Published in final edited form as:

Mol Biol Rep. 2013 October ; 40(10): 5769–5779. doi:10.1007/s11033-013-2680-6.

Susceptibility Gene search for Nephropathy and related Traits in Mexican Americans

Farook Thameem^{1,*}, Issa A. Kawalit^{1,4}, Sharon G. Adler³, and Hanna E. Abboud^{1,2}

¹Division of Nephrology, Department of Medicine, University of Texas Health Science Center, San Antonio, TX 78229, USA

²South Texas Veterans Healthcare System, San Antonio, TX 78229, USA

³Division of Nephrology and Hypertension, Los Angeles Biomedical Research Institute, Torrance, CA 90502, USA

⁴Division of Nephrology; Department of Medicine, University of Jordan, Amman (11942) Jordan

Abstract

The rising global epidemic of diabetic nephropathy (DN) will likely lead to increase in the prevalence of cardiovascular morbidity and mortality posing a serious burden for public health care. Despite greater understanding of the etiology of diabetes and the development of novel treatment strategies to control blood glucose levels, the prevalence and incidence rate of DN is increasing especially in minority populations including Mexican Americans. Mexican Americans with type 2 diabetes (T2DM) are three times more likely to develop microalbuminuria, and four times more likely to develop clinical proteinuria compared to non-Hispanic whites. Furthermore, Mexican Americans have a six fold increased risk of developing renal failure secondary to T2DM compared to Caucasians. Prevention and better treatment of DN should be a high priority for both health-care organizations and society at large. Pathogenesis of DN is multi-factorial. Familial clustering of DN-related traits in MAs show that DN and related traits are heritable and that genes play a susceptibility role. While, there has been some progress in identifying genes which when mutated influence an individual's risk, major gene(s) responsible for DN are yet to be identified. Knowledge of the genetic causes of DN is essential for elucidation of its mechanisms, and for adequate classification, prognosis, and treatment. Self-identification and collaboration among researchers with suitable genomic and clinical data for meta-analyses in Mexican Americans is critical for progress in replicating/identifying DN risk genes in this population. This paper reviews the approaches and recent efforts made to identify genetic variants contributing to risk for DN and related phenotypes in the Mexican American population.

Keywords

Diabetic Nephropathy; Mexican Americans; ACR; GFR; Type 2 Diabetes; linkage; candidate gene; SNPs

*Corresponding author: Farook Thameem Ph.D., Assistant Professor of Medicine, Division of Nephrology; Department of Medicine, The University of Texas Health Science Center at San Antonio, 7703 Floyd Curl Drive, San Antonio, Texas 78229; USA, Phone: +1-210-567-4787, FAX: +1-210-567-4712, thameem@uthscsa.edu.

Introduction

Diabetic nephropathy (DN) is a progressive microvascular complication of diabetes in which patients exhibit persistent proteinuria, hypertension, renal failure, and increased morbidity and premature mortality largely as a result of cardiovascular disease. Diabetes has now become the most common cause of end-stage renal disease (ESRD) necessitating dialysis or renal transplantation. Diabetes accounts for ~55% of patients treated for ESRD each year, including dialysis and renal transplantation and is anticipated to reach 70% by 2015 [1]. Given the overwhelming burden of DN worldwide, it is of particular importance to understand the pathogenesis of DN and to identify new therapeutic approaches which alone or in combination with standard therapy have the potential to prevent or delay its progression thereby improving kidney and patient survival. Despite more aggressive treatment of diabetes, the incidence and prevalence rates of kidney disease continue to increase in a subset of the population suggesting that a subgroup of patients remain at high risk for kidney disease in diabetes. Furthermore, marked racial variation has been observed in the prevalence and familial aggregation of DN with high rates in minority populations including Mexican Americans [2].

Mexican Americans (MA) represents the largest and fastest growing minority population in the US and constitutes approximately 66% of US Hispanics [3]. The MA population has a greater prevalence of type 2 diabetes (T2DM) and associated complications compared with non-Hispanic whites [4]. MA with T2DM lose renal function at a faster rate than African Americans and Non-Hispanic whites [5]. MA has 6.1 times higher incidence of treatment for ESRD than non-Hispanic whites and T2DM accounts for 93% of ESRD cases in MA [6]. The incidence rate of ESRD is rising in the Hispanic population and Texas has the highest number of MA patients with ESRD. The higher incidence of ESRD in MA combined with an increased incidence over time and longer survival after ESRD onset, indicate that this ethnic group will comprise a large portion of diabetic ESRD in the future and predict an increasing burden to healthcare, if prevention measures are not instituted in this population [7].

Although the higher incidence of DN in MA may be due to differences in access to healthcare, and prevalence of modifiable lifestyle risk factors, ethnic variation in inherited predisposition to DN is also considered as an important factor. Understanding and identifying susceptibility genes underlying the pathogenesis of DN and related traits may help explain the ethnic differences in triggering the defective allele with differential disease risk and guide preventive and therapeutic efforts in MA. The aim of this review is to provide an overview of the recent efforts and findings on the genetic determinants of DN and related phenotypes such as changes in albumin-to-creatinine ratio (ACR) and estimated glomerular filtration rate (GFR) in the MA population.

Over the last decade, both the dichotomous (DN) and quantitative (ACR, GFR) linkage and the genetic association approaches have been successfully applied to detect genes underlying the development of DN in MA as part of several large epidemiological studies including the Family Investigation of Nephropathy and Diabetes (FIND), San Antonio Family Diabetes/Gallbladder Disease (SAFDGS), San Antonio Family Heart Study (SAFHS), and the

National Health and Nutrition Examination Survey III (NHANES III). Of many strategies available to decipher the genetic architecture of DN, primarily genome-wide linkage, candidate gene, and mapping by admixture linkage disequilibrium (MALD) analysis have been adopted in these MA cohorts.

Heritability of DN phenotypes in MA

Heritability is a measure of a trait's variability due to genetic factors and is estimated using the phenotypic correlations between relatives in families. Heritability estimates of ACR and GFR in MA ranged from 0.24 to 0.27 and 0.21 to 0.52, respectively [8–11], and are similar in magnitude to heritability estimates reported in other populations [12–14]. These data indicate that ACR and GFR are heritable in MA, and it is reasonable to screen for QTL influencing these traits.

Genome-wide linkage scan for DN in MA

Genome-wide linkage scan is employed to identify genetic regions co-segregating with DN and related traits in MA families utilizing hundreds of evenly spaced genetic markers such as microsatellites/SNPs. A major advantage of this approach is that it provides the potential benefit of explaining a larger proportion of the heritability of disease or traits of interest.

FIND is a multicenter study designed to identify and characterize susceptibility genes influencing both dichotomous (DN) and quantitative (ACR and GFR) traits in four ethnic groups that includes European Americans, African Americans, Native Indians and MA. Patient recruitment and data collection procedures of FIND are published elsewhere [15]. In brief, FIND is predominantly a sibling pair study. Families of probands with DN having a diabetic sibling with or without nephropathy were recruited.

Using 404 microsatellite markers genotype information on 397 full sibling pairs including 225 MA sib pairs, suggestive evidence for linkage of DN was observed on chromosomes 7q21.3, 10p15.3, 14q23.1, and 18q22.3 after accounting for sex effects [16]. The linkage findings are in concordance with the previous reports for the presence of DN susceptibility genes on chromosomes 7q, 10p, and 18q [17–19]. However, linkage scan only in MA participants failed to identify a genetic region significantly linked to DN. Subsequently, Igo et al., [20] performed a genome-wide linkage scan for DN susceptibility loci in the FIND study using the genotypic data of 5,500 SNPs (Illumina panel IV) on 1623 subjects from 1,235 nuclear and extended pedigrees that included 632 subjects from 478 pedigrees of MA participants. DN trait was adjusted for sex. While the genome scan identified a suggestive linkage of DN to a genetic region on 7p (LOD=1.81) across all four populations, chromosome 11p15 was found to be linked (LOD=2.2) to DN only in MA participants. A fine mapping of the 11p15 region should help identify the causal variants for DN susceptibility in MA participants of the FIND study.

Genome-wide linkage scan for albuminuria in MA

Increased ACR represents an intermediate and quantitative phenotype that predicts the progression of DN. Whole-genome screens have been conducted to localize genes

influencing urinary ACR in MA cohorts of the FIND, SAFDGS and SAFHS. In the FIND study, linkage of ACR was performed as a quantitative trait using 404 microsatellite markers information available on the 883 sibling pairs including 520 MA sib pairs. After accounting for the covariate effects of sex and age at T2DM diagnosis, the Haseman-Elston linkage test identified the strongest linkage signals for ACR occurring on chromosomes 2q14.1, 7q21.1, and 15q26.3 [16]. However, linkage analysis failed to identify a chromosomal region significantly linked to ACR only in MA participants examined. Subsequently, using the genotypic data of 5,500 SNPs and the ACR data of 1316 MA participants of the FIND study, Igo et al., [20] identified a suggestive evidence for linkage of ACR with a LOD score of 2.29 ($P = 0.00058$) occurring on chromosome 22q12 near the nonmuscle myosin heavy chain 9 (*MYH9*) / apolipoprotein L1 (*APOL1*) gene region. Age and sex were used as covariates. The 22q chromosomal region was reported to be linked with ACR in European Caucasian who participated in the Joslin type 2 diabetes study [13]. In addition, several epidemiological studies have reported a significant association between the genetic variants of *MYH9/APOL1* and ESRD and focal segmental glomerulosclerosis in African Americans [21–25]. Fine mapping of the 22q12 region should help to identify causal variants influencing ACR in MA participants of the FIND study.

Genetic variants influencing ACR are being investigated in MA participants of the SAFDGS cohort. The MA family members recruited for the SAFDGS and the data collection procedures used were reported previously [26]. In brief, probands of SAFDGS were low income MA with T2DM, and all 1st, 2nd, and 3rd degree relatives of probands were invited to participate in the study. Utilizing the genotypic data of 379 microsatellite markers and ACR data available on 335 MA subjects from 27 SAFDGS families, a quantitative ACR linkage scan was performed by variance component analytical tools implemented in the program SOLAR. This genome scan identified the strongest evidence for linkage of ACR occurring on human chromosome 15q12 (LOD=3.1) at the *GABRB3* marker region after accounting for age and sex as covariates [21]. The LOD score of 3.1 dropped down slightly, when hypertension (LOD=3.0) and diabetes (LOD=2.5) were included as additional covariates. Of the positional candidate genes located within the linkage interval on 15q12, the tight junction protein 1 (*TJPI*) and gremlin 1 (*GREM1*), were further investigated for DNA sequence variants that may contribute to the linkage of ACR. However, the DNA sequence variants examined from *TJPI* and *GREM1* genes failed to exhibit significant association with ACR indicating that the genetic variants examined in these two loci do not play a major role in regulating ACR levels in MA recruited for the SAFDGS [27, 28].

A susceptibility gene search for ACR has also been conducted in another independent cohort of MA who participated in the SAFHS cohort. SAFHS family member recruitment and a variety of metabolic, hemodynamic, anthropometric, and demographic data collection procedures from more than 40 extended families were described previously [29]. Probands and family members were randomly recruited for the SAFHS from a census tract in San Antonio of low-income MA regardless of preexisting medical conditions. SAFHS was established to identify and characterize genes influencing cardiovascular disease-risk factors. We performed a linkage scan to localize the genetic region harboring susceptibility genes for ACR utilizing 404 microsatellite marker information and the urinary ACR data

available (N=486; 26 families) in a subset of MA participants of the SAFHS. After accounting for the covariate effects of age, sex, BMI, triglycerides and systolic blood pressure (SBP), suggestive evidence for linkage of ACR was found to localize on chromosome 20q12 (LOD score of 3.1) near marker *D20S481*. The LOD score increased to 3.5 after including hypertension status as covariate for SBP, but it decreased to 2.8 when diabetes status was replaced for SBP [30]. We subsequently repeated the linkage scan in a larger sample size of 848 participants including the 486 subjects who participated in the original ACR linkage scan [30]. Interestingly, linkage of ACR in a larger sample size was also observed localizing in the same marker region on 20q12 with a LOD score of 2.94 after accounting for the covariate effects of age, sex, BMI, blood pressure medication and diabetic duration [10]. Shike et al [31] mapped a QTL for albuminuria in a diabetic mouse model to a chromosomal region that is syntenic with the region on human chromosome 20q12. In addition, genome scans have linked the 20q12 region with T2DM [32, 33]. Potential positional candidate genes located on 20q12 include hepatocyte nuclear factor 4alpha (*HNF4A*), a transcription factor and a regulator of wide range of metabolic processes. Dysfunction of *HNF4A* is associated with maturity-onset diabetes of young I and DN [34, 35]. Taken together, fine mapping of 20q12 region is clearly needed to validate and narrow down the candidate interval to enable identification of causative genes influencing ACR levels.

Genome-wide linkage scan for GFR in MA

Equations that estimate GFR facilitate the diagnosis, evaluation and management of chronic kidney disease (CKD). GFR varies between individuals and is influenced by genetic and environmental factors (T2DM) and their interactions. Because eGFR is a continuous trait, its use as an intermediate endo-phenotype in genetic studies has the advantage to offer more precision and more information about CKD severity than the threshold-defined CKD stages. Both the quantitative linkage and the genetic association approaches in MA cohorts (FIND/SAFDGS/SAFHS) have been applied to identify QTL influencing GFR variability.

Schelling et al [11] used the GFR estimates derived from the Modification of Diet in Renal Disease (MDRD) equation and the genotypic data of 404 microsatellite markers on 507 MA subjects in the QTL influencing eGFR levels in the FIND study. After accounting for the effects of diabetes duration and angiotensin converting enzyme inhibitor/angiotensin receptor blocker use, suggestive evidence for linkage of eGFR localized on chromosomes 1q43 (LOD=3.8), 2p13 (LOD=3.0), 7q36.1 (LOD=4.2), 8q21.3 (LOD=4.0) and 18q23 (LOD=2.1) [11]. The 18q23 region was previously reported to be linked with DN in the FIND study [9]. Several studies have also reported an association between DN and the genetic variants of the carnosine dipeptidase 1 (*CNDP1*) gene located on 18q23 [36]. Refining the 18q23 linkage region would help identifying causal variants for eGFR/DN in the FIND participants.

In an attempt to localize genes influencing changes in GFR in MA participants of the SAFDGS, we estimated renal function using both the Cockcroft-Gault (creatinine clearance) and the MDRD (eGFR) formulae [9]. While, both estimates of renal function exhibited significant heritability, only the creatinine clearance showed significant genotype and

diabetes (G×T2DM) interaction effects in our population [9]. Subsequently, we performed a multipoint 382 microsatellite marker linkage scan on both measures of renal function (creatinine-clearance and eGFR) available on 453 subjects using models that did not include $G \times T2DM$ interaction effects (Model 1) and that included $G \times T2DM$ interaction effects (Model 2) after adjustment for the trait-specific covariates effect (Creatinine clearance = diabetes, diabetes duration, SBP, and antihypertensive medication; eGFR = diabetes, diabetes duration, SBP, antihypertensive medication, and BMI). Linkage of creatinine clearance identified a region on chromosome 2q35-37 near the markers D2S1363-D2S427 for both models. However, the significant LOD score of 3.3 was obtained from model 2 which accounted for the interaction influences [9]. In addition, suggestive evidence for linkage of both creatinine clearance (LOD = 2.9) and eGFR (LOD 2.6) was found to occur only for Model 1 (without $G \times T2DM$ interaction) on chromosome 9q21 between markers D9S922 and D9S1120. Our findings suggest that the genes located on 2q and 9q could differentially influence renal function in diabetic and nondiabetic environments in MA participants of the SAFDGS [9].

Of the positional candidate genes located within the 2q35-37 region, insulin receptor substrate 1 (*IRS1*) was screened for genetic variants. Of 19 variants that we identified and genotyped within the *IRS1* gene, Bayesian quantitative trait nucleotide analysis indicated the presence of strong association between the Gly(972)Arg variant of *IRS1* and creatinine clearance. The Gly(972)Arg variant contributed to 26% of the linkage signal on 2q. Expression of the *IRS1* mutant Arg972 in human mesangial cells significantly reduced insulin-stimulated phosphorylation of *IRS1* and Akt kinase. Carriers of Arg972 had significantly decreased renal function values [37]. A potential mechanism by which the Arg972 variant of *IRS1* contribute to decline in renal function is by impaired insulin receptor (IR) / *IRS1* signaling in renal mesangial cells (MC) and afferent arteriolar microvascular smooth muscle cells (VSMC), major regulators of GFR [37]. Alteration or loss of contractile activity due to impaired insulin signaling in MC/VSMC by Arg972 may lead to alteration in glomerular hemodynamics and glomerular injury thereby favoring the decreased renal function. Together, we speculate that the Gly(972)Arg variant is one of the functional variants influencing renal function in the *IRS1* or is in linkage disequilibrium with major functional variant(s) flanking *IRS1* [37]. Fine mapping of 2q35-37 and 9q21 regions would eventually identify causal variants influencing renal function in MA participants of the SAFDGS.

GFR susceptibility loci have also been investigated in the SAFHS cohort. Utilizing eGFR and the genotypic data (417 microsatellite markers) available on 848 subjects from the SAFHS, genome scan identified the strongest evidence for linkage of eGFR to chromosome 9q21 near the marker *D9S922* with a LOD score of 3.9 [26]. It is interesting to note that linkage of eGFR in SAFDGS cohort was also localized to the same 9q21 region [9]. Most importantly, a recent GWAS in Europeans identified a GFR susceptibility locus on 9q21 [38]. Taken together, the available data from both genome-wide linkage and association studies from two ethnically diverse populations provide evidence for the existence of eGFR susceptibility loci on chromosome 9q21. Potential positional candidate genes located within the 9q21 GFR linkage interval include the transient receptor potential (*TRP*)*M3*, *TRPM6* and

the solute carrier (*SLC*)28A3 genes. Fine mapping of 9q21 region in both SAFDGS and SAFHS cohorts are essential to identify causal variants influencing variation in eGFR.

Biological candidate gene analysis for DN phenotypes in MA

Examining genetic variation within the known biological candidate genes and their association with disease risk is a potential alternate approach to elucidate the genetic basis of DN phenotypes. Difference in allele frequencies between case and control subjects or mean trait values by genotype is a strength of this approach. A number of biological candidate genes have been investigated to explain the genetic susceptibility to DN and related phenotypes in MA cohorts.

Genetic variants in engulfment and cell motility 1 (*ELMO1*) and *CNDP1* were reported to be associated with DN in European Americans, African Americans, Asians, and American Indians [36, 39–41]. Carnosinase degrades carnosine (β -alanyl-L-histidine), which has been ascribed a renal protective effect as a scavenger of reactive oxygen species. ELMO-1 is known to be involved in fibrogenesis, inducing the profibrotic TGF β and matrix synthesis. We therefore examined whether the genetic variants reported to be associated with DN in the *CNDP1* and *ELMO1* loci are also associated with DN in MA who participated in the mapping by admixture linkage disequilibrium (MALD) study, an integral case-control study design of the FIND (FIND-MALD). The tagging SNPs selected from 8 candidate genes including *CNDP1* and *ELMO1* were genotyped in 455 cases (T2DM patients with nephropathy) and 437 controls (T2DM patients without nephropathy). Although our association analysis failed to replicate an association between genetic variants of *CNDP1* and *ELMO1* and DN, a novel association between a SNP pair involving rs2146098 and rs6659783 from the hemicentin 1 (*HMCN1*) gene and DN was identified in MA participants of the FIND-MALD study [42].

Genes involved in the regulation of blood pressure, endothelial function, anti-oxidants and lipid metabolism were investigated as potential susceptibility candidate genes of DN-related traits in MA participants of the SAFDGS and SAFHS. Genes encoding components of the renin–angiotensin–aldosterone system (RAAS) have received special attention, due to the central role of this system in regulating blood pressure, sodium balance and renal hemodynamics. Candidate genetic variants of RAAS components such as the insertion/deletion polymorphisms of angiotensin converting enzyme I (*ACE-I/D*), the M235T variant of angiotensinogen (*AGT-M235T*), and the A1166C polymorphism of angiotensin II type 1 receptor (*AT1R-A1166C*) genes have been extensively associated with cardiovascular-renal disease risk in many studies [43]. When these three candidate polymorphisms of RAAS components were examined for their association with ACR and GFR in SAFDGS and SAFHS cohorts, only the *AGT-M235T* polymorphism exhibited a nominal association with GFR values in SAFHS after accounting for the effects of diabetes, duration of diabetes, blood pressure measures and antihypertensive medications [44, 45]. The results suggest that the RAAS candidate polymorphisms examined do not have a major effect on the DN-related phenotypes in the SAFDGS and SAFHS participants.

Endothelial dysfunction is strongly associated with the development and progression of DN. Of the genes involved in endothelial function, the *eNOS* gene encoding endothelial nitric oxide synthase gained attention as it regulates nitric oxide production in the endothelium. Candidate genetic variants of *eNOS* such as the T-786C (rs2070744), Glu298Asp (rs1799983), and 27 bp variable number of tandem repeats (27 bp-VNTR-a/b) polymorphisms of *eNOS* are thought to alter nitric oxide production and contribute to the development of vascular and renal disease risk [46]. We examined the effect of these three polymorphisms on individual's susceptibility to variation in ACR and GFR in SAFDGS and SAFHS participants. While the association analysis failed to find an association between the examined *eNOS* polymorphisms and ACR and GFR in SAFDGS, the 27 bp-VNTR-a/b variant exhibited a modest association with ACR after accounting for the trait specific covariate effects in SAFHS subjects [47, 48]. Overall, the data suggest that the genetic variants examined at the *eNOS* locus do not influence variation in ACR/GFR in SAFDGS and SAFHS participants.

Anti-oxidative and anti-atherogenic activities of the paraoxonase (PON) family genes have been documented. Alteration of paraoxonase 2 (PON2) enzyme activity due to genetic variations in the *PON2* gene [Arg148Gly (rs11545942); Cys311Ser (rs7493); an intronic variant A/C (rs12794795)] is thought to influence the development of oxidative stress thereby contributing to the pathophysiology of microvascular complications of diabetes [49]. Investigation of these three candidate polymorphisms at the *PON2* gene in both SAFDGS and SAFHS cohorts indicated only a nominal association between Cys311Ser and ACR levels in SAFDGS suggesting that the variants examined at the *PON2* locus do not play a major role in variation in ACR and GFR levels in these cohorts [50, 51]

Genetic variants influencing changes in ACR and eGFR levels have also been investigated in MA participants as part of a large epidemiological study the Third National Health and Nutrition Examination Survey (NHANES III), a population-based and nationally representative survey of the US population. Ned et al [52] recently examined whether selected 60 candidate polymorphisms within 27 inflammatory response genes are associated with ACR in 1570 MA participants from the NHANES III. While, a variant (rs1143623) residing within the interleukin 1 β (*IL1 β*) gene exhibited a significant association with the dichotomous trait albuminuria (odds ratio of 1.46; CI: 1.28–1.67), variants residing within *IL1 β* (rs1143623), C-reactive protein (*CRP*; rs1800947) and *NOS3* (rs2070744) were also found to be nominally associated with quantitative ACR $\geq$ 30 mg/g in NHANES III study [52].

Apolipoprotein E (ApoE) is known to play a central role in lipid metabolism. The lipid profiles or atherogenic factors are, in part, determined by three common isoforms (E2, E3, E4) encoded by three alleles (ϵ 2, ϵ 3, ϵ 4) in exon 4 of the *APOE* gene. The ϵ 2 allele of *ApoE* has been associated with an increased risk of DN [53]. Chu et al [54] investigated whether the ϵ 2/ ϵ 4 alleles of *APOE* are associated with measures of renal function in 1,656 MA participants from the NHANES III. Association analysis failed to find a statistically significant association between *APOE* variants and renal function estimated either by MDRD or CKD-EPI (Chronic Kidney Disease Epidemiology Collaboration) equations.

Admixture mapping for DN susceptibility loci in MA

Because the prevalence and incidence of disease risk differs among ethnic groups, admixture mapping is considered as a powerful tool to localize genetic regions containing ancestry-linked traits when the distribution of the susceptibility genes is different among the founding populations. It uses a set of ancestry-informative markers (AIMs) with large allele frequency differences in the two parental populations to effectively identify genetic variant(s) that distinguish the two parental groups and co-segregate with the disease [55]. Admixture mapping has been successful in identifying genetic risk loci influencing non diabetic-ESRD [21].

Because MA are an admixed population, a genome wide SNP panel of 8,144 AIMs was recently developed to distinguish the chromosomal segments of MA and Amerindian and European ancestries [56]. Subsequently, a genome-wide admixture mapping was conducted to localize DN susceptibility genes using a subset of 4300 AIMs markers generated from the 1154 MA participants of the FIND-MALD study comprised of 664 cases (T2DM with nephropathy) and 490 controls (T2DM without nephropathy). After accounting for the effects of covariates age, sex, diabetes duration, and recruitment site, ADMIXMAP identified significant evidence for linkage of DN occurring on 2q36.3-37.1 with a Z score of 4.3 ($P = 1.45 \times 10^{-5}$) [55, 56]. Suggestive evidence for linkage of DN was also observed on 1p35, and 14q32 in FIND-MALD. Potential positional candidate genes located on 2q36-37 include Delta-and-Notch-like EGF-related receptor (*DNER*) and *IRS1*. The chromosomal region 2q36 harboring *IRS1* was previously reported to be linked with eGFR in MA participants of the SAFDGS [9, 37]. Fine mapping of 2q36 region followed by a demonstration of biological relevance are required to ascertain the significance of the findings in MA participants of the FIND-MALD.

Concluding remarks

Table 1 summarizes the localization of genetic regions that may harbor susceptibility genes for DN (2q36, 11p15; FIND), ACR [(15q12; SAFDGS), (20q12; SAFHS), (22q12; FIND)], and renal function [(1q43, 2p13, 7q36.1, 8q13.3, 18q23; FIND), (2q36, 9q21; SAFDGS), 9q21; SAFHS)] in the MA cohorts examined. As expected for a complex trait, multiple linkage peaks have been observed for DN and related traits in MA participants. Disappointingly, there is very little overlap in the detected loci between different studies and related traits. This may be due to heterogeneous study populations, differences in methods and statistical modeling strategies, pedigree structures recruited, ascertainment criteria, definitions of DN and measurement of kidney function, diabetes duration, and covariates used for analysis (Table 1).

While the functional relevance of most of the linkage findings needs to be established and replicated, genetic regions suggestively linked with DN, ACR and GFR within MA cohorts suggest that multiple loci may be linked with DN and its contributing phenotypes. Genes that regulate dichotomous trait (DN) may differ from those controlling quantitative traits (ACR and eGFR). In addition, genes regulating eGFR may differ from those controlling ACR. However, suggestive evidence for a shared genetic association was also detected for

DN phenotypes in the MA cohorts. For example, variation in eGFR was found to be linked to chromosome 9q21 in two independent MA cohorts; SAFDGS [9] and SAFHS [10]. The chromosomal region 2q36 linked to DN in the FIND-MALD study [57, 58] was also found to be linked with eGFR in SAFDGS [9]. Furthermore, QTL influencing eGFR, serum creatinine, and creatinine clearance were linked to chromosome 9q21 in SAFHS cohort suggestive of localization of a common gene(s) on 9q21 that may regulate albuminuria and renal function-related traits [10]. Linkage of eGFR on 18q22 in MA participants of the FIND study is consistent with linkage peaks in genome scans for DN in European American [18] and African American [19] populations, as well as the FIND linkage scan for a composite DN phenotype [16]. Although, identification of genes independently regulating DN phenotypes as well as a possible gene co-regulating these traits located within linkage peaks that replicate between multiple studies are needed, linkage results emanating from these reports on DN and related phenotypes represent a first step toward improving our knowledge of the mechanisms underlying genetic susceptibility to DN phenotypes in MA.

Genetic variants examined at the biological candidate genes for their association with DN phenotypes have yielded conflicting results within MA or in other ethnic groups. Absence of major effect observed between DN phenotypes and the candidate variants residing within *ELMO1*, *CNDP1*, *APOE*, *RAS*, *eNOS*, and *PON2* may be related to ethnic differences in their frequencies, genetic heterogeneity, coupled with study design flaws such as population stratification, sample sizes lacking statistical power, use of confounding variables, and evaluation of inadequate numbers of polymorphisms in genes to determine their true involvement. However, genetic variants investigated and their frequencies observed in MA cohorts may contribute to future meta-analyses validating these polymorphisms. Furthermore, ethnic differences in gene frequencies may assist in the search to identify causal genes within and across populations.

Future direction

To accelerate the gene discovery process, high throughput scanning of MA families followed by replication in other population should eventually permit identification of causal variants influencing DN phenotypes within the loci identified. Genome-wide association studies (GWAS) have recently emerged as a novel technology to identify common risk variants (> 5% MAF) for complex traits and diseases. However, the functional relevance of these common variants identified thus far is largely unknown. Such observations have led to concepts of ‘missing heritability’ and ‘synthetic associations’, highlighting the role of rare causal variants (< 5% MAF) that are undetectable using the current GWAS [59]. Next generation exome sequencing was subsequently introduced as a complementary approach to GWAS with the possibility of identifying rare coding variants with large effects. Such exome sequencing efforts has helped creation of large database of low frequency variants allowing researchers to evaluate these variants as candidate genetic determinants of DN. Advances in exome sequencing have now set the stage for applying whole-genome sequencing (WGS) to identify rare causal variants to facilitate clinical diagnosis and personalized disease-risk profiling of individuals. WGS in a large pedigree is the current frontier of hereditary disease. Large pedigrees with multiple affected subjects or subjects with extreme values are enriched with multiple copies of rare variants providing statistically

significant evidence for both co-segregation and the effect of those variants [60]. Furthermore, the use of large pedigrees with multiple affected subjects is homogeneous and better than using a combination of multiple smaller pedigrees.

Taken together, WGS in large MA pedigrees with multiple affected subjects is very crucial at this stage to search for rare variants that co-segregate and have a substantially higher likelihood of representing risk allele for DN phenotypes. The SAFHS and SAFDGS cohorts are comprised of multigenerational pedigrees of MA descent. SAFHS participants represent 45 pedigrees with up to 171 individuals in a pedigree, across 5 generations [61]. Similarly, SAFDGS represent 39 pedigrees with up to 66 individuals in a pedigree across five generations [62]. A variety of metabolic, hemodynamic, anthropometric, and demographic variables are available from both cross-sectional and longitudinal data collected for these extended large pedigrees of MA. While, few patients in these cohorts have DN, WGS in these pedigrees would help to identify gene(s) influencing changes in GFR and ACR. Recruitment of new large multinuclear/extended families with multiple DN subjects is costly. Since the FIND cohort is comprised of sibpairs with and without DN [15], extending this cohort to include all relatives may be cost effective and would represent an excellent utilization of resources for the future WGS in MA.

Consideration must be given to other sources of variation including gene–gene, and gene–environment interactions as well as copy number variation, in addition to improving statistical methods addressing lack of power, phenotypic and population heterogeneity and multiple testing. Nonetheless, phenotypic data should be subjected to the same level of quality control as genotypic data, because poorly defined phenotypes lead to loss of power and inability to replicate findings. A new estimate of GFR was recently validated by CKD-EPI formula in the MA participants of the NHANES study [63]. Inker et al [64] have recently demonstrated that GFR estimated using a combined creatinine–cystatin C (GFR-Cre-CysC) equation performed better than equations based on either of these markers alone. Therefore, eGFR may be recalculated using CKD-EPI / GFR-Cre-CysC formulae in MA cohorts to validate the findings. In all cohorts, ACR, and eGFR were determined based on a single random sample collection, although validation with serial measures would be ideal.

Finally, emerging evidence from animal models and tissue culture experiments suggests that the complex interplay of epigenetic factors interacting with genes and environment play a critical role in susceptibility to DN [65]. Future studies are required to elucidate the role of epigenetic modification in human DN. Methods should be developed to identify more efficient ways to combine data from sources such as GWAS/WGS, epigenetics, and gene expression to successfully identify and characterize DN susceptibility genes.

In conclusion, identification of novel diagnostic and better prognostic markers for the prevention and better treatment of DN in the MA population and in other ethnic groups should be a high priority for both health-care organizations and society at large. Genetic approaches and statistical methods together with the examined DN phenotypes have contributed considerably to our understanding of the genetic basis of DN in MA. The results of linkage and association analyses obtained within MA cohorts will be valuable complements to the analyses of future WGS data that together should accelerate

identification of causative genes for DN phenotypes. Better understanding and awareness of the disparities of DN risk genes by race and ethnicity may further help clinicians and public health professionals to develop culturally sensitive interventions, prevention programs, and services specifically targeted toward risk burdens in MA population. With the existing knowledge and the development of robust and novel high throughput technologies, we envision promising innovative therapeutic potential in a near future for this devastating complication of diabetes.

Acknowledgements

This study was supported by Grant-in-Aid from the American Heart Association, Carl W. Gottschalk Research Scholar award from the American Society of Nephrology, Norman S. Coplan award from the Satellite Healthcare, San Antonio area foundation, and Diabetes Action Research and Education Foundation. This work was also supported by the grants from the National Institute of Diabetes, Digestive and Kidney Diseases (5U01DK085524), VA-Merit Review and the National Center for Research Resources contracts UL1 RR025767 and KL2 RR025766 for the Institute for Integration of Medicine and Science. This work was also supported in part by the National Center for Advancing Translational Science through UCLA CTSI Grant UL1TR000124 and by the National Institute of Diabetes, Digestive, and Kidney Diseases grants R01 DK071185 and U01 DK57249.

References

1. Burrows NR, Ki Y, Geiss LS. Incidence of treatment for end-stage renal disease among individuals with diabetes in the U.S. continues to decline. *Diabetes Care*. 2010; 33:73–77. [PubMed: 20040673]
2. Palmer ND, Freedman BI. Insights into the genetic architecture of diabetic nephropathy. *Curr Diab Rep*. 2012; 12:423–431. [PubMed: 22573336]
3. Ventura H, Piña IL, Lavie CJ. Hypertension and antihypertensive therapy in Hispanics and Mexican Americans living in the United States. *Postgrad Med*. 2011; 123:46–57. [PubMed: 22104453]
4. Cusi K, Ocampo GL. Unmet needs in Hispanic/Latino patients with type 2 diabetes mellitus. *Am J Med*. 2011; 124:S2–S9. [PubMed: 21939795]
5. Garza R, Medina R, Basu S, Pugh JA. Predictors of the rate of renal function decline in non-insulin-dependent diabetes mellitus. *Am J Nephrol*. 1997; 17:59–67. [PubMed: 9057955]
6. Pugh JA, Medina RA, Cornell JC, Basu S. NIDDM is the major cause of diabetic endstage renal disease. More evidence from a tri-ethnic community. *Diabetes*. 1995; 44:1375–1380. [PubMed: 7589841]
7. Pugh JA. Diabetic nephropathy and end-stage renal disease in Mexican Americans. *Blood Purif*. 1996; 14:286–292. [PubMed: 8873954]
8. Bhandari K, Duggirala R, Kawalitt I, Arya R, et al. Genome-wide search for albuminuria susceptibility genes in Mexican Americans. *Diabetes*. 2001; 50:A240–A241.
9. Puppala S, Arya R, Thameem F, Arar NH, et al. Genotype by diabetes interaction effects on the detection of linkage of glomerular filtration rate to a region on chromosome 2q in Mexican Americans. *Diabetes*. 2007; 56:2818–2828. [PubMed: 17698600]
10. Arar NH, Voruganti SV, Nath SD, Thameem F, et al. A Genome-Wide Search for Linkage to Chronic Kidney Disease in a Community-based Sample: The SAFHS. *Nephrol Dial Transplant*. 2008; 23:3184–3191. [PubMed: 18443212]
11. Schelling JR, Abboud HE, Nicholas SB, et al. Family Investigation of Nephropathy and Diabetes Research Group. Genome-wide scan for estimated glomerular filtration rate in multi-ethnic diabetic populations: the Family Investigation of Nephropathy and Diabetes (FIND). *Diabetes*. 2008; 57:235–243. [PubMed: 18003762]
12. Forsblom CM, Kanninen T, Lehtovirta M, Saloranta C, et al. Heritability of albumin excretion rate in families of patients with Type II diabetes. *Diabetologia*. 1999; 42:1359–1366. [PubMed: 10550421]
13. Krolewski AS, Poznik GD, Placha G, Canani L, et al. A genome-wide linkage scan for genes controlling variation in urinary albumin excretion in type II diabetes. *Kidney Int*. 2006; 69:129–136. [PubMed: 16374433]

14. Kiryluk K. Quantitative genetics of renal function: tackling complexities of the eGFR phenotype in gene mapping studies. *Kidnet Int.* 2008; 74:1109–1112.
15. Knowler WC, Coresh J, Elston RC. Family Investigation of Nephropathy and Diabetes Research Group. The Family Investigation of Nephropathy and Diabetes (FIND): design and methods. *J Diabetes Complications.* 2005; 19:1–9. [PubMed: 15642484]
16. Iyengar SK, Abboud HE, Goddard KA, et al. Family Investigation of Nephropathy and Diabetes Research Group. Genome-wide scans for diabetic nephropathy and albuminuria in multiethnic populations: the family investigation of nephropathy and diabetes (FIND). *Diabetes.* 2007; 56:1577–1585. [PubMed: 17363742]
17. Imperatore G, Hanson RL, Pettitt DJ, Kobes S, et al. Sib-pair linkage analysis for susceptibility genes for microvascular complications among Pima Indians with type 2 diabetes. Pima Diabetes Genes Group. *Diabetes.* 1998; 47:821–830. [PubMed: 9588456]
18. Vardarli I, Baier LJ, Hanson RL, Akkoyun I, et al. Gene for susceptibility to diabetic nephropathy in type 2 diabetes maps to 18q22.3-23. *Kidney Int.* 2002; 62:2176–2183. [PubMed: 12427143]
19. Bowden DW, Colicigno CJ, Langefeld CD, Sale MM, et al. A genome scan for diabetic nephropathy in African Americans. *Kidney Int.* 2004; 66:1517–1526. [PubMed: 15458446]
20. Igo RP Jr, Iyengar SK, Nicholas SB, Goddard KA, et al. Genomewide linkage scan for diabetic renal failure and albuminuria: the FIND study. *Am J Nephrol.* 2011; 33:381–389. [PubMed: 21454968]
21. Kao WH, Klag MJ, Meoni LA, Reich D, et al. MYH9 is associated with nondiabetic endstage renal disease in African Americans. *Nat Genet.* 2008; 40:1185–1192. [PubMed: 18794854]
22. Kopp JB, Smith MW, Nelson GW, Johnson RC, et al. MYH9 is a major-effect risk gene for focal segmental glomerulosclerosis. *Nat Genet.* 2008; 40:1175–1184. [PubMed: 18794856]
23. Freedman BI, Hicks PJ, Bostrom MA, Comeau ME, et al. Non-muscle myosin heavy chain 9 gene MYH9 associations in African Americans with clinically diagnosed type 2 diabetes mellitus-associated ESRD. *Nephrol Dial Transplant.* 2009; 24:3366–3371. [PubMed: 19567477]
24. Genovese G, Tonna SJ, Knob AU, Appel GB, et al. A risk allele for focal segmental glomerulosclerosis in African Americans is located within a region containing APOL1 and MYH9. *Kidney Int.* 2010; 78:698–704. [PubMed: 20668430]
25. McDonough CW, Palmer ND, Hicks PJ, Roh BH, et al. A genome-wide association study for diabetic nephropathy genes in African Americans. *Kidney Int.* 2011; 79:563–572. [PubMed: 21150874]
26. Puppala S, Dodd GD, Fowler S, Arya R, et al. A genomewide search finds major susceptibility loci for gallbladder disease on chromosome 1 in Mexican Americans. *Am J Hum Genet.* 2006; 78:377–392. [PubMed: 16400619]
27. Lehman DM, Leach RJ, Johnson-Pais T, Hamlington J, et al. Evaluation of tight junction protein 1 encoding zona occludens 1 as a candidate gene for albuminuria in a Mexican American population. *Exp Clin Endocrinol Diabetes.* 2006; 114:432–437. [PubMed: 17039425]
28. Thameem F, Puppala S, He X, Arar NH, et al. Evaluation of Gremlin1 (GREM1) as a candidate susceptibility gene for albuminuria-related traits in Mexican Americans with Type 2 Diabetes. *Metabolism.* 2009; 58:1496–1502. [PubMed: 19577778]
29. MacCluer JW, Stern MP, Almasy L, Atwood LA, et al. Genetics of atherosclerosis risk factors in Mexican Americans. *Nutr Rev.* 1999; 57:S59–S65. [PubMed: 10391028]
30. Arar N, Nath S, Thameem F, Bauer R, et al. Genome-wide scans for microalbuminuria in Mexican Americans: the San Antonio Family Heart Study. *Genet Med.* 2007; 9:80–87. [PubMed: 17304049]
31. Shike T, Gohda T, Tanimoto M, Kobayashi, et al. Chromosomal mapping of a quantitative trait locus for the development of albuminuria in diabetic KK/Ta mice. *Nephrol Dial Transplant.* 2005; 20:879–885. [PubMed: 15769825]
32. Klupa T, Malecki MT, Pezzolesi M, Ji L, et al. Further evidence for a susceptibility locus for type 2 diabetes on chromosome 20q13.1-q13.2. *Diabetes.* 2000; 49:2212–2216. [PubMed: 11118028]
33. Permutt MA, Wasson J, Love-Gregory L, Ma J, et al. Searching for type 2 diabetes genes on chromosome 20. *Diabetes.* 2002; 51:S308–S315. [PubMed: 12475768]

34. Ryffel GU. Mutations in the human genes encoding the transcription factors of the hepatocyte nuclear factor (HNF)1 and HNF4 families: functional and pathological consequences. *J Mol Endocrinol.* 2001; 27:11–29. [PubMed: 11463573]
35. Niehof M, Borlak J. HNF4 alpha and the Ca-channel TRPC1 are novel disease candidate genes in diabetic nephropathy. *Diabetes.* 2008; 57:1069–1077. [PubMed: 18184923]
36. Mooyaart AL, Valk EJ, van Es LA, Bruijn JA, et al. Genetic associations in diabetic nephropathy: a meta-analysis. *Diabetologia.* 2011; 54:544–553. [PubMed: 21127830]
37. Thameem F, Puppala S, Schneider J, Bhandari BK, et al. The Gly(972)Arg variant of Human Insulin Receptor Substrate 1 (IRS1) gene is associated with variation in Glomerular Filtration Rate (GFR) likely through impaired insulin receptor signaling. *Diabetes.* 2012; 61:2385–2393. [PubMed: 22617042]
38. Köttgen A, Pattaro C, Böger CA, Fuchsberger C, et al. New loci associated with kidney function and chronic kidney disease. *Nat Genet.* 2010; 42:376–384. [PubMed: 20383146]
39. Leak TS, Perlegas PS, Smith SG, Keene KL, et al. Variants in intron 13 of the ELMO1 gene are associated with diabetic nephropathy in African Americans. *Ann Hum Genet.* 2009; 73:152–159. [PubMed: 19183347]
40. Hanson RL, Millis MP, Young NJ, Kobes S, et al. ELMO1 variants and susceptibility to diabetic nephropathy in American Indians. *Mol Genet Metab.* 2010; 101:383–390. [PubMed: 20826100]
41. Ahluwalia TS, Lindholm E, Groop LC. Common variants in CNDP1 and CNDP2, and risk of nephropathy in type 2 diabetes. *Diabetologia.* 2011; 54:2295–2302. [PubMed: 21573905]
42. Kim S, Abboud HE, Pahl MV, Tayek J, et al. Examination of association with candidate genes for diabetic nephropathy in a Mexican American population. *Clin J Am Soc Nephrol.* 2010; 5:1072–1078. [PubMed: 20299368]
43. Wong C, Kanetsky P, Raj D. Genetic polymorphisms of the RAS-cytokine pathway and chronic kidney disease. *Pediatr Nephrol.* 2008; 23:1037–1051. [PubMed: 18481112]
44. Thameem F, Puppala S, Arar N, Blangero J, et al. Genetic polymorphisms in the reninangiotensin system (RAS) genes and their association analysis with type 2 diabetes and related traits in Mexican Americans. *Diabetes Res Clin Pract.* 2008; 79:e14–e16. [PubMed: 17936396]
45. Thameem F, Voruganti VS, He X, Nath SD, et al. Genetic variants in the reninangiotensin system genes are associated with cardiovascular-renal-related risk factors in Mexican Americans. *Hum Genet.* 2009; 124:557–559. [PubMed: 18985387]
46. Zeng Z, Li L, Zhang Z, Li Y, et al. A meta-analysis of three polymorphisms in the endothelial nitric oxide synthase gene (NOS3) and their effect on the risk of diabetic nephropathy. *Hum Genet.* 2010; 127:373–381. [PubMed: 20049477]
47. Thameem F, Puppala S, Arar NH, Stern MP, et al. Endothelial nitric oxide synthase (eNOS) gene polymorphisms and their association with type 2 diabetes-related traits in Mexican Americans. *Diab Vasc Dis Res.* 2008; 5:109–113. [PubMed: 18537098]
48. Nath SD, He X, Voruganti VS, Blangero J, et al. The 27-bp repeat polymorphism in intron 4 (27 bp-VNTR) of endothelial nitric oxide synthase (eNOS) gene is associated with albumin to creatinine ratio in Mexican Americans. *Mol Cell Biochem.* 2009; 331:201–205. [PubMed: 19468830]
49. Calle R, McCarthy MI, Banerjee P, Zeggini E, et al. Paraoxonase 2 (PON2) polymorphisms and development of renal dysfunction in type 2 diabetes: UKPDS 76. *Diabetologia.* 2006; 49:2892–2899. [PubMed: 17096118]
50. Thameem F, Puppala S, Arar NH, Farook VS, et al. The Cys(311)Ser polymorphism of paraoxonase 2 (PON2) is associated with albumin-to-creatinine ratio (ACR) in Mexican Americans. *Nephrol Dial Transplant.* 2008; 23:416–417. [PubMed: 17940058]
51. Thameem F, He X, Voruganti VS, Nath SD, et al. Evaluation of polymorphisms in paraoxonase 2 (PON2) gene and their association with cardiovascular-renal disease risk in Mexican Americans. *Kidney Blood Press Res.* 2009; 32:200–204. [PubMed: 19546579]
52. Ned RM, Yesupriya A, Imperatore G, Smelser DT, et al. Inflammation gene variants and susceptibility to albuminuria in the U.S. population: analysis in the Third National Health and Nutrition Examination Survey (NHANES III), 1991–1994. *BMC Med Genet.* 2010; 11:155–169. [PubMed: 21054877]

53. Liberopoulos E, Siamopoulos K, Elisaf M. Apolipoprotein E and renal disease. *Am J Kidney Dis.* 2004; 43:223–233. [PubMed: 14750087]
54. Chu AY, Parekh RS, Astor BC, Coresh J, et al. Association of APOE polymorphism with chronic kidney disease in a nationally representative sample: a Third National Health and Nutrition Examination Survey (NHANES III) Genetic Study. *BMC Med Genet.* 2009; 10:108. [PubMed: 19852818]
55. Seldin MF, Pasaniuc B, Price AL. New approaches to disease mapping in admixed populations. *Nat Rev Genet.* 2011; 12:523–528. [PubMed: 21709689]
56. Tian C, Hinds DA, Shigeta R, Adler SG, et al. A genomewide single-nucleotide polymorphism panel for Mexican American admixture mapping. *Am J Hum Genet.* 2007; 80:1014–1023. [PubMed: 17557415]
57. Seldin MF, Tian C, Pahl M, et al. The FIND Consortium. Identification of chromosomal risk loci for diabetic nephropathy in Mexican-Americans using mapping by admixture linkage disequilibrium: The Family Study of Nephropathy and Diabetes (FIND) Study. *JASN.* 2008; 19:57A.
58. Adler S, Pahl M, Abboud H, Nicholas S, et al. Mexican-American admixture mapping analyses for diabetic nephropathy in type 2 diabetes mellitus. *Semin Nephrol.* 2010; 30:141–149. [PubMed: 20347643]
59. Gibson G. Rare and common variants: twenty arguments. *Nat Rev Genet.* 2012; 13:135–145. [PubMed: 22251874]
60. Wijsman. The role of large pedigrees in an era of high-throughput sequencing. *Hum Genet.* 2012; 131:1555–1563. [PubMed: 22714655]
61. Blangero J, Diego VP, Dyer TD, Almeida M, et al. A kernel of truth: statistical advances in polygenic variance component models for complex human pedigrees. *Adv Genet.* 2013; 81:1–31. [PubMed: 23419715]
62. Lehman DM, Hamlington J, Hunt KJ, Leach RJ, et al. A novel missense mutation in ADRB3 increases risk for type 2 diabetes in a Mexican American family. *Diabetes Metab Res Rev.* 2006; 22:331–336. [PubMed: 16444766]
63. Levey AS, Stevens LA, Schmid CH, Zhang YL, et al. A new equation to estimate glomerular filtration rate. *Ann Intern Med.* 2009; 150:604–612. [PubMed: 19414839]
64. Inker LA, Schmid CH, Tighiouart H, Eckfeldt JH, et al. Estimating glomerular filtration rate from serum creatinine and cystatin C. *N Engl J Med.* 2012; 367:20–29. [PubMed: 22762315]
65. Reddy MA, Natarajan R. Epigenetics in Diabetic Kidney Disease. *J Am Soc Nephrol.* 2011; 22:2182–2185. [PubMed: 22021712]

Table 1
Summary of genome-wide linkage scans for DN phenotypes in Mexican American Cohorts

Cohorts	Trials	Subjects/ pedigrees	Linkage markers	Covariates used	Linkage analysis	Chromosome locations	LOD/ Z Score	Linkage/ flanking Markers	Positional genes	References
FIND	DN	632 / 478	Illumina IV panel (5500 SNPs)	sex	Haseman- Elston test	11p15	2.2	-	-	13
FIND- MALD	DN	1154 (664 DN and 490 T2DM)	4300 Ancestry informatory markers	Age/sex/ diabetes/ duration/ recruitment site/ admixture mapping	ADMIX- MAP	2q37 1p35 14q32	4.3 (Z) 3.1 3.0	rs721941 rs1564720 rs4267246	<i>IRS1/DNER</i> <i>HDAC1</i> <i>DICER1</i>	55, 56
FIND	ACR	1316/ 478	Illumina IV panel (5500 SNPs)	Age/sex	Haseman- Elston test	22q12	2.3	rs735853	<i>MYH9</i>	13
SAFDGS	ACR	335 / 27	379 microsatellites	Age/sex/ hypertension	Variance component	15q12	3.0	<i>GABRB3</i>	<i>TJP1/ GREM1</i>	21
SAFHS	ACR	486 / 26	404 microsatellites	Age/sex/ BMI/triglycerides/ hypertension	Variance component	20q12	3.5	<i>D20S481</i>	<i>HNF4A</i>	25
SAFHS	ACR	848 / 26	417 microsatellites	Age/sex/ BMI/ diabetes duration/ BP- Med	Variance component	20q12	2.9	<i>D20S481</i>	<i>HNF4A</i>	26
FIND	eGFR	507 / 196	404 microsatellites	diabetes duration/ BP- Med	Haseman- Elston test	1q43, 2p13, 7q36.1, 8q13, 18q22	3.8 3.0 4.2 4.0 2.1	D1S235- D1S1609 D2S1352- D2S441 7S3070- D7S3058 D8S1136- D8S1119 D18S1371- D18S1390	<i>GREM2</i> <i>IL7</i> <i>CNDP1</i>	32
SAFDGS	eGFR	453 / 29	382 microsatellites	Diabetes/ diabetes duration/ SBP/BP- Med	Variance component	2q36q21	3.3 2.9	D2S1363- D2S427 D9S922- D9S1120	<i>IRS1/D</i> <i>NER</i> <i>TRPM 6</i>	34

Cohorts	Trials	Subjects/ pedigrees	Linkage markers	Covariates used	Linkage analysis	Chromosome locations	LOD/ Z Score	Linkage/ flanking Markers	Positional genes	References
SAFHS	eGFR	848 / 26	417 microsatellites	Age/sex/ BMI/ diabetes duration/ BP- Med	Variance component	9q21	3.9	D9S922	TRPM6	26

FIND-Family Investigation of Nephropathy and Diabetes; MALD-Mapping by Admixture Linkage Disequilibrium; SAFDGS-San Antonio Family Diabetes; SAFHS-San Antonio Family Heart Study