additional clinical sites responsible for study participant enrollment, retention, and protocol implementation under the guidance of the respective PCC.

6.2. DATA COORDINATING CENTER

The DCC is located at the University of Michigan Health System and Arbor Research Collaborative for Health, both located in Ann Arbor, MI. The DCC contributes content area expertise and shares in scientific leadership of CureGN. The DCC has developed a communication infrastructure that includes meetings, teleconferences, electronic mail and bulletins, interactive web-based encounters, and written correspondence. The DCC assists in preparation of scientific publications. The DCC has the major responsibility of creating and maintaining the study database and data collection systems for CureGN, ongoing evaluation of data quality and performance monitoring of the PCCs, and statistical analyses of the data. The DCC also maintains a comprehensive Manual of Procedures that will govern the conduct of the study.

6.3. STEERING COMMITTEE

The primary governing body of the study is the Steering Committee, which includes each of the Principal Investigators of the PCCs, the Principal Investigator of the DCC, a Chairperson appointed by the NIDDK, and the NIDDK Project Officers. Each PCC, the DCC, the NIDDK, and the Steering Committee chair has one vote for decisions brought to the Steering Committee. The Steering Committee is charged to develop, approve, and update the study protocol as needed, and to develop policies for the study pertaining to access to participant data and specimens, ancillary studies, performance standards, and publications and presentations. The Steering Committee meets to discuss study progress and to resolve problems arising during study conduct. The Steering Committee may establish subcommittees to further develop or manage specific components of the study, such as ancillary studies or publications. Working groups may also be established, e.g., to prepare manuscripts and presentations.

7. STUDY MANAGEMENT

7.1. Data Collection, Data Collection Forms, and Data Entry

The DCC will utilize the web-based *CureGNLink* as the data management nucleus for the CureGN studies. *CureGNLink* is a database platform developed by Arbor Research Collaborative for Health.

The DCC will utilize *CureGNLink* to create electronic case report forms to capture all relevant study data for the core study and all investigational/research protocols that are developed and implemented during the course of the study. The *CureGNLink* system allows real-time monitoring of study data for protocol adherence, quality assurance, AE reporting, discrepancy reporting, and other trends.

7.2. Data Management

Study data will be entered into the electronic data entry system by study coordinators at each study site. These data will be encrypted and transferred to the DCC and stored on a secure server at Arbor Research Collaborative for Health. Access to the server and data entry system is limited and requires a unique username and password combination. The servers are backed up daily and physically stored in a locked facility. All analysis of the data sets will utilize de-identified (coded) data sets. Transfer of batch data from site-specific databases or other electronic data sources will be assessed individually for each clinical site based on feasibility and data quality.

7.3. Quality Control and Database Management

The first steps in ensuring protocol compliance are good protocol design and careful orientation of study personnel. Prior to study initiation at any of the PCCs, the DCC will organize a Training and Certification session for study coordinators/data entry personnel.

The electronic data entry system will have built-in data checks as part of study quality assurance. Protocol compliance will be assessed by monitoring the submission of data at required intervals. Data inconsistencies and discrepancy reports will be reviewed by the Clinical Monitors so that necessary queries can be generated and sent to the PCC study sites for verification and resolution.
Periodic requests may be generated for the submission of random source documents to assess the quality of data acquisition and data entry at each site. In addition, the Clinical Monitor or Project Manager will visit lead PCC at least once a year to review source documents, monitor regulatory compliance, and assess protocol adherence. The lead PCC will be responsible for data quality and data entry timeliness at their sub-sites.

In addition to source document verification, the Clinical Monitor and Project Manager will produce reports from the database to look for inconsistencies in submitted data, particularly for repeated measures data elements, even if data do not fall outside of built-in validation routines. Studies of intra-subject and inter-subject data variability by PCC as well as intra-center and inter-center data variability will be used to further ascertain random or systematic data quality issues.

7.4. Data Security/Data Transfer

Personnel at each study center will collect and enter data into the web-based data entry system. The following data security contingencies are in place:

- Compliance with Industry Standards Regarding Data Security (HIPAA and 21 CFR Part 11)
- Audit trails are maintained for all activity and all changes to any data element
- All servers, web servers, firewalls, etc. are configured and maintained according to industry best practice guidelines for backup, security, continuity of operations, and protection of PHI
- All data are available only to authorized users from each site after secure login with encryption, with all site activity audited at the user level
- All transmissions between the Internet and the database are encrypted using a 128-bit encryption algorithm
- There is a comprehensive security plan in place

Detailed instructions on the use of the database platform, data element definitions, and a code list will be provided in a Manual of Procedures. Each study site will be provided a copy of this manual, and the entire manual will be available on the study web site, and in the Help area of the database user interface.

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676 # 9. APPENDICES