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CLINICAL RESEARCH

Implementation of a Kidney Genetic Service Into the Diagnostic Pathway for Patients With Chronic Kidney Disease in Canada

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Introduction: Genetic kidney disease (GKD) accounts for 10% to 20% of chronic kidney disease (CKD). Genetic testing using gene panel or targeted exome sequencing (ES) can confirm GKD; however, integration into clinical practice has been hampered by small studies, selective populations, and data predominately derived from research settings. Using prespecified clinical referral criteria and a diagnostic pipeline, we performed a prospective cohort study describing diagnostic efficacy and clinical utility of genetic assessment in patients with CKD.

Methods: We analyzed a prospective cohort of 300 participants (256 families) referred to a kidney genetics clinic, between March 2020 and March 2024. Testing strategies included gene panels, and if negative or unsuitable, targeted ES analysis. Testing was performed for the detection of variants in genes known to cause CKD.

Results: We identified a causative variant in 33% of families (85/256). Diagnostic yield increased from 23% ($n = 70/300$) from gene panel alone, to 34% ($n = 103/300$) with comprehensive testing. The median time from first diagnosis of CKD to genetic assessment was long at 10.4 years. Following genetic assessment, the median time to receive a positive genetic result was 2.9 months. Multiple levels of clinical utility were recorded in patients receiving a genetic diagnosis, varying across CKD subtype.

Conclusion: Instituting referral guidelines and a standardized testing algorithm established a genetic diagnosis in one-third of participants, providing insight into the viability of integrating genetic assessment in the CKD diagnostic pathway. Considering the potential for clinical utility, strategies to reduce the time from CKD diagnosis to genetics assessment are needed.

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Genetic kidney disease (GKD) is increasingly recognized as a primary cause of CKD in adults, with data now indicating that 10% to 20% of CKD is due to single gene mutations.[1,2] Data show that nephrologists often report uncertainty in recognizing

which patients need a genetic assessment and find it difficult to navigate the genetic testing process.[3–5] To streamline genetic referrals, it is important to develop clinical referral guidelines and clear testing algorithms. For example, a recent systematic review by our team shows that the application of selection criteria can result in the diagnosis of GKD in a high proportion of patients with CKD.[6] Studies consistently show that the probability of diagnosing GKD is higher in the presence of genetic risk factors, such as a family history, extrarenal disease, or certain subtypes of CKD.[7–10] Conversely, without clear testing guidelines, the diagnostic yield after testing can be extremely low.[11] When considering which genetic test is the most appropriate in patients with CKD, it is important to consider that there are now over 600 monogenic causes of CKD described.[12] Technological improvements have made it possible to effectively screen for variants in hundreds of kidney disease genes through targeted gene panel, ES, or genome sequencing[7,13] at ever reducing costs.[14]

Despite the increasing recognition of the burden of GKD and data demonstrating the cost-effectiveness when performed early in the diagnostic pathway,[9,15] genetic testing has not been routinely integrated into the CKD diagnostic pathway. The current practice usually entails conducting genetic assessments only after exhausting all other diagnostic avenues. By shifting genetic assessment to an earlier point in the diagnostic pathway and shortening the time to diagnosis, we can potentially offer a more cost-effective diagnostic strategy.[15] In addition to avoiding invasive tests, genetic assessment can improve patient outcomes by facilitating a more rapid and accurate diagnosis, allowing for genetic counseling, and permitting institution of disease-specific treatment.[16] For this reason, we developed clinical referral guidelines to provide guidance on when a genetic assessment should be considered. We hypothesize that clinical referral guidelines can streamline genetics assessment in patients with CKD, resulting in a high diagnostic yield following genetic testing.

Here, we report the implementation experience of a dedicated kidney genetic clinic in an urban academic center in Canada. Using predefined clinical referral guidelines and a diagnostic pipeline, we detail the outcomes of 300 patients assessed in this clinic over a 4-year period, describing the diagnostic efficacy, testing modalities, and clinical utility of genetic testing in CKD.

METHODS

Recruitment

The procedures followed in this study were approved by the Western University Research Ethics Board (#115160). In total, 322 participants were referred to a dedicated kidney genetic clinic, of which 300 study participants were recruited. This clinic is in London, Ontario, Canada and serves an area with a population of >2.5 million, in Southwestern Ontario, Canada. The clinic was established in March 2020 and data was collected from March 1, 2020, to March 4, 2024. Clinical referral guidelines for kidney genetic assessment were initially developed through a review of the literature and were based on the best available evidence as of December 2019 (Table 1).[17–37] We shared these guidelines with the nephrology, urology, pediatric, and genetic communities in our service area 3 months before establishing the clinic and then annually thereafter (Table 1).[17–37] Informed consent was obtained from all participants or the substitute decision maker. Following informed consent, we recorded clinical and pedigree data using a standardized questionnaire with additional data extracted from the medical records. Ethnicity was self-reported, and race was documented after clinical assessment. Additional clinical data obtained during the study period was integrated into the analysis.

Study Design

All patients were reviewed in a dedicated kidney genetics clinic if they met any 1 of the clinical referral criteria for genetics assessment (Table 1).[17–37] The team comprised of a nephrologist with expertise in GKD, genomic analysis and interpretation, a genetic counsellor, and a nephrology trainee. Patients were recruited based as either the affected index or affected family members. We defined the time of CKD onset as the date of first review by a nephrologist. We used the Kidney Disease Improving Global Outcomes definition of CKD and end-stage kidney disease (ESKD).[38] Following recruitment and genetic assessment, patients were divided into diagnostic categories based on the presumed etiology of CKD (Figure 1). If the etiology of CKD was known, the diagnostic pathway pursued was a phenotypic-driven gene panel, performed in a Clinical Laboratory Improvement Amendments–certified laboratory. If the etiology of CKD was unknown, a comprehensive testing strategy was employed. Ontario has a publicly funded health care system which covers the cost of gene panel testing for eligible patients. Because of the lack of provincial funding for comprehensive gene panel testing from March 2020 to December 2021, the comprehensive testing strategy was exome sequencing (ES) analysis (i.e., primary ES). From January 2022 to March 2024, a comprehensive kidney disease panel was employed as the first-line diagnostic test for participants with CKD of unknown etiology. Access to clinical ES for patients

Table 1. Referral criteria for the kidney genetics clinic

Major referral criteria	Subcategory of Referral Criteria	Source of evidence based on best available in December 2019
Positive family history	1. Chronic kidney disease (CKD) 2. Recurrent kidney stones 3. Nephrocalcinosis 4. Hypertension 5. Nephropathy or nephritis including Alport Syndrome (suspected or confirmed) 6. Consanguinity	Connaughton *et al.*,[7] 2019, Lata *et al.*[10] 2018 Halbritter *et al.*,[17] 2018, Halbritter *et al.*,[18] 2015 Daga *et al.*[19] 2018, Dickson and Sayer,[20] 2020 Connaughton and Hildebrandt[21] 2020 Kashtan *et al.*,[22] 2018 Braun *et al.*,[23] 2016
Young age of CKD onset	Early onset CKD at age < 50 yrs	Mann *et al.*,[24] 2019
CKD of unknown etiology which includes the following criteria	1. Ultrasound imaging that demonstrates small echogenic kidneys 2. Kidney biopsy not feasible due to bilateral small echogenic kidneys 3. Kidney biopsy had indeterminant findings	Hays *et al.*,[25] 2020
Structural abnormalities in the kidney or genitourinary tract consistent with congenital anomalies of the kidney and urinary tract including the following:	1. Renal agenesis 2. Renal hypodysplasia 3. Vesicoureteral reflux	van der Ven *et al.*,[26,27]2018
Cystic kidney disease	1. Normal sized cystic kidneys or atypical cystic kidney disease 2. Medullary cystic kidney disease 3. Suspicion of Nephronophthisis 4. Cysts of uncertain etiology 5. Polycystic kidney disease where there is unclear or negative family history. 6. Uncertainty regarding the diagnosis due to either atypical or mild features. 7. Provider discretion where a genetic diagnosis may have an impact on clinical management (i.e., living related donor selection, preconception, or prenatal screening, screening of at-risk family members etc.)	Braun and Hildebrandt,[28] 2017 Bleyer *et al.*,[29]1993 Wolf and Hildebrandt,[30] 2011 Cornec-Le Gall *et al.*,[31] 2018 Armstrong and Thomas,[32] 2019 Grosse and Khoury,[33] 2006
Persistent electrolyte disturbances consistent with renal tubular dysfunction and no other cause identifiable where secondary causes have been excluded	1. Recurrent hypokalemia or hyperkalemia (of unknown cause) 2. Renal tubular acidosis (definitive or possible with clinical and biochemical evidence of disease) 3. Hypercalciuria (of unknown cause) 4. Hypomagnesemia (of unknown cause) 5. Other persistent electrolyte disturbances of unknown causes	Ashton *et al.*,[34] 2018 Alexander *et al.*,[35] 2019 García-Castaño *et al.*,[36] 2019 Wolf, [37] 2017 Wolf, [37] 2017
Extrarenal features of disease	1. Evidence of a multisystem disease 2. Early onset gout with renal impairment 3. CKD in the presence of extrarenal features including but not limited to the following: a. Retinitis pigmentosa or other eye pathologies b. Intellectual disability/learning disability c. Seizure disorder d. Dextrocardia e. Hepatobiliary disease such as congenital hepatic fibrosis f. Syndromic features such as polydactyly, clinodactyly and other skeletal abnormalities g. Deafness onset at age < 50 yrs	Connaughton *et al.*,[7] 2018

CAKUT, congenital anomalies of the kidneys and urinary tract; CKD, chronic kidney disease.
Major referral criteria are provided with additional specific subcategories of each referral criteria. Please note these criteria were devised based on the best available evidence at the time (December 2019). Following analysis of these data, further review, and refinement of the referral criteria, incorporating all new data from January 2020 is required.
Patients must meet at least 1 of the major referral criteria by meeting at least 1 of the following subcategory referral criteria to be accepted for genetic assessment.

with CKD at the time of this study was limited. As a result, in most cases, ES was performed in a research laboratory in patients whose etiology remained unknown following gene panel testing (comprehensive panel included), so called secondary exome sequencing analysis. As per standard practice, all the variants identified on research-based testing were subsequently confirmed in a Clinical Laboratory Improvement Amendments–certified laboratory by targeted variant testing. Clinical grade primary exome sequencing analysis was performed in 6 cases, all of whom met the provincial criteria for exome analysis (Supplementary Table S1). Three participants had microarray performed because of the presence of developmental delay and/or learning disability. Targeted-variant testing was performed for affected family members with known familial variant(s).

All genetic results were reviewed by a multidisciplinary team, which included a physician with expertise in kidney genetics, a genetic counselor, and a

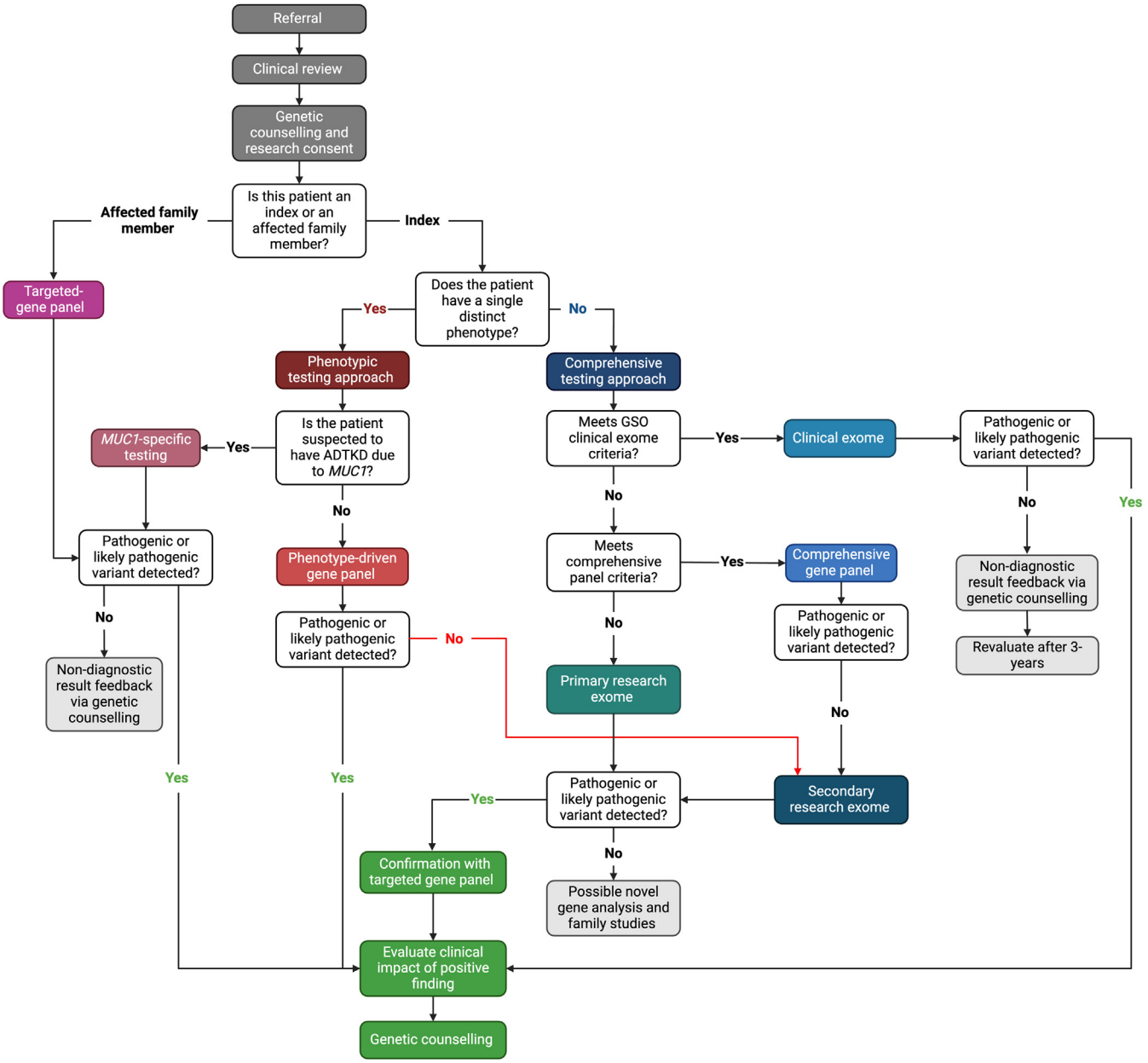

Figure 1. Study design from patient referral to genetic testing result disclosure. Index participants are defined as the first participant in a family to be recruited to the study. Pathogenic and likely pathogenic variants are defined as per the American College of Medical Genetics Guidelines.[39] Primary research exome is defined as research exome sequencing as the first-line test, whereas secondary research exome is defined as exome sequencing as a second-line test after a negative panel test. Clinical exomes were performed through Genome-wide Sequencing Ontario (GSO), when the participant met the provincial indications for testing (Table S1).

researcher with expertise in genomics analysis and sequencing interpretation. We defined solved participants as those in whom a pathogenic or likely pathogenic variant was identified following the American College of Medical Genetics Guidelines[39]; and following our multidisciplinary team review, the genotype was concordant with the disease phenotype. The solve rate was defined as the proportion of solved participants divided by the total number of participants tested. For these analyses, we reported both patient ($N = 300$) and familial ($N = 256$) solve rates. In patients who remained unsolved after initial testing or in whom variants of uncertain significance (VUS) were identified, routine reanalysis was performed at an 18-month interval.[40] Suspicious VUS were defined as variants that matched the phenotype but did not meet the American College of Medical Genetics criteria for pathogenicity.[39] During reanalysis, all new data were integrated into the analysis of all previously identified VUSs, which in some cases warranted a change in variant classification. New data included segregation data or family studies, new clinical data, including information gathered through

reverse phenotyping, data on population frequency, functional data, or reports of exact variant data in the literature.

Genetic Testing

All the genetic testing results disclosed to patients were confirmed in a Clinical Laboratory Improvement Amendments–certified laboratory (Supplementary Table S2). Participants with clinical characteristics of autosomal dominant tubulointerstitial kidney disease (ADTKD; positive family history with an autosomal dominant mode of transmission, CKD, and bland urinary sediment) underwent targeted gene testing for the +C *MUC1* variant.[41] ES was performed using Agilent SureSelect (Agilient Technologies, Santa Clara, CA) enrichment followed by sequencing on the Illumina Novaseq6000 platform (Illumina, Inc., San Diego, CA) following standardized protocols. Exome data was overlapped with a list of 694 monogenic genes known to cause kidney disease developed by our laboratory (Supplementary Table S3). To annotate ES data, we used an internally designed custom automated workflow in CLC Bio genomics workbench (CLC bio version 12, QIAGEN Digital Insights) to perform sequence alignment, call variants, and to produce targeted region coverage statistics for variant discovery.[42] To annotate variant data, the ANNOVAR pipeline with customized downloads and filtering strategies was used as previously described.[43] Details are in the Supplementary Methods.

Health Outcomes Related to Clinical Utility

Health outcome measures were captured after disclosure of genetic results and repeated at study close (March 2024) across 6 predefined levels of clinical utility. This was performed by the multidisciplinary team immediately following the disclosure of genetic testing results and repeated at study close through medical chart review. The predefined measures of clinical utility were adapted from a toolkit devised by Hayeems *et al.*,[44] which was developed using the Fryback and Thornbury model for evidence collection for clinical utility of genomics sequencing.[44] These health outcome measures assess 6 levels of clinical utility, adapted to capture all health outcomes pertaining to patients with CKD. Health outcome measures include the following: (i) diagnostic accuracy to determine if genetic testing resulted in a diagnosis, dual diagnosis, or partial diagnosis; (ii) diagnostic thinking efficacy, including prognostic clarity, family history revised or refined, use of additional consultations, laboratory tests, or laboratory imaging, and/or avoidance of additional tests; (iii) therapeutic efficacy including referral to genetic counseling, initiation, alteration, or cessation of a treatment; referral to a subspecialist; change in management, including surgical procedure (i.e., kidney transplantation) or surveillance; and referral for educational services, clinical trials, or support group; (iv) patients outcomes efficacy, including resolution of diagnostic confusion and reduction in diagnostic odyssey for patient and/or family; (v) societal efficacy, including cascade testing and family member risk identification; and (vi) retrospective clinical utility is an additional level of clinical utility, which recorded the potential or possible health outcomes *if* testing had been performed earlier in the diagnostic pathway. We report the clinical utility as a ratio of the events occurring in solved patients (including incidental findings) ($n = 105$), and separately in unsolved (negative result or VUS) patients ($n = 182$). Patients with pending results ($n = 13$) were not included in these analyses.

Statistical Analysis

Descriptive statistics were expressed using frequencies and proportions. We planned an *a priori* analysis of risk factors associated with GKD, including positive family history, presence of extrarenal features or multisystem disease, ESKD at time of testing, ESKD onset at age < 50 years versus ≥ 50 years, age of onset of CKD, pediatric versus adult onset, and onset of CKD at age < 18, 18 to 34, and ≥ 35 years. Using the binom package in R version 4.3.2,[45] we used univariate Pearson's Chi-square tests and reported proportion with 95% confidence intervals; significance threshold was $P < 0.05$. For comparison of differences across groups, we used 2-way analysis of variance. We also assessed the time to a genetic diagnosis in participants with a positive genetic testing result, by determining the time from a clinical CKD diagnosis to both time of genetic assessment and time to disclosure of a positive test result ($n = 103$). In all participants ($N = 300$), we determined the time from referral to genetic assessment. For the quantitative data, we inspected a histogram to determine data distribution. For normally distributed data, we reported mean and standard deviation (SD). For nonnormal distribution, we reported the median and interquartile range (IQR).

RESULTS

Referral and Recruitment

In a 4-year period, 322 patients meeting referral criteria were assessed in the kidney genetics clinic (Figure 2). Twenty-two patients declined participation and therefore, were not included in the analysis. Females were more likely to decline participation (59%) with a median age of 56 (IQR: 29) years. Of those who declined, 15 did proceed with clinical genetic testing

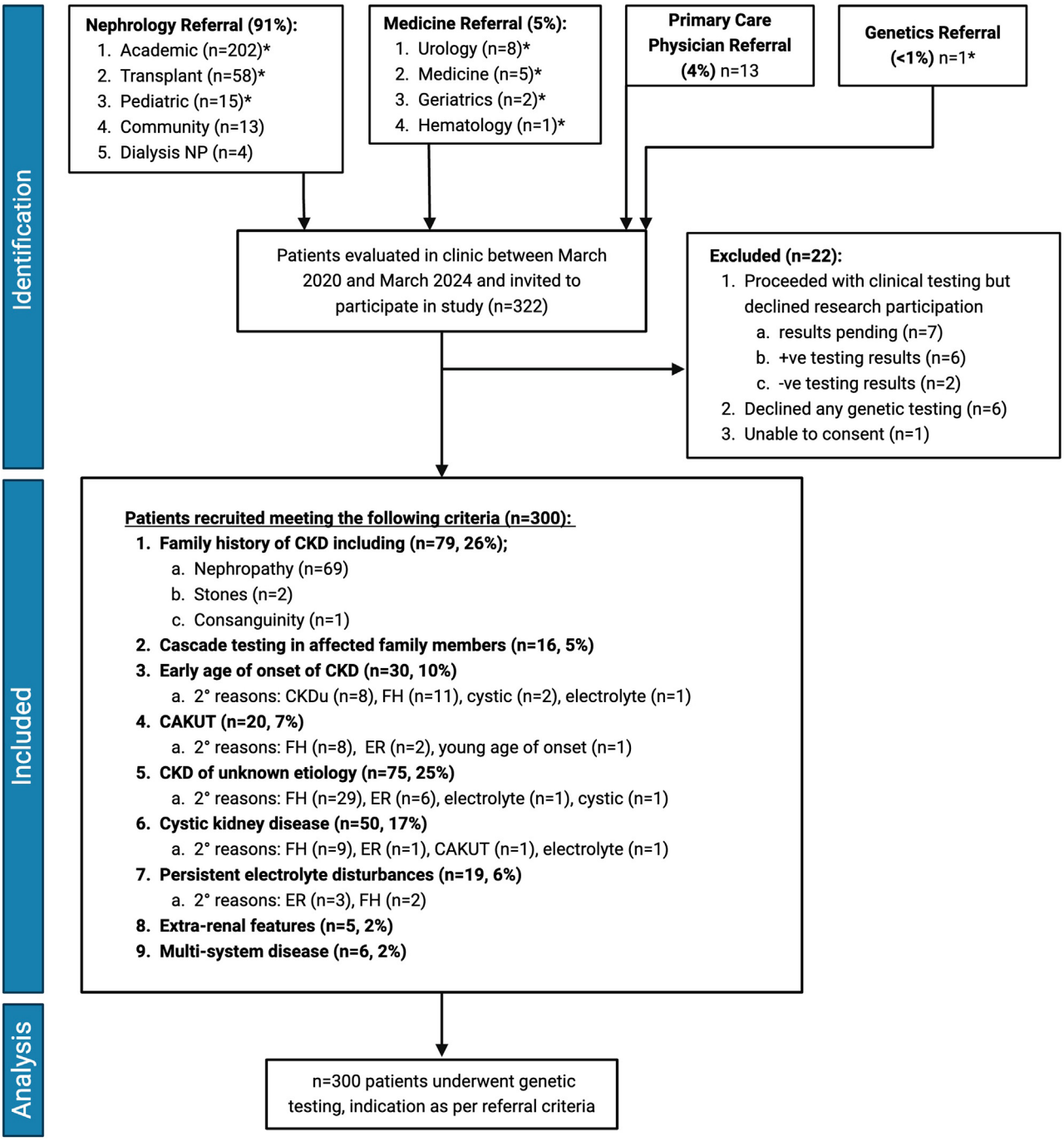

Figure 2. Strengthening the reporting of observational studies in epidemiology (STROBE) flowchart for participant referral and recruitment. Referrals from nephrology consisted of; academic (referral from an academic center), transplant (referral from transplant nephrology center for assessment of potential transplant recipient or someone who has received a transplant in the past), pediatric (referral from pediatric nephrology), community (referral from a community nephrologist), and dialysis nurse practitioner (referral from a dialysis nurse practitioner). Each participant had a primary reason for referral to the kidney genetic clinic, which is their main concern for genetic risk, and in some cases, a secondary (2°) reason was also provided by the referring doctor, which would also increase their risk of genetic kidney disease. The asterisk (*) represents referrals from clinics associated with an academic center affiliated with a university. CAKUT, congenital anomalies of the kidney and urinary tract; CKD, chronic kidney disease; CKDu, chronic kidney disease of unknown etiology; ER, extrarenal features; FH, family history; stones; nephrocalcinosis and nephrolithiasis; 2°, secondary reason for referral.

with a diagnostic yield of 27% ($n = 6/22$), whereas 7 declined any form of genetic testing following assessment. The major indications for referral were a positive family history of CKD ($n = 95$, 32%) and CKD of unknown etiology ($n = 75$, 25%), with many participants ($n = 65$, 22%) meeting multiple indications for kidney genetics assessment (Table 1).[17–37] The number of referrals increased from 2020 to 2024

Table 2. Cohort characteristics

	Total		Adult-onset CKD		Pediatric-onset CKD	
	300 (256)		232 (197[a])		68 (68[a])	
Clinical characteristic	***n***	**%**	***n***	**%**	***n***	**%**
Sex						
Males	148	49.3	119	51.3	29	42.6
Females	152	50.7	113	48.7	39	57.4
Race						
Asian	24	8.0	19	8.2	5	7.4
Black	11	3.7	9	3.9	2	2.9
Hispanic	10	3.3	9	3.9	1	1.5
Indigenous	4	1.3	4	1.7	0	0
More than one race	3	1.0	3	1.3	0	1.5
White	183	61.0	148	63.8	35	51.5
Unknown	65	21.7	40	17.2	25	36.8
CKD						
Mean CKD age of onset (years ± SD)	32.2 ± 18.9	-	38.8 ± 15.8	-	8.6 ± 7.0	
ESKD						
ESKD present	124	41.3	100	43.1	24	35.3
Mean ESKD age of onset (years ± SD)	37.5 ± 17.7	-	41.5 ± 16.0	-	16.4 ± 8.9	
ESKD modality						
Kidney Transplant (single)	54	43.5	42	42.0	12	50.0
More than 1 kidney transplant	13	10.5	9	9.0	4	16.7
Hemodialysis only	34	27.4	32	32.	2	8.3
Peritoneal dialysis only	7	5.7	7	7.0	0	0
Hemodialysis with previous failed kidney transplant	16	12.9	13	13.0	3	12.5
Peritoneal dialysis with previous failed kideny transplant	13	10.5	10	10.0	3	12.5
Participants with failed kideny transplant	14	11.3	8	8.0	6	25.0
Reason for kidney transplant failure						
Recurrence of primary disease	3	21.4	1	12.5	2	33.3
Rejection	7	50.0	4	50.0	3	50.0
Unknown	4	28.6	3	37.5	1	16.7
Etiology of CKD						
Unknown	156	52.0	120	51.7	36	52.9
Presumed	144	48.0	112	48.3	32	47.1
Presumed etiology of CKD						
Hypertension	13	9.0	10	8.9	3	9.4
Diabetes	9	6.3	9	8.0	0	0
TKD	5	3.5	5	4.5	0	0
Cystic	33	22.9	31	27.7	2	6.3
CAKUT	23	16.0	13	11.6	10	31.3
GN	45	31.3	31	27.7	14	43.8
aHUS	3	2.1	3	2.7	0	0
Tubulopathy	4	2.8	3	2.7	1	3.1
Kidney Stones disease	6	4.2	4	3.6	2	6.3
Other	3	2.1	3	2.7	0	0
Other characteristics						
Prior native kidney biopsy	80	26.7	60	25.9	20	29.4
Extrarenal features	194	64.7	156	67.2	38	55.9
Positive family history	206	68.7	158	68.1	48	70.6
Total solved participants	103	34.3	76	32.8	27	39.7

aHUS, atypical hemolytic uremic syndrome; CAKUT, congenital anomalies of the kidney and urinary tract; CKD, chronic kidney disease; Cystic, cystic kidney disease; ESKD, end-stage kidney disease; GN, glomerulopathy; TKD, tubulointerstitial kidney disease.
[a]For 9 families, 1 family member had pediatric disease onset whereas the other family members had adult disease onset, therefore the number of families when separating by age of CKD onset is 9 higher than the overall number of families.
Numbers at the top of the graph represent total participants (*n*) and families (*n*) in brackets.

(Supplementary Figure S1) and included referral from nephrology (91%), internal medicine (5%), primary care physicians (4%), and clinical genetics (<1%) (Figure 2). The median wait time from receipt of referral to review in the clinic was 4.1 months (IQR: 5.2).

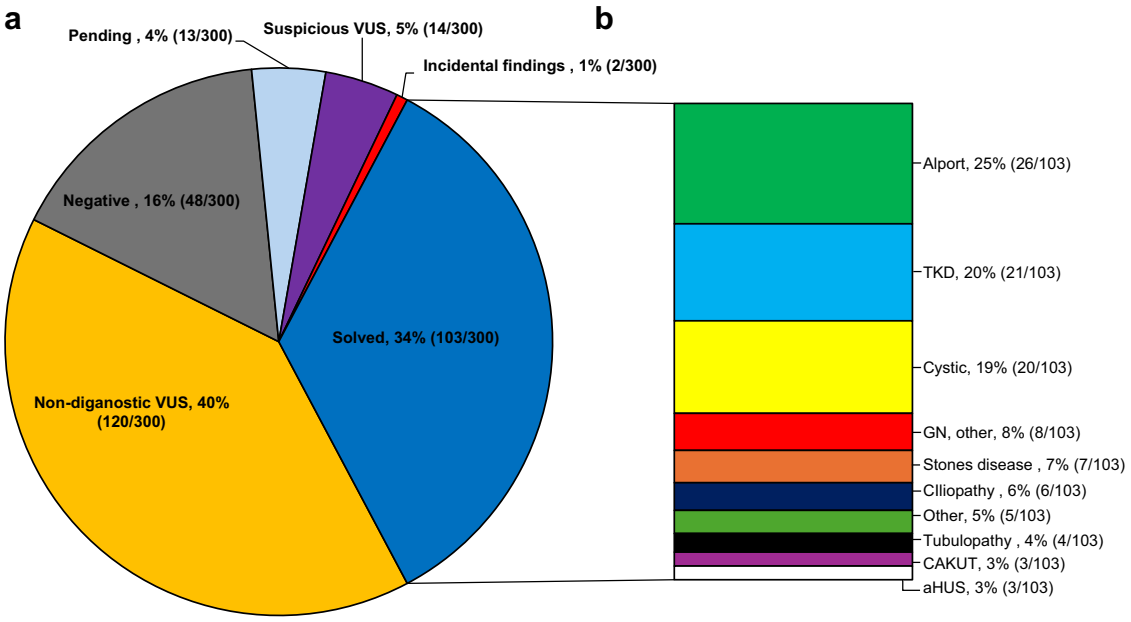

Figure 3. Overall diagnostic yield. (a) the overall diagnostic yield for 300 tested participants. The solved group represents participants with likely pathogenic or pathogenic diagnostic variants; the suspicious variant of unknown significance (VUS) group are participants with a suspicious VUS that cannot be upgraded; the pending group are participants with genetic testing sent out but results are pending as of March 4, 2024; the nondiagnostic VUS group are participants with a VUS that does not match their phenotype and is not suspected to be disease causing; the negative group are participants with no genetic findings. (b) Phenotypes identified in the 103 solved participants, including atypical hemolytic uremic syndrome (aHUS); Alport syndrome; congenital anomalies of the kidney and urinary tract (CAKUT); ciliopathies, including nephronophthisis; cystic kidney disease, including polycystic; glomerulopathy (GN), excluding Alport syndrome; and other kidney diseases, including amyloidosis, stones disease (nephrocalcinosis and nephrolithiasis), tubulointerstitial kidney disease (TKD), and tubulopathies.

Diagnostic Yield

In total, 300 participants from 256 families were included (Table 2). The mean age of CKD and ESKD onset was 32.2±18.9 and 37.5±17.7 years, respectively. There were slightly more females (n = 152/300) than males. Overall, 34% (n = 103/300) of participants from 33% of families (n = 85/256) received a genetic diagnosis (Figure 3a, Supplementary Tables S5 and S6). In solved participants, 96 had a likely pathogenic or a pathogenic variant detected on initial analysis. VUS were identified in 40% (n = 120/300) of the participants. Six participants had a suspicious VUS that was subsequently upgraded to a pathological classification following reanalysis. After reanalysis, 5% (n = 14/300) remain with a suspicious VUS, pending data from either family segregation studies or further functional data. These variants were not included in the overall diagnostic yield (Supplementary Table S7). Two participants had incidental findings of pathogenic variants in genes not related to their kidney disease (Supplementary Table S8).

Phenotypes and Genotypes Identified

The highest diagnostic yield was observed in participants referred with an assumed diagnosis of Alport Disease before genetic testing at 86% (n = 6/7), followed by ADTKD (80%, n = 4/5). Participants with CKD of unknown etiology had a diagnostic yield of 34% (n = 53/156) (Table 3). Alport Disease was the most common genetic diagnosis after testing (25%, n = 26/103), followed by ADTKD (20%, n = 21/103), and cystic kidney disease (19%, n = 20/103) (Figure 3b). Often, the specific genetic diagnosis was not suspected before genetic testing; for example, in genetically confirmed Alport cases, 77% (n = 20/26) were not

Table 3. Diagnostic yield per pretesting phenotype

Pretesting phenotype	Diagnostic yield (%)	Solved participants (n)	Total participants (n)
Alport disease	86	6	7
TKD	80	4	5
Other kidney diseases	67	2	3
Cystic kidney disease	52	17	33
Tubulopathies	50	2	4
CKD of unknown etiology	34	53	156
Nephrolithiasis/ nephrocalcinosis	34	2	6
aHUS	34	1	3
Other glomerulopathies	24	9	38
Hypertensive nephropathy	23	3	13
CAKUT	17	4	23
Diabetic nephropathy	0	0	9
Overall	34	103	300

aHUS, atypical hemolytic uremic syndrome; CAKUT, congenital anomalies of the kidney and urinary tract; TKD, tubulointerstitial kidney disease.

clinically diagnosed with Alport before genetic testing. In total, diagnoses were made in 39 genes, with variants in the following 8 genes reported most frequently: *MUC1*, *PKD1*, *COL4A3*, *COL4A5*, *UMOD*, *COL4A4*, *NPHP1*, and *APOL1* (Supplementary Figure S2). The remaining 31 genes were reported ≤2 times with 28 (72%) genes reported only once. Two participants had dual gene diagnoses because of digenic Alport syndrome (Supplementary Table S5).

Testing Modality

In line with clinical practice in our jurisdiction at the time of this study, most participants (65%, $n = 196/300$) had a phenotypic-driven gene panel as their first line test (Figure 4). In the remaining 104, a phenotypic driven approach was not appropriate because of ambiguity regarding the specific CKD subtype (i.e., CKD of unknown etiology), and therefore proceeded directly to a comprehensive testing strategy. The diagnostic yield after phenotypic driven panel in appropriately selected participants was 36% (70/196); however, employing a phenotypic-driven gene panel alone would have resulted in a lower diagnostic yield for the entire cohort of 23% ($n = 70/300$). By proceeding with a comprehensive testing strategy (i.e., comprehensive gene panel, ES and/or *MUC1* testing, if clinically indicated), we were able to confirm a genetic diagnosis in an additional 33 participants, thereby increasing the diagnostic yield from 23% to 34% ($n = 103/300$). For participants suspected of having ADTKD, targeting *MUC1* testing had a positivity rate of 67% ($n = 14/21$). Considering our initial phenotypic driven approach to testing, participants often required >1 genetic test, with 21% ($n = 21/102$) of the solved cohort undergoing ≥2 genetic tests to achieve a genetic diagnosis. Overall, the median number of tests per participants was 1.2 in the solved cohort, 1.8 in participants in whom a VUS was detected, and 1.5 in participants who, at the time of study close, had negative genetic testing results (Supplementary Figure S3). Our data showed a trend toward increased utilization of comprehensive testing strategies, including comprehensive gene panels and clinical ES over time (Supplementary Figure S4).

Risk Factors Associated With GKD

Families reporting a positive family history of CKD had a higher diagnostic yield (41% vs. 20%, $P < 0.001$). Almost 50% ($n = 147/300$) of this cohort had extrarenal features reported before genetic testing. The diagnostic yield, although numerically higher in the presence of extrarenal features, was not statistically different (38% vs. 31%, $P = 0.18$). Subgroup analysis shows that liver disease (55% vs. 32%, $P = 0.04$) and genitourinary manifestations (100% vs. 0%, $P = 0.05$) were associated with genetic CKD (Supplementary Table S9). Most participants had adult-onset disease (77%, $n = 230/300$). The diagnosis yield was similar in participants with pediatric onset CKD (<18 years) and CKD onset at age 18 to 34 years

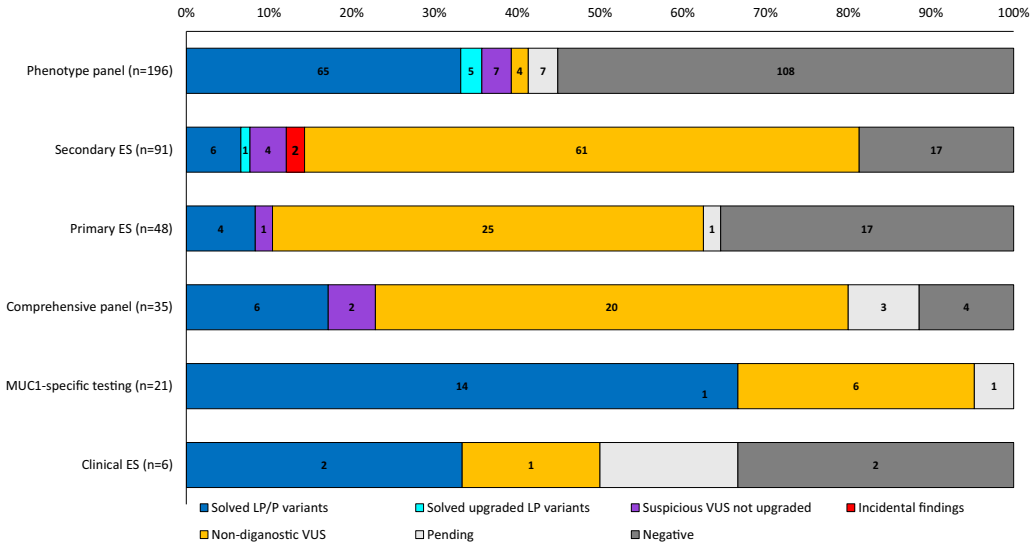

Figure 4. Testing modality and solve rate per modality. Represents the percent of participants solved using each modality. The numbers on the bars represent the number of participants tested. Phenotype-driven panels are clinical genetic tests, including a list of genes of a specific phenotype and performed by a Clinical Laboratory Improvement Amendments–certified laboratory. Primary exome is defined as research exome sequencing as the first-line test, whereas secondary exome is defined as exome sequencing as a second-line test after a negative panel test. Comprehensive panels are clinical tests that analyze for variants in 329 kidney disease genes. Clinical exomes are clinical tests completed by the Genome-wide Sequencing Ontario. *MUC1*-specific testing is testing for the +C *MUC1* variant causing autosomal dominant tubulointerstitial kidney disease. ES, exome sequencing; LP/P, likely pathogenic or pathogenic; VUS, variant of unknown significance.

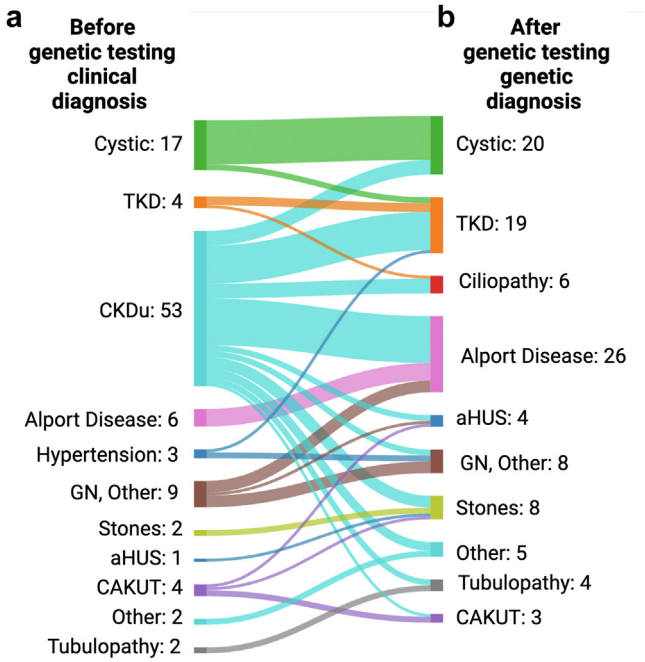

Figure 5. Reclassification rate for all solved participants ($n = 103$). (a) represents the participants' diagnosis before genetic testing and (b) represents the diagnosis after genetic testing (their confirmed genetic diagnosis). Phenotypes include atypical hemolytic uremic syndrome (aHUS); Alport syndrome; congenital anomalies of the kidney and urinary tract (CAKUT); ciliopathies, including nephronophthisis; cystic kidney disease, including polycystic; glomerulopathy (GN), excluding Alport syndrome; hypertension (hypertensive kidney disease); and other kidney diseases (including amyloidosis), stones disease (nephrocalcinosis and nephrolithiasis), tubulointerstitial kidney disease (TKD), and tubulopathies. This figure was created using sankeymatic.com and biorender.com.

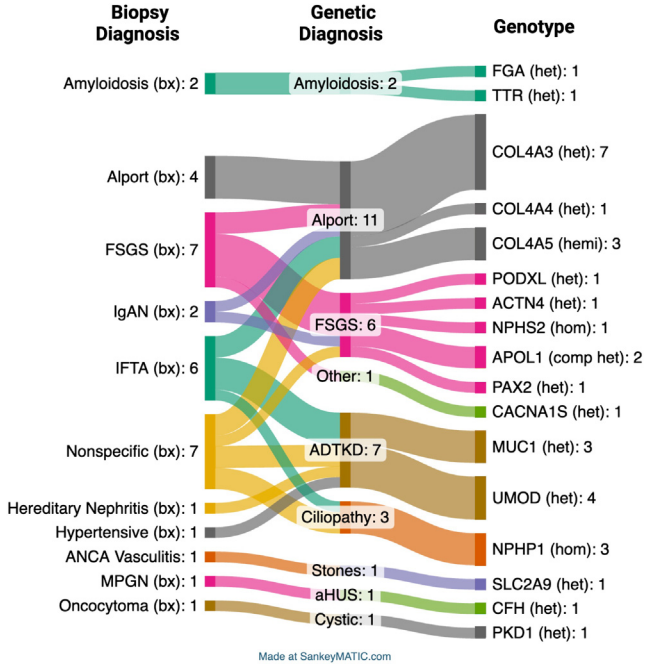

Figure 6. Comparison of biopsy diagnosis to genetic diagnosis. Solved participants with a biopsy available ($n = 34/100$). The left side represents the participants' diagnosis after biopsy, the center represents the diagnosis after genetic testing (genetic diagnosis), and the right side represents the specific genotypes participants were diagnosed with. Zygosity is represented after each gene by; comp het (compound heterozygous), hemi (hemizygous), het (heterozygous), and hom (homozygous). aHUS, atypical hemolytic uremic syndrome; CAKUT, congenital anomalies of the kidney and urinary tract; cystic, cystic kidney disease, FSGS, focal segmental glomerulosclerosis; IFTA, interstitial fibrosis and tubular atrophy; IgAN, IgA nephropathy; MPGN, membranoproliferative glomerulonephritis; TBM, thin basement membrane; TKS, tubulointerstitial kidney disease; TMA, thrombotic microangiopathy. This figure was created using sankeymatic.com and biorender.com.

(40% and 39%, respectively). Diagnostic yield was lower at 27% in participants with CKD onset at age >35 years. Diagnostic yield was higher at 38% in those who had reached ESKD at time of testing; however, there was no difference based on age of ESKD onset <50 or ≥50 years (Supplementary Table S10).

Reclassification

Sixty-seven of the 103 solved participants (65%) were reclassified into a different diagnostic category after genetic testing. The largest reclassification rates were with an *a priori* diagnosis of (CKD of unknown etiology $n = 53$) with reclassification into 10 different phenotypic categories after testing (Figure 5). Three participants with hypertensive nephropathy were reclassified as having genetic glomerulopathies ($n = 2$) and ADTKD ($n = 1$). No participants had a before testing diagnosis of ciliopathies; however, after testing 6% of the solved cohort ($n = 6/103$) were reclassified into this diagnostic category.

Biopsy Diagnosis Compared to Genetic Diagnosis

We compared kidney biopsy findings, when available, in the solved cohort to determine if the histopathology findings on native kidney biopsy were consistent with the molecular diagnosis post–genetic testing. Thirty-three solved participants ($n = 33/103$, 32%) underwent kidney biopsy prior to genetic testing, with a complete biopsy report available in 23 (Supplementary Table S11). Overall, 36% ($n = 12/33$) of participants had a kidney biopsy report consistent with the genetic diagnosis (Figure 6). Eleven participants were diagnosed with Alport syndrome after genetic testing, 4 had a confirmed histopathological diagnosis pre–genetic testing, whereas 7 were reclassified post–genetic testing. The pretesting diagnosis in participants with genetically confirmed Alport syndrome included, IgA nephropathy ($n = 1$), focal segmental glomerulosclerosis ($n = 2$), nonspecific biopsy findings ($n = 2$), and interstitial fibrosis and tubular atrophy

Table 4. Mean age to diagnosis, referral, and genetic diagnosis in solved patients in years, stratified by participant type and testing type

Participant and testing type	Median age of CKD diagnosis, yrs (IQR)	Median age of recruitment, yrs (IQR)	Median age of genetic diagnosis, yrs (IQR)	Median time from CKD diagnosis to recruitment, yrs (IQR)	Median time from recruitment to genetic diagnosis, mo, (IQR)	Median time from clinical diagnosis of CKD to genetic diagnosis, yrs (IQR)
Overall (n = 103)	28.2 (22.4)	39.1 (27.0)	40.9 (27.8)	10.4 (15.5)	2.9 (6.3)	10.7 (15.6)
Participant type						
Index (n = 87)	28.2 (24.0)	39.1 (27.0)	40.9 (27.4)	10.8 (15.7)	2.4 (4.4)	11.0 (15.2)
Affected family member (n = 16)	27.0 (15.4)	42.0 (27.3)	42.1 (27.9)	10.0 (16.4)	5.8 (7.4)	10.6 (16.3)
Testing modality						
Comprehensive gene pane (n = 7)	21.3 (19.6)	44.9 (23.9)	44.7 (23.7)	14.7 (14.5)	1.7 (1.4)	15.1 (14.3)
Phenotypic gene panel (n = 62)	29.9 (30.3)	39.1 (27.9)	40.9 (29.3)	10.4 (14.2)	2.0 (2.4)	10.5 (14.7)
Targeted gene panel (n = 8)	26.3 (22.3)	52.2 (22.3)	53.3 (22.2)	16.3 (25.8)	5.7 (9.9)	17.1 (25.6)
Primary ES (n = 6)	27.3 (12.9)	32.8 (15.7)	33.4 (15.2)	6.1 (8.2)	8.0 (2.1)	6.8 (9.1)
Secondary ES (n = 7)	17.6 (27.1)	34.7 (27.7)	35.7 (27.9)	15.3 (19.3)	6.4 (7.4)	16.2 (19.2)
MUC1 targeted testing (n = 13)	28.2 (8.7)	36.1 (25.7)	37.0 (26.2)	8.5 (13.2)	7.8 (8.9)	8.9 (12.9)
Stratified by time from CKD diagnosis to genetic diagnosis						
Genetics-First <1 yr (n = 7)	26.8 (29.3)	27.2 (28.9)	27.7 (29.0)	0.4 (0.4)	2.0 (2.6)	0.5 (0.7)
Early 1–5 yrs (n = 22)	28.5 (35.0)	29.7 (35.0)	30.5 (34.8)	3.1 (2.9)	2.5 (7.1)	3.3 (2.6)
Late 5–10 yrs (n = 16)	32.0 (35.0)	38.5 (33.0)	38.6 (33.9)	6.3 (2.2)	3.7 (9.2)	6.7 (1.8)
Late 10–20 yrs (n = 31)	24.8 (20.1)	38.9 (18.5)	39.1 (18.4)	13.5 (4.7)	2.3 (6.8)	13.5 (4.6)
Late >20 yrs (n = 27)	20.5 (24.5)	55.4 (27.7)	55.9 (27.5)	24.3 (12.8)	3.2 (7.4)	24.6 (13.5)

CKD, chronic kidney disease; ES, exome sequencing; IQR, interquartile range; *MUC1*, mucin-1 specific gene testing.

(n = 2). For participants who had nondiagnostic kidney biopsies pre–genetic testing, the post testing diagnoses included, genetically confirmed Alport Disease (29%, n = 2/7), focal segmental glomerulosclerosis because of recessively inherited *APOL1* risk alleles (14%, n = 1/7), ADTKD (29%, n = 2/7), and ciliopathies (29%, n = 2/7).

Time to a Genetic Diagnosis

For participants who received a genetic diagnosis (n = 103), the median time from initial CKD diagnosis to genetic assessment was 10.4 years (IQR: 15.5). Affected family members had a shorter wait time than the index participant (10.6; IQR: 16.3 vs. 11.0; IQR: 15.2 years). Reflective of the increase in the availability of genetic testing following establishment of the kidney genetics clinic in 2020, time from initial diagnosis of CKD to genetic diagnosis was shorter in those diagnosed with CKD in the last 5 years, at 3.1 years (IQR: 2.9) compared with participants diagnosed with CKD >5 years from time of genetic assessment (Table 4). Only 7 participants (7%) with confirmed GKD were referred for genetic assessment within 1 year of initial diagnosis of CKD. After genetic assessment, the median time to receive a positive genetic diagnosis was 2.9 months (IQR: 6.3). Comprehensive gene panels provided the shortest return of results (1.7 months, IQR: 1.4), whereas exome analysis had the longest return of results with a median time of 7.8 months (IQR: 8.9) (Table 4).

Clinical Utility

Clinical utility was reported in a high proportion of participants in whom a genetic diagnosis was confirmed (Figure 7). This included the ability to avail of genetic counseling (99%), prognostic clarity (97%), and initiation of education services (90%). In 75% of participants, at risk family members could now avail themselves of cascade testing, leading to a reduction in the diagnostic odyssey for family members in 46%. A change in management was reported in 35%, with direct treatment changes initiated in 33%, following confirmation of a genetic diagnosis. In terms of health resource utilization, 17% were able to avoid additional testing, whereas others required additional subspecialist consultation, imaging, or laboratory testing (62%, 12% and 10%, respectively). In 21%, genetic testing informed risk in a biologically related living donors, whereas 10% of participants had a change in kidney transplantation management (Supplementary Table S12). The proportion of participants and the type of clinical utility measures recorded post establishment of a genetic diagnosis varied across the CKD subtypes

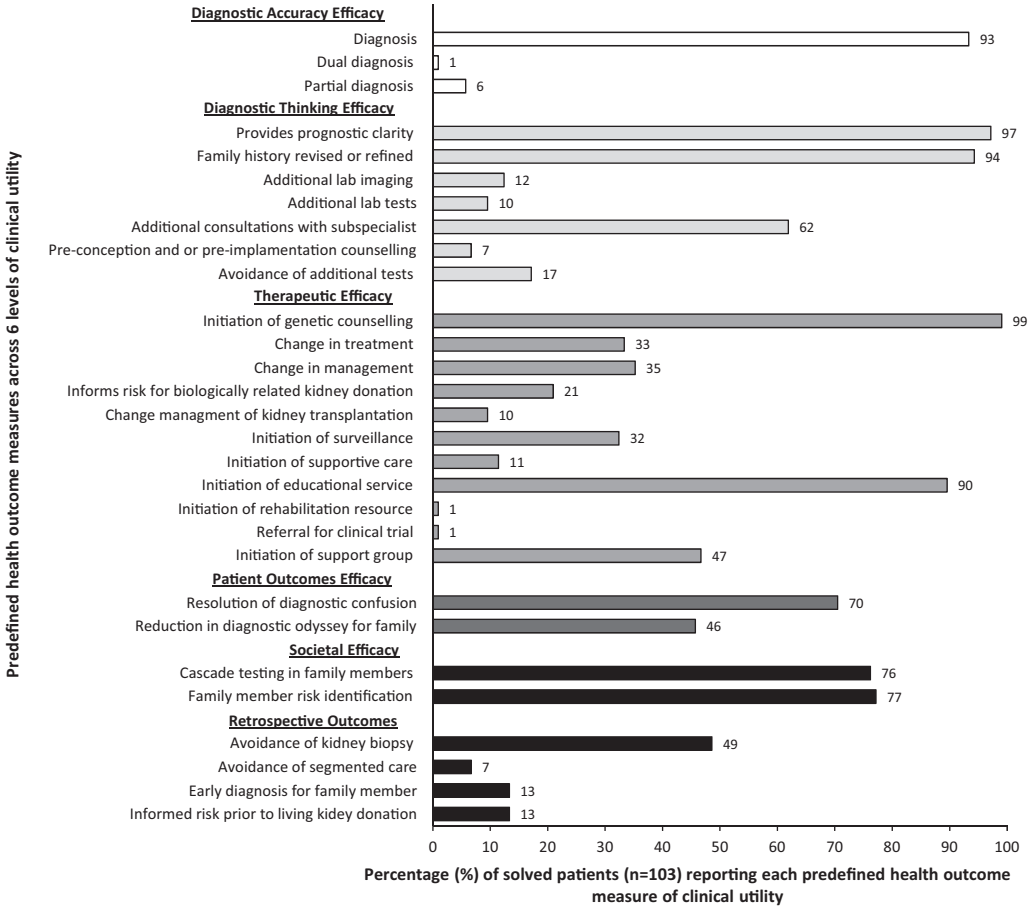

Figure 7. Measured of clinical utility following establishment of a genetic diagnosis post genetic testing. Health outcome measures were recorded across 6 predefined levels of clinical utility using measurements of clinical utility adopted from a toolkit devised by Hayeems *et al.*[44] 2020. The reported health outcomes were measured in 105 patients following genetic testing and establishment of a genetic diagnosis (including incidental findings). All health outcome measures were captured post disclosure of genetic testing results. Legend: Diagnosis, a genetic diagnosis was confirmed post genetic testing by the identification of a pathogenic or likely pathogenic variant in a gene known to cause CKD. Dual diagnosis, more than 1 genetic diagnosis. Partial diagnosis, a genetic testing identified a pathogenic or likely pathogenic variant in a gene known to cause CKD which is exact part of the observed phenotype but not the entire disease spectrum. CKD, chronic kidney disease; LKD, living kidney donation.

(Supplementary Figure S5). Treatment implications were recorded in 35 participants (33%) and included guidance of therapy following establishment of a diagnosis of Alport disease, eculizumab therapy in the peri-transplant period, treatment following establishment of the genetic cause of amyloidosis, treatment following confirmation of hereditary thrombotic thrombocytopenic purpura, commencement of coenzyme Q10 therapy following confirmation a coenzyme Q10 deficiency because of a recessive *COQ2* mutation, and confirmation of the diagnosis of hypophosphatemic rickets because of *PHEX* mutation resulting in commencement of disease specific therapy with burosumab (Supplementary Table S12).

DISCUSSION

By using predefined, clinical referral guidelines and a testing algorithm, 103 out of 300 participants with CKD (34%), referred to a dedicated kidney genetics clinic, received a genetic diagnosis. This yield is higher than previously reported in the general CKD population of 10% to 20%,[1,2] which is reflective of the more selective criteria we employed to determine eligibility for genetic testing. Similar to groups in other jurisdictions,[9,46–48] we demonstrate the importance, in clinical practice, of establishing clearly defined inclusion criteria for genetic testing in CKD. For example, in Italy, Becherucci *et al.*[9] demonstrated high success in identifying patients with GKD (i.e., diagnostic yield of 67%) by instituting 7 categories of selection criteria. Conversely, as previously shown, when testing proceeds without clear clinical guidelines, diagnostic yield can be low at 1.8%.[11] Considering data outlining the probability of a genetic diagnosis based on clinical characteristics, such as the presence of genetic risk factors, or the subtype of CKD,[6] it is vital that we continue to integrate these data into clinical care, to

inform the optimal genetic testing approach in patients with CKD.

At present, one of the cited barriers to accessing genetic testing for patients with CKD is the lack of knowledge among nephrologists on the genetic testing process, with >50% stating they would prefer a multidisciplinary kidney genetics clinic.[5] In Canada, similar to studies in the USA,[46] we highlight the success of the kidney genetics clinic as a model for service delivery for improving diagnosis in patients with suspected GKD. Our testing algorithm resulted in a median time to diagnosis of 2.9 months (IQR: 6.3), following genetic assessment. Time to receiving a diagnosis is an important barrier in the implementation of genetics in nephrology, because a late or delayed diagnosis can limit the maximum clinical utility of the testing result.[49] Overall, we demonstrate that by instituting clinical referral criteria to a kidney genetic clinic, we can successfully identify a high proportion of patients with GKD, with a short turnaround time for genetic testing results.

Although participants in this study received a genetic diagnosis quickly after genetics assessment (median 2.9 months), many spent years in the diagnostic odyssey before receiving a referral to a kidney genetics clinic. Our data show long wait times for genetic assessment from initial diagnosis of CKD (median > 10 years), with few participants being referred for assessment within 1 year of diagnosis of CKD. This long wait time is particularly prevalent in participants diagnosed with CKD >5 years from time of genetic assessment, which is likely reflective of the historic lack of access to genetic testing for patients with CKD.[25] However, considering the establishment of the kidney genetic clinic in 2020, it is noteworthy that participants diagnosed with GKD within this timeframe, still have a median wait time from CKD diagnosis to genetic assessment of 3.1 years. Bern *et al.*[50] demonstrated that among nephrology trainees in the USA, 15% stated they had little or no training in GKD, with 50% stating they did not have enough training to feel competent in the assessment of patients with GKD. The long wait-time for genetic assessment in CKD may therefore be indicative, in part, of underrecognition and underreferral of patients with CKD for genetic testing. This finding therefore warrants further assessment as a potential implementation barrier for kidney genetics.

The testing strategies employed in this study were based on the funding model currently available in our jurisdiction, and in most cases started with phenotypic driven gene panel testing, resulting in a genetic diagnosis in 23% of cases. We demonstrate that by proceeding with a more comprehensive approach to testing, particularly in participants with CKD of unknown cause or in whom gene panel testing was negative, we can further increase diagnostic yield by an additional 11%. Currently, the estimated prevalence of CKD of unknown cause is 10% to 20% within the general CKD population, with up to 20% attributable to a genetic cause.[12] Our data therefore highlights the importance of "reflexing" to comprehensive genomic testing after negative phenotypic driven testing, which can ultimately provide answers for patients, who would otherwise remain without a formal diagnosis.[49] Noteworthy is the finding that >20% of participants (n = 21/102), in whom a genetic diagnosis was established, required ≥2 genetic tests prior to confirmation of GKD. The proportion requiring ≥2 genetic tests is even higher in participants in whom a VUS was detected (n = 88, 66%). This finding raises important questions regarding the cost-effectiveness of different genetic testing approaches. For example, in the Australian context, although associated with higher initial costs ($1600 more costly per patient), comprehensive testing using genomic analysis has been found to be more cost-effective in the long-term through enhanced diagnostic yield, with an incremental cost-effectiveness ratio of AU$5991 per additional diagnosis.[15] Integrating quality of life measures, genomic testing resulted in an incremental cost-effectiveness ratio of up to AU$10,823 per quality-adjusted life years gained.[15] In Italy, data demonstrate that by integrated early exome analysis, as a first tier investigation in the CKD diagnostic pathway of eligible candidates, a cost saving per diagnosis of approximately €1500-€2000 can be achieved.[9] Taken together, these findings suggest that a more comprehensive approach to genetic testing may be the optimal testing strategy in appropriately selected patients, if instituted earlier in the diagnostic pathway for patients with CKD.

Current guidelines have conflicting recommendations on age criteria for genetic testing in nephrology; some prioritize young age of onset, whereas others suggest that there should be no upper age limit.[51,52] These conflicting recommendations are in part related to the historical testing bias, where younger participants—often with clinically more severe phenotypes—were more frequently included in research studies. Similar to previous studies,[7,10,13] we did not find a significant difference in diagnostic yield in pediatric (< 18 years) versus young adult onset (18–34 years), although there was a decrease in diagnostic yield in those with CKD onset at age ≥ 35 years. To address the potential publication bias, studies are increasingly including older adults. For example, Dahl *et al.*[2] recently demonstrated a mean age of genetic

diagnosis of 48 years in adults with CKD, including adults aged 18 to 92 years in their analyses.[4] Herein, we include a large proportion of older adults, with 42% ($n = 127$) aged $\geq$ 50 years at the time of genetic assessment. Interestingly, we found no difference stratifying by age of onset of ESKD <50 years or ≥50 year, which may indicate that the severity of disease rather than age of onset is more indicative of an underlying genetic etiology. These findings therefore suggest that age-based criteria alone in accessing genetic assessment warrants caution; and supports the current Kidney Disease Improving Global Outcomes guidelines that there should be no upper-age limit for monogenic kidney disease.[51]

Another cited implementation barrier for kidney genetics is the perceived lack of clinical utility following the establishment of a genetic diagnosis.[3] In this study, similar to reports by Dahl *et al.*,[2] we record a high level of clinical utility after establishment of a genetic diagnosis. The exact impact of the diagnosis does, however, vary across different subtypes of CKD. For example, treatment change was observed more commonly in participants with glomerular kidney disease; resolution of diagnostic confusion was seen in a higher proportion of participants with ciliopathies; cascade testing in at-risk family members occurred at a higher rate following the establishment of ADTKD. In addition, by using a comprehensive toolkit to assess clinical utility, we provide a more holistic assessment, incorporating not only clinical health outcomes but also patient level and societal outcomes. One limitation is that the assessment of clinical utility occurred at the time of disclosure of genetic results and at the end of study, thereby limiting the ability to assess the impact over time. Future studies are required to longitudinally assess the impact of a genetic diagnosis over time.

A key finding, considering the delayed diagnosis observed in patients with GKD, was the retrospective clinical utility and the potential impact of a genetic diagnosis *if* testing had been performed at an earlier stage in the diagnostic pathway. For example, we found that in 69% of participants with GKD, a native kidney biopsy could have been avoided *if* genetic testing had been performed at an earlier point in the diagnostic pathway. Consistent with findings by other groups,[53] we found that in a high proportion of participants (64%), histopathological findings on biopsy were inconsistent with the ultimate genetic diagnosis. Considering that >20% ($n = 7/33$) of participants underwent an inconclusive kidney biopsy prior to the consideration of genetic testing, our findings suggest that genetic assessment should, in appropriately selected cases, be considered the first-line diagnostic test. This approach could negate the need for nondiagnostic invasive procedures, and ultimately be associated with enhanced economic and patient level outcomes.

The main limitation of this study is that we report data from a single center, thereby reducing generalizability to other centers. In addition, because our inclusion criteria preselect for certain clinical features, for example extrarenal features, there is a reduction in generalizability to a general CKD population and limits our ability to assess for between group variations. This is however, one of the first reports to detail the implementation of a kidney genetics clinics and the direct integration of genetic testing in the diagnostic pathway for CKD in Canada, thereby providing a template for similar models of care. The clinical referral guidelines were developed in 2019, based on the best available data at the time. Although they provide reproducible inclusion criteria, we suggest routine, periodic review, to ensure inclusion of all contemporaneous data in the field of kidney genetics.

CONCLUSION

Overall, we show that implementation of standardized clinical referral guidelines and a testing algorithm in a dedicated kidney genetics clinic, can provide a high diagnostic yield and clinical utility in patients with CKD. With most patients waiting over a decade for genetic assessment, our data suggest that maximum clinical utility will only be achieved with earlier assessment and confirmation of a genetic diagnosis. Overall, our data support genetic testing using a more comprehensive genomic testing approach as a first line test in the diagnostic pathway for patients with CKD meeting eligibility criteria for genetic testing.

DISCLOSURE

All the authors declared no competing interests.

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DATA AVAILABILITY STATEMENT

Additional data can be shared by contacting the senior author upon reasonable request.

AUTHOR CONTRIBUTIONS

This project was conceived by DMC. Data curation was completed by CS, MA, CB, JW, ADM, SC, SR, GAO, CC, LVN, AP, AD, and KK. Formal analysis was completed by CS, JW, AAD, AP, AD, VL, and KK. DMC was responsible for funding acquisition. CS, JW, ADM, AP, AD, and KK completed investigations. DMC, RAH, and AJB were responsible for methodology development. CS, CB, and SR were responsible for project administration. Resources were provided by MC, AC, GF, LG, AAH, SSH, HI, AKJ, AMJ, KL, KM, FR, PSR, APS, MAW, AJB, and RAH. Software was provided by JW and RAH. Supervision and validation were provided by RAH and DMC. Visualizations were created by CS. Writing of the original draft was completed by CS and DMC; review and editing were completed by CS, SR, CC, AC, GF, LG, AKJ, AMJ, LM, PSR, APS, AJB, and DMC. All the authors provided final approval of the version to be submitted.

SUPPLEMENTARY MATERIAL

Supplementary File (PDF)

Supplementary Methods. DNA extraction, Gene panels, *MUC1* testing, exome sequencing, gene list and BED file generation, Variant annotation, Variant analysis, CNV analysis, and Web resources.

Supplementary References.

Figure S1. Pathogenic and likely pathogenic genes in all solved patients ($n = 103$).

Figure S2. Clinical consequences of a positive genetic testing result.

Figure S3. Number of tests per patient subdivided into variant types.

Figure S4. Number of tests per year stratified by testing type.

Figure S5. Clinical utility following the establishment of a genetic diagnoses post genetic testing in patients with chronic kidney disease, stratified by subtype of chronic kidney disease.

Table S1. Genome-wide Sequencing Ontario eligibility criteria for clinical exome sequencing.

Table S2. List of gene panels ordered through Clinical Laboratory Improvement Amendments–approved laboratories.

Table S3. Research based exome sequencing analysis gene list.

Table S4. STROBE Statement: checklist of items that should be included in reports of observational studies.

Table S5. Patients solved for pathogenic and likely pathogenic variants.

Table S6. Patients with initial suspicious variants of uncertain significance upgraded to likely pathogenic following review.

Table S7. Suspicious variants of uncertain significance that cannot be upgraded at this time.

Table S8. Incidental findings not related to primary kidney disease.

Table S9. Prevalence of extrarenal features.

Table S10. Statistical analysis of risk factors associated with genetic kidney disease diagnosis.

Table S11. Native kidney biopsy reports for solved patients ($n = 33$).

Table S12. Health outcomes of genetic testing.

STROBE Statement

This clinical prospective study was performed and written in accordance with the Strengthening the Reporting of Observational studies in Epidemiology (STROBE) guidelines for cohort studies (Supplementary Table S4).

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