Integrated bioinformatics analysis reveals novel key biomarkers and potential candidate small molecule drugs in diabetic nephropathy

Harish Joshi

Endocrine and Diabetes Care Center, Hubballi, Karnataka 580029, India. <https://orcid.org/0000-0002-3817-5194>

Basavaraj Vastrad

Department of Pharmaceutics, SET'S College of Pharmacy, Dharwad, Karnataka, 580002, India.
<https://orcid.org/0000-0003-2202-7637>

Nidhi Joshi

Dr. D. Y. Patil Medical College, Kolhapur, 416006, Maharashtra, India. <https://orcid.org/0000-0001-8067-3448>

Anandkumar Tengli

Department of Pharmaceutical Chemistry, JSS College of Pharmacy, Mysuru and JSS Academy of Higher Education & Research, Mysuru- 570015, Karnataka, India <https://orcid.org/0000-0001-8076-928X>

Chanabasayya Vastrad (✉ channu.vastrad@gmail.com)

Biostatistics and Bioinformatics, Chanabasava Nilaya, Bharthinagar, Dharwad, Karnataka, 580001, India. <https://orcid.org/0000-0003-3615-4450>

Iranna Kotturshetti

Department of Ayurveda, Rajiv Gandhi Education Society's Ayurvedic Medical College, Ron 562209, Karnataka, India <https://orcid.org/0000-0003-1988-7345>

Research Article

Keywords: bioinformatics analysis, protein-protein interaction network, differentially expressed genes, diabetic nephropathy, novel biomarkers

Posted Date: December 22nd, 2020

DOI: <https://doi.org/10.21203/rs.3.rs-132705/v1>

License: © ⓘ This work is licensed under a Creative Commons Attribution 4.0 International License.
[Read Full License](#)

Abstract

The underlying molecular mechanisms of diabetic nephropathy (DN) have yet not been investigated clearly. In this investigation, we aimed to identify key genes involved in the pathogenesis and prognosis of DN. We selected expression profiling by high throughput sequencing dataset GSE142025 from Gene Expression Omnibus (GEO) database. The differentially expressed genes (DEGs) between DN and normal control samples were analyzed with limma package. Gene ontology (GO) and REACTOME enrichment analysis were performed using ToppGene. Then we established the protein-protein interaction (PPI) network, miRNA-DEG regulatory network and TF-DEG regulatory network. The diagnostic values of hub genes were performed through receiver operating characteristic (ROC) curve analysis. Finally, the candidate small molecules as potential drugs to treat DM were predicted using molecular docking studies. Through expression profiling by high throughput sequencing dataset, a total of 549 DEGs were detected including 275 up regulated and 274 down regulated genes. Biological process analysis of functional enrichment showed these DEGs were mainly enriched in cell activation, response to hormone, cell surface, integral component of plasma membrane, signaling receptor binding, lipid binding, immunoregulatory interactions between a lymphoid and a non-lymphoid cell and biological oxidations. DEGs with high degree of connectivity (MDFI, LCK, BTK, IRF4, PRKCB, EGR1, JUN, FOS, ALB and NR4A1) were selected as hub genes from protein-protein interaction (PPI) network, miRNA-DEG regulatory network and TF-DEG regulatory network. The ROC curve analysis confirmed that hub genes were high diagnostic values. Finally, the significant small molecules were obtained based on molecular docking studies. Our results indicated that MDFI, LCK, BTK, IRF4, PRKCB, EGR1, JUN, FOS, ALB and NR4A1 could be the potential novel biomarkers for GC diagnosis prognosis and the promising therapeutic targets. The present study may be crucial to understanding the molecular mechanism of DN initiation and progression.

Introduction

Diabetic nephropathy (DN) is a common and devastating microvascular complication of the kidneys induced by diabetes mellitus [1]. The incidence of DN is reported to be 30% to 40% patients with diabetes [2] and is the main cause of end - stage renal disease throughout the world in both developed and developing countries [3]. Numerous risk factors may affect DN progression [4]; however, how these factors affect the development of DN requires further study and no effective method has been developed. Despite important developments toward an understanding of the pathophysiology of DN, early diagnosis, therapeutic interference, and underlying molecular pathogenesis hover a require [5]. Therefore, enlighten the rare nature belonging to DN is predominant in expand therapies to improve patient outcome.

The exact mechanisms of DN are still unknown. A number of investigation have reported possible roles of some genes and pathways such as UCP1-3 [6] and JAK/STAT3 signaling pathway [7] in the development of DN. However, these reports only concentrated on any certain molecule, gene or pathway, ignoring that the development process involves aberrant expression of a variety of genes and pathways,

among which some proteins might interact with other proteins and thus play an essential role in the DN [8]. Hub genes may act as prognostic or diagnostic biomarkers or treatment targets for DN [9]. Therefore, it is urgent to search new biomarkers for DN with a powerful genome-wide technology.

The high-throughput platforms for analysis of gene expression, such as high throughput RNA sequencing, are increasingly valued as promising tools in medical field with great clinical applications: molecular diagnosis, prognosis prediction and new drug targets discovery [10]. The Gene Expression Omnibus (GEO) is a database and online resource for the gene expression of any species. In the current investigation, expression profiling by high throughput sequencing dataset (GSE142025) was downloaded. In total, there are 28 DN samples and 8 normal control samples datasets available. A data processing standard was used to filter the DEGs on the limma package of R language, followed by Gene Ontology (GO) and pathway enrichment analyses using ToppGene software. The DEGs protein-protein interaction (PPI) network and modular analysis were integrated using InnateDB interactome software to identify hub genes in DN. The regulatory network of miRNA and TF was constructed and the target genes with high degree of connectivity were selected. Receiver operating characteristic (ROC) curve analysis was used to predicting power of the gene signature. Molecular docking experiment was implemented for selected hub genes. This study will enhance our understanding of the molecular mechanisms of DN.

Materials And Methods

Data source

The DN expression profiling by high throughput sequencing dataset GSE142025 was downloaded from the NCBI GEO database (<http://www.ncbi.nlm.nih.gov/geo>) [11]. The dataset GSE142025 was based on the GPL20301 platform (Illumina HiSeq 4000 (Homo sapiens)), including 28 DN samples and 8 normal control samples.

Identification of DEGs

Identify a gene that are differentially expressed across experimental conditions, was used to identify DEGs in the GSE142025 dataset with the limma package of R language, which had been processed, normalized and transformed. An adjusted P-value was retrieved by implement the Benjamini-Hochberg false discovery rate (FDR) correction on the original P-value, and a fold change threshold was preferred based on our plan to target on statistically significant DEGs [12]. Only genes with a fold change > 1.35 for up regulated genes and fold change < -1.24 for down regulated genes, and adjusted P-value < 0.05 were considered as statistically significant DEGs. A volcano plot and heat map of the identified DEGs was also constructed, using an R package.

Gene ontology and pathway enrichment analysis of DEGs

In the current investigation, the significant enrichment analysis of DEGs was assessed based on the Gene Ontology (GO) and REACTOME using the ToppGene (ToppFun)

(<https://toppgene.cchmc.org/enrichment.jsp>) [13], an online tool for functional annotation analysis. GO analysis (<http://geneontology.org/>) [14] is a accepted, effective method for annotating genes and gene products, and for identifying unique biological aspect of high-throughput genome or transcriptome data , including 3 categories: biological process (BP), cellular component (CC) and molecular function (MF). REACTOME database (<https://reactome.org/>) [15] is a knowledge database for the assignment of specific pathways to sets of DEGs, thus linking-omics data with higher-order functional information. Comprehensively mapping genes to relevant biological annotations in databases such as ToppGene is critical for the success of any high-throughput gene functional analysis. An FDR of <0.05 was set as the cut-off.

Construction of protein-protein interaction (PPI) network

The online database InnateDB interactome (<https://www.innatedb.com/>) [16] was used to construct a PPI network of the proteins encoded by DEGs. Then, Cytoscape software (Version 3.8.1, National Institute of General Medical Sciences) [17] was utilized to perform protein interaction association network analysis and analyze the interaction correlation of the candidate proteins encoded by the DEGs in DN. Next, the Network Analyzer plugin for Cytoscape was applied to calculate node degree [18], betweenness centrality [19], stress centrality [20] and closeness centrality [21]. Finally, the PEWCC1 (<http://apps.cytoscape.org/apps/PEWCC1>) [22] for Cytoscape was used to collect the significant modules in the PPI network complex.

Integrated regulatory network construction

The integrated regulatory network of miRNAs (microRNAs) and TFs (transcription factors) was constructed based on standardized integration of numerous high-throughput datasets. It granted a plan including a set of hub genes, miRNAs and TFs for analyzing multi-level regulation in DN. The miRNAs associated with DEGs were selected from miRNet (<https://www.mirnet.ca/>) [23] Database (The integration database of TarBase, miRTarBase, miRecords, miRanda (S mansoni only), miR2Disease, HMDD, PhenomiR, SM2miR, PharmacomiR, EpimiR, starBase, TransmiR, ADmiRE, and TAM 2.0), and TFs associated with DEGs were selected from NetworkAnalyst database (<https://www.networkanalyst.ca/>) [24] Database (The integration database of JASPAR). The miRNA-DEG regulatory network and TF-DEG regulatory network were constructed by using Cytoscape software, which is open source software for visualizing complex networks.

Validation of the hub genes

The diagnostic value of validated hub genes was assessed using receiver operating characteristic (ROC) curve analysis using the pROC in R with GLM prediction model [25] and area under the curve (AUC) was calculated.

Molecular docking studies

The module SYBYL-X 2.0 perpetual software were used for Surflex-Docking of the designed molecules. The molecules were sketched by using ChemDraw Software and imported and saved in sdf. format using openbabel free software. The protein structures of CyclinB1 (CCNB1) its co-crystallisedprotein of PDB code 4Y72, 5H0V and Four and half LIM domains 2 (FHL2) its NMR structure of proteins 2D8Z and 2EHE was retrieved from Protein Data Bank [26-27]. Together with the TRIPOS force field, GasteigerHuckel (GH) charges were added to all designed derivatives for the structure optimization process. In addition, energy minimization was carried out using MMFF94s and MMFF94 algorithm process. Protein processing was carried out after the incorporation of protein. The co-crystallized ligand and all water molecules were removed from the crystal structure; more hydrogens were added and the side chain was set. TRIPOS force field was used for the minimization of structure. The compounds' interaction efficiency with the receptor was represented by the Surflex-Dock score in kcal / mol units. The interaction between the protein and the ligand, the best pose was incorporated into the molecular area. The visualisation of ligand interaction with receptor is done by using discovery studio visualizer.

Results

Identification of DEGs

DN and normal control samples (28 and 9, respectively) were first analyzed. Limma was used to analyze the series of each chip and to identify the DEGs. Following analysis of GSE142025 dataset, 549 DEGs (275 up regulated and 274 down regulated) genes were identified (Fig. 1. and Table 1). The results of the cluster analysis of DEGs revealed significant differences between the DN and normal control samples (Fig. 2).

Gene ontology and pathway enrichment analysis of DEGs

The identified DEGs were uploaded to the online software ToppGene for GO and REACTOME pathway enrichment analyses and results are listed in Table 2 and Table 3. The results of the GO analysis revealed that up regulated genes were significantly enriched in BP, including cell activation and regulation of immune system process, whereas down regulated genes were significantly enriched in response to hormone and ion transport. In terms of CC, the up regulated genes were enriched in cell surface and intrinsic component of plasma membrane, whereas down regulated genes were enriched in integral component of plasma membrane and nuclear chromatin. In terms of MF, the up regulated genes were enriched in signaling receptor binding and identical protein binding, whereas down regulated genes were enriched in lipid binding and transporter activity. REACTOME pathway analysis revealed that the up regulated genes were highly associated with pathways including immunoregulatory interactions between a lymphoid and a non-lymphoid cell, and innate immune system, whereas down regulated genes were significantly enriched in biological oxidations and GPCR ligand binding.

Construction of protein-protein interaction (PPI) network

The DEG expression profiles in DN were constructed according to the information in the InnateDB interactome database. The PPI network of DEGs is consisted of 2718 nodes and 4477 edges (Fig. 3A). There are 10 genes selected as hub genes, such as MDFI, LCK, BTK, IRF4, PRKCB, EGR1, JUN, FOS, ALB and NR4A1 are listed in Table 4. A two significant modules were obtained from PPI network of DEGs using PEWCC1, including 15 nodes and 36 edges (Fig. 3B) and 7 nodes and 12 edges (Fig. 3C). Gene ontology and pathway enrichment analysis revealed that genes in these modules were mainly involved in innate immune system, tmmunoregulatory interactions between a lymphoid and a non-lymphoid cell, cell activation, regulation of immune system process, cell surface, response to hormone, cytokine signaling in immune system, metabolism of proteins and nuclear chromatin.

Integrated regulatory network construction

The miRNA-DEG regulatory network had 8997 interactions (involving 1973 miRNAs and 248 DEGs) (Fig. 4A). Moreover, COL1A1 was targeted by 178 miRNAs (ex, hsa-mir-4492), IRF4 was targeted by 140 miRNAs (ex, hsa-mir-4319), MYBL2 was targeted by 83 miRNAs (ex, hsa-mir-637), PRKCB was targeted by 81 miRNAs (ex, hsa-mir-1261), IL2RB was targeted by 54 miRNAs (ex, hsa-mir-4300), JUN was targeted by 144 miRNAs (ex, hsa-mir-3943), EGR1 was targeted by 132 miRNAs (ex, hsa-mir-548e-3p), ZFP36 was targeted by 130 miRNAs (ex, hsa-mir-6077), FOS was targeted by 105 miRNAs (ex, hsa-mir-5586-5p) and DUSP1 was targeted by 97 miRNAs (ex, hsa-mir-4458) are listed in Table 5. The TF-DEG regulatory network had 1954 interactions (involving 81 TFs and 250 DEGs) (Fig. 4B). Moreover, IRF4 was targeted by 10 TFs (ex, NFATC2), LCK was targeted by 10 TFs (ex, YY1), RET was targeted by 10 TFs (ex, NR2C2), MAP1LC3C was targeted by 10 TFs (ex, MAX), IL2RB was targeted by 8 TFs (ex, PDX1), ATF3 was targeted by 19 TFs (ex, TP53), EGR1 was targeted by 16 TFs (ex, ARID3A), JUNB was targeted by 15 TFs (ex, SRF), FOS was targeted by 13 TFs (ex, CREB1) and PTPRO was targeted by 13 TFs (ex, NR3C1) are listed in Table 5.

Validation of the hub genes

A ROC curve was plotted to evaluate the diagnostic value of MDFI, LCK, BTK, IRF4, PRKCB, EGR1, JUN, FOS, ALB and NR4A1 (Fig. 5). The AUCs for the 10 genes were 0.946, 0.853, 0.835, 0.871, 0.804, 0.991, 0.964, 0.964, 0.839 and 0.982, respectively.

Molecular docking studies

In the current research, the docking simulation was conducted to recognize the active site conformation and major interactions responsible for complex stability with the binding sites receptor. Novel molecules containing thiazolidindioneheterocyclic ring were designed and performed docking studies using Sybyl X 2.1 drug design software. Molecules containing thiazolidindioneheterocyclic ring is designed based on the structure of the pioglitazone, is most commonly used alone or in combination with other antidiabetic drug. In diabetic nephropathy pregablin is used to relieve the pain and is taken as a standard. The proteins which are over expressed in diabetic nephropathy are selected for docking studies. The X- RAY crystallographic structure of one proteins of each over expressed PRKCB its co-crystallised protein of PDB

code 5T5T and IRF4 its co-crystallised protein of PDB code 5UTZ were selected for docking. The investigations of designed molecules were performed to identify the potential molecule. The most of the designed molecules obtained C-score greater than 5 and are active having the c-score greater than 5 are said to be an active, among total of 56 designed molecules few molecules have excellent good binding energy (C-score) greater than 8 respectively. Few of the designed molecules SALPYR 12, SALISO 11 and SLPYR 13 (Fig. 6) shown excellent binding score of 8.518, 8.437 and 8.083 with 5T5T and no molecules with excellent binding score with 5UTZ respectively. Few of the molecules BENZPYR 7 and BENZPYR 1 obtained with moderate binding score of 4.5897 and 5.5881 with 5T5T and molecules of SALISO 2, SALISO 8 & SALPYR 4 with binding score of 5.8485, 5.5892 & 5.5222 with PDB code 5UTZ respectively. Molecules of BENZPYR 01, BENZPYR 5, BENZPYR 2 and SALPYR 7 shown weak binding score of 4.5881, 4.4581, 4.4092 and 4.3984 and the molecules SALPYR 11, BENZISO 5, BENZISO 7 and BENZISO 13 with binding score 3.604, 3.5198, 3.5005 and 3.4955 with 5T5T and 5UTZ, the values are depicted in Table 6. The binding score of the predicted molecules are compared with that of the standard Pregablin used in diabetic nephropathy, the standard obtained moderate binding score with 5T5T and weak binding score with 5UTZ respectively. The molecule 26 has highest binding score its interaction with protein 5T5T and hydrogen bonding and other bonding interactions with amino acids are depicted by 3D and 2D figures (Fig. 7 and Fig. 8).

Discussion

DN remains end-stage renal disease worldwide because of its complicated molecular mechanisms and cellular heterogeneity, and its prevalence rise every year [28]. Therefore, recognition of DN may offer clinicians novel tools that can be used to treat the disease. Extensive genomic investigations showing the effects of genes have accepted noticeable attention. Many potential and valuable genes must be identified to develop the clinical outcome for DN patients. However, the number of specific molecular biomarkers that can be used to show therapeutic effects is still limited, and prognostic factors are essential for the treatment of DN patients. Therefore, to diminish mortality and develop DN prognosis, there is a critical demand for the screening of molecular biomarkers of DN.

To better understand the genetic modifications occurring during DN advancement, bioinformatics methods were used to extract data from the GSE7803 and GSE142025 expression profiling by high throughput sequencing. In this investigation, we identified 549 DEGs (275 up regulated and 274 down regulated) between DN and normal control. Xie et al [29] and Zhou et al [30] demonstrate that increased activity of polymorphic CFHR1 and RGS1 genes play a key role in nephropathy progression. Previous investigations report that polymorphic GREM1 gene plays an essential role in progression of DN [31]. Sun et al [32] find that CCL19 is responsible for renal inflammation and fibrosis in DN. Martinelli-Boneschi et al [33] revealed that polymorphic COL6A5 gene is involved in neuropathic chronic itch, but this gene might be liable for progression of DN. Hall et al [34] observed that the expression of CIDEA (cell death inducing DFFA like effector c) play key role in obesity, but this gene might be involved in DN progression. NR4A1 drives DN growth through mitochondrial fission and mitophagy [35]. Recent study has reported that low expression of NR4A2 is associated with myocardial infarction [36], but this gene might be linked

with progression of DN. EGR1 is required for fibrosis and inflammatory response in DN [37]. ATF3 expression has been implicated in DN [38]. Polymorphic NR4A3 gene may contribute to type 2 diabetes progression [39], but this gene might be associated with development of DN. KLK1 functions in DN progression can be used for predicting the progression and prognosis of the disease [40].

A series of DEGs were discovered to be enriched in the GO functions and pathways. SERPINA3 [41], IKZF1 [42], BTK (Bruton tyrosine kinase) [43], C1QA [44], CD1C [45] and CCL13 [46] have a key role in lupus nephritis, but these genes might be liable for advancement of DN. TNFSF14 [47], ITGAL (integrin subunit alpha L) [48], PLAC8 [49], ADRA2A [50], CCL21 [51], ALOX5 [52], CNR2 [53], COL1A1 [54], WNT7A [55], SLAMF1 [56], CD3D [57], LTF (lactotransferrin) [58], MIR27B [59], PDK4 [60], UCN3 [61], PCK1 [62], CEL (carboxyl ester lipase) [63], TRPM6 [64], MTTP (microsomal triglyceride transfer protein) [65], CYP2C8 [66] and CYP3A4 [67] have important role in the progression of type 2 diabetes via inflammation, but these genes might be crucial role in DN progression. Zhang et al [68], Ellenbroek et al [69], Guo et al [70] and Tillmanns et al [71] have shown that MZB1, LAIR1, MIR142 and FAP (fibroblast activation protein alpha) modulating mitochondrial function and alleviating inflammation in myocardial infarction, but these genes might be involved in progression of DN. IRF4 plays a key role in the obesity-induced insulin resistance [72], but this gene might be responsible for development of DN. MDK (midkine) [73], CCR2 [74], SAA1 [75], C3 [76], CD19 [77], CCR5 [78], CXCR3 [79], FABP4 [80], GDF15 [81], IGF2 [82], IGFBP1 [83] and IL6 [84] are important in the progression of DN through inflammation. A previous study has shown that UBASH3A [85], SIRPG (signal regulatory protein gamma) [86], IKZF3 [87], CD1D [88], CD2 [89], CD48 [90], CD247 [91] and CYP27B1 [92] are liable for progression of type 1 diabetes through inflammation, but these genes might be key for progression of DN. SIT1 [93], JAML (junction adhesion molecule like) [94], TIMP1 [95], PRKCB (protein kinase C beta) [96], MMP7 [97], WNT7B [98], WNT10A [99], DUSP1 [100], WT1 [101], APOC3 [102], ERFF1 [103], HCN2 [104], MME (membrane metalloendopeptidase) [105], STRA6 [106], SLC12A3 [107] and GC (GC vitamin D binding protein) [108] expedites epithelial to mesenchymal transition and renal fibrosis in DN. Previous studies have found CFD (complement factor D) [109], DOCK2 [110], LYZ (lysozyme) [111], CD5L [112], SCARA5 [113], VCAN (versican) [114], GDF5 [115], SFRP2 [116], BTG2 [117], ZFP36 [118], GPR3 [119], OLR1 [120], PM20D1 [121] and UGT2B7 [122] to be expressed in obesity, but these genes might be liable for advancement of DN. Polymorphic FCRL3 [123], FCGR2B [124], COMP (cartilage oligomeric matrix protein) [125], ERF (erythroferrone) [126] and NPHS1 [127] expression can be altered by inflammation, which might involved in nephropathy. The expression of COL1A2 [128], LCK (LCK proto-oncogene, Src family tyrosine kinase) [129], LCN2 [130] and APOB (apolipoprotein B) [131] are key for progression of diabetic retinopathy, but these genes might be essential for DN development. COL3A1 [132], PER1 [133], JUN (Jun proto-oncogene, AP-1 transcription factor subunit) [134], SLC26A4 [135], F2RL3 [136], CYP4A11 [137] and CYP4F2 [138] play an important role in the hypertension, but these genes might be involved in progression of DN.

Based on the PPI network and module analysis, we obtained top hub genes in the whole network. Onions et al. [139] showed that ALB (albumin) was potential biomarkers of DN and disease progression. Novel biomarkers such as MDFI (MyoD family inhibitor), FOS (Fos proto-oncogene, AP-1 transcription factor

subunit), SH2D1A, SLA2, TRAT1, CD3E, JUNB and FOSB might play an important role in the development of DN.

Based on the miRNA-DEG regulatory network and TF-DEG regulatory network, we obtained target in the whole network. Recent investigation reported that the dysregulated activity of MYBL2 was associated with myocardial infarction progression [140], but this gene might be linked with progression of DN. Many investigation have reported the hsa-mir-637 [141] and NR3C1 [142] were linked with progression of hypertension, but these genes might be liable for advancement of DN. Wang et al. [143] noted that hsa-mir-1261 was associated with development of DN. Li et al [144] reported that hsa-mir-4458 expression was an independent marker of prognosis in myocardial infarction, but this gene might be involved in DN. Keller et al [145], Fujimoto et al [146] and Xu et al [147] demonstrated that expression of NFATC2, PDX1 and CREB1 were involved in type 2 diabetes, but these genes might be key for progression of DN. Du et al. [148], Zhang et al. [149] and Zhao et al. [150] found that YY1, TP53 and SRF (serum-response factor) played a key role in DN through the epithelial–mesenchymal transition. Novel targets such as IL2RB, hsa-mir-4492, hsa-mir-4319, hsa-mir-4300, hsa-mir-3943, hsa-mir-548e-3p, hsa-mir-6077, hsa-mir-5586-5p, RET (ret proto-oncogene), MAP1LC3C, PTPRO (protein tyrosine phosphatase receptor type O), NR2C2, MAX (myc-associated factor X) and ARID3A might be responsible for progression of DN.

In the present investigation, the DEGs of DN and normal control samples were analyzed to achieve a better understanding of DN. GO and pathway enrichment analyses of DEGs were applied, and the protein-protein interaction (PPI) network, module and miRNA-DEG regulatory network and TF-DEG regulatory network of these DEGs were also constructed. ROC analysis and molecular docking experiments conducted. The aim of this investigation was to identify essential genes and pathways in DN using bioinformatics analysis, and then to explore the intrinsic mechanisms of DN and distinguish new potential diagnostic and therapeutic biomarkers of DN. We anticipated that these investigations will provide further insight of DN pathogenesis and advancement at the molecular level.

Declarations

Acknowledgement

I thank Weijia Zhang, Icahn School of Medicine at Mount Sinai, Renal, New York, USA, very much, the author who deposited their microarray dataset, GSE133684, into the public GEO database.

Conflict of interest

The authors declare that they have no conflict of interest.

Ethical approval

This article does not contain any studies with human participants or animals performed by any of the authors.

Informed consent

No informed consent because this study does not contain human or animals participants.

Availability of data and materials

The datasets supporting the conclusions of this article are available in the GEO (Gene Expression Omnibus) (<https://www.ncbi.nlm.nih.gov/geo/>) repository. [(GSE142025) (<https://www.ncbi.nlm.nih.gov/geo/query/acc.cgi?acc=GSE142025>)]

Consent for publication

Not applicable.

Competing interests

The authors declare that they have no competing interests.

Author Contributions

H.J - Methodology and validation

B.V - Writing original draft, and review and editing

N.J - Software and resources

A.T - Formal analysis and validation

C.V- Investigation and resources

I.K - Supervision and resources

Authors

Harish Joshi ORCID ID: 0000-0002-3817-5194

Basavaraj Vastrad ORCID ID: 0000-0003-2202-7637

Nidhi Joshi ORCID ID: 0000-0001-8067-3448

Anandkumar Tengli ORCID ID: 0000-0001-8076-928X

Chanabasayya Vastrad ORCID ID: 0000-0003-3615-4450

Iranna Kotturshetti ORCID ID: 0000-0003-1988-7345

References

1. Papadopoulou-Marketou N, Chrousos GP, Kanaka-Gantenbein C. Diabetic nephropathy in type 1 diabetes: a review of early natural history, pathogenesis, and diagnosis. *Diabetes Metab Res Rev*. 2017;33(2):10.1002/dmrr.2841. doi:1002/dmrr.2841
2. Umanath K, Lewis JB. Update on Diabetic Nephropathy: Core Curriculum 2018. *Am J Kidney Dis*. 2018;71(6):884-895. doi:1053/j.ajkd.2017.10.026
3. Qi C, Mao X, Zhang Z, Wu H. Classification and Differential Diagnosis of Diabetic Nephropathy. *J Diabetes Res*. 2017;2017:8637138. doi:1155/2017/8637138
4. Wang G, Ouyang J, Li S, Wang H, Lian B, Liu Z, Xie L. The analysis of risk factors for diabetic nephropathy progression and the construction of a prognostic database for chronic kidney diseases. *J Transl Med*. 2019;17(1):264. doi:1186/s12967-019-2016-y
5. Heerspink HJ, Perkins BA, Fitchett DH, Husain M, Cherney DZ. Sodium Glucose Cotransporter 2 Inhibitors in the Treatment of Diabetes Mellitus: Cardiovascular and Kidney Effects, Potential Mechanisms, and Clinical Applications. *Circulation*. 2016;134(10):752-772. doi:1161/CIRCULATIONAHA.116.021887
6. Lindholm E, Klannemark M, Agardh E, Groop L, Agardh CD. Putative role of polymorphisms in UCP1-3 genes for diabetic nephropathy. *J Diabetes Complications*. 2004;18(2):103-107. doi:1016/S1056-8727(03)00019-9
7. Sun MY, Wang SJ, Li XQ, Shen YL, Lu JR, Tian XH, Rahman K, Zhang LJ, Nian H, Zhang H. CXCL6 Promotes Renal Interstitial Fibrosis in Diabetic Nephropathy by Activating JAK/STAT3 Signaling Pathway. *Front Pharmacol*. 2019;10:224. doi:3389/fphar.2019.00224
8. Yang F, Cui Z, Deng H, Wang Y, Chen Y, Li H, Yuan L. Identification of miRNAs-genes regulatory network in diabetic nephropathy based on bioinformatics analysis. *Medicine (Baltimore)*. 2019;98(27):e16225. doi:1097/MD.00000000000016225
9. Zeng M, Liu J, Yang W, Zhang S, Liu F, Dong Z, Peng Y, Sun L, Xiao L. Identification of key biomarkers in diabetic nephropathy via bioinformatic analysis. *J Cell Biochem*. 2018;10.1002/jcb.28155. doi:1002/jcb.28155
10. Liao W, Jordaan G, Nham P, Phan RT, Pelegrini M, Sharma S. Gene expression and splicing alterations analyzed by high throughput RNA sequencing of chronic lymphocytic leukemia specimens. *BMC Cancer*. 2015;15:714. doi:1186/s12885-015-1708-9
11. Clough E, Barrett, T. The Gene Expression Omnibus Database. *Methods. Mol. Biol*. 2016,1418,93-110. doi:1007/978-1-4939-3578-9_5
12. Ferreira JA. The Benjamini-Hochberg method in the case of discrete test statistics. *Int J Biostat*. 2007;3(1):. doi:2202/1557-4679.1065
13. Chen J, Bardes EE, Aronow BJ, Jegga AG. ToppGene Suite for gene list enrichment analysis and candidate gene prioritization. *Nucleic Acids Res*. 2009;37(Web Server issue):W305-W311. doi:1093/nar/gkp427
14. Thomas PD. The Gene Ontology and the Meaning of Biological Function. *Methods Mol Biol*. 2017;1446:15-24. doi:1007/978-1-4939-3743-1_2

15. Fabregat A, Jupe S, Matthews L, Sidiropoulos K, Gillespie M, Garapati P, Haw R, Jassal B, K€orninger F, May B et al The Reactome Pathway Knowledgebase. *Nucleic Acids Res.* 2018;46(D1):D649–D655. doi:1093/nar/gkx1132
16. Breuer K, Foroushani AK, Laird MR, Chen C, Sribnaia A, Lo R, Winsor GL, Hancock RE, Brinkman FS, Lynn DJ. InnateDB: systems biology of innate immunity and beyond–recent updates and continuing curation. *Nucleic Acids Res.* 2013;41(Database issue):D1228-D1233. doi:1093/nar/gks1147
17. Shannon P, Markiel A, Ozier O, Baliga NS, Wang JT, Ramage D, Amin N, Schwikowski B, Ideker T Cytoscape: a software environment for integrated models of biomolecular interaction networks. *Genome Res* 2003;13(11):2498-2504. doi:1101/gr.1239303
18. Przulj N, Wagle DA, Jurisica I. Functional topology in a network of protein interactions. *Bioinformatics.* 2004;20(3):340–348. doi:1093/bioinformatics/btg415
19. Nguyen TP, Liu WC, Jordán F. Inferring pleiotropy by network analysis: linked diseases in the human PPI network. *BMC Syst Biol.* 2011;5:179. Published 2011 Oct 31. doi:1186/1752-0509-5-179
20. Shi Z, Zhang B. Fast network centrality analysis using GPUs. *BMC Bioinformatics.* 2011;12:149. doi:1186/1471-2105-12-149
21. Fadhal E, Gamielien J, Mwambene EC. Protein interaction networks as metric spaces: a novel perspective on distribution of hubs. *BMC Syst Biol.* 2014;8:6. doi:1186/1752-0509-8-6
22. Zaki N, Efimov D, Berengueres J. Protein complex detection using interaction reliability assessment and weighted clustering coefficient. *BMC Bioinformatics.* 2013;14:163. doi:1186/1471-2105-14-163
23. Fan Y, Xia J (2018) miRNet-Functional Analysis and Visual Exploration of miRNA-Target Interactions in a Network Context. *Methods Mol Biol* 1819:215-233. doi:1007/978-1-4939-8618-7_10
24. Zhou G, Soufan O, Ewald J, Hancock REW, Basu N, Xia J (2019) NetworkAnalyst 3.0: a visual analytics platform for comprehensive gene expression profiling and meta-analysis. *Nucleic Acids Res* 47:W234-W241. doi:1093/nar/gkz240
25. Robin X, Turck N, Hainard A, Tiberti N, Lisacek F, Sanchez JC, Müller M. pROC: an open-source package for R and S+ to analyze and compare ROC curves. *BMC Bioinformatics* 2011;12:77. doi:1186/1471-2105-12-77
26. Khalaf RA, Sabbah D, Al-Shalabi E, Al-Sheikh I, Albadawi G, Abu Sheikha G. Synthesis, Structural Characterization and Docking Studies of Sulfamoyl- Phenyl Acid Esters as Dipeptidyl Peptidase-IV Inhibitors. *Curr Comput Aided Drug Des.* 2018;14(2):142-151. doi:10.2174/1573409914666180308164013
27. Xie Z, Wang G, Wang J, Chen M, Peng Y, Li L, Deng B, Chen S, Li W. Synthesis, Biological Evaluation, and Molecular Docking Studies of Novel Isatin-Thiazole Derivatives as α -Glucosidase Inhibitors. *Molecules.* 2017;22(4):659. doi:3390/molecules22040659
28. Kishore L, Kaur N, Singh R. Distinct Biomarkers for Early Diagnosis of Diabetic Nephropathy. *Curr Diabetes Rev.* 2017;13(6):598-605. doi:2174/1573399812666161207123007
29. Xie J, Kiryluk K, Li Y, Mladkova N, Zhu L, Hou P, Ren H, Wang W, Zhang H, Chen N, et al. Fine Mapping Implicates a Deletion of CFHR1 and CFHR3 in Protection from IgA Nephropathy in Han Chinese. *J*

- Am Soc Nephrol. 2016;27(10):3187-3194. doi:1681/ASN.2015111210
30. Zhou XJ, Nath SK, Qi YY, Sun C, Hou P, Zhang YM, Lv JC, Shi SF, Liu LJ, Chen R, et al. Novel identified associations of RGS1 and RASGRP1 variants in IgA Nephropathy. *Sci Rep*. 2016;6:35781. doi:1038/srep35781
31. McKnight AJ, Patterson CC, Pettigrew KA, Savage DA, Kilner J, Murphy M, Sadlier D, Maxwell AP et al. A GREM1 gene variant associates with diabetic nephropathy. *J Am Soc Nephrol*. 2010;21(5):773-781. doi:1681/ASN.2009070773
32. Sun J, Wang J, Lu W, Xie L, Lv J, Li H, Yang S. MiR-325-3p inhibits renal inflammation and fibrosis by targeting CCL19 in diabetic nephropathy. *Clin Exp Pharmacol Physiol*. 2020;10.1111/1440-1681.13371. doi:1111/1440-1681.13371
33. Martinelli-Boneschi F, Colombi M, Castori M, Devigili G, Eleopra R, Malik RA, Ritelli M, Zoppi N, Dordoni C, Sorosina M, et al. COL6A5 variants in familial neuropathic chronic itch. *Brain*. 2017;140(3):555-567. doi:1093/brain/aww343
34. Hall AM, Brunt EM, Klein S, Finck BN. Hepatic expression of cell death-inducing DFFA-like effector C in obese subjects is reduced by marked weight loss. *Obesity (Silver Spring)*. 2010;18(2):417-419. doi:1038/oby.2009.236
35. Sheng J, Li H, Dai Q, Lu C, Xu M, Zhang J, Feng J. NR4A1 Promotes Diabetic Nephropathy by Activating Mff-Mediated Mitochondrial Fission and Suppressing Parkin-Mediated Mitophagy. *Cell Physiol Biochem*. 2018;48(4):1675-1693. doi:1159/000492292
36. Liu H, Liu P, Shi X, Yin D, Zhao J. NR4A2 protects cardiomyocytes against myocardial infarction injury by promoting autophagy. *Cell Death Discov*. 2018;4:27. doi:1038/s41420-017-0011-8
37. Zha F, Qu X, Tang B, Li J, Wang Y, Zheng P, Ji T, Zhu C, Bai S. Long non-coding RNA MEG3 promotes fibrosis and inflammatory response in diabetic nephropathy via miR-181a/Egr-1/TLR4 axis. *Aging (Albany NY)*. 2019;11(11):3716-3730. doi:18632/aging.102011
38. Zhang H, Liang S, Du Y, Li R, He C, Wang W, Liu S, Ye Z, Liang X, Shi W, et al. Inducible ATF3-NFAT axis aggravates podocyte injury. *J Mol Med (Berl)*. 2018;96(1):53-64. doi:1007/s00109-017-1601-x
39. Mohammad BS, Alireza N, Ramin S. The effect of NR4A3-rs12686676 and XBP1-rs2269577 polymorphisms on type 2 diabetes mellitus susceptibility in an Iranian population: Case-control study. *Gene Reports* 21 (2020) 100854. doi:1016/j.genrep.2020.100854
40. Riad A, Zhuo JL, Schultheiss HP, Tschöpe C. The role of the renal kallikrein-kinin system in diabetic nephropathy. *Curr Opin Nephrol Hypertens*. 2007;16(1):22-26. doi:1097/MNH.0b013e328011a20c
41. Turnier JL, Brunner HI, Bennett M, Aleed A, Gulati G, Haffey WD, Thornton S, Wagner M, Devarajan P, Witte D, et al. Discovery of SERPINA3 as a candidate urinary biomarker of lupus nephritis activity. *Rheumatology (Oxford)*. 2019;58(2):321-330. doi:1093/rheumatology/key301
42. Zhang YM, Zhou XJ, Cheng FJ, Qi YY, Hou P, Zhao MH, Zhang H. Association of the IKZF1 5' UTR variant rs1456896 with lupus nephritis in a northern Han Chinese population. *Scand J Rheumatol*. 2017;46(3):210-214. doi:1080/03009742.2016.1194458

43. Kong W, Deng W, Sun Y, Huang S, Zhang Z, Shi B, Chen W, Tang X, Yao G, Feng X, et al. Increased expression of Bruton's tyrosine kinase in peripheral blood is associated with lupus nephritis. *Clin Rheumatol*. 2018;37(1):43-49. doi:1007/s10067-017-3717-3
44. Racila DM, Sontheimer CJ, Sheffield A, Wisnieski JJ, Racila E, Sontheimer RD. Homozygous single nucleotide polymorphism of the complement C1QA gene is associated with decreased levels of C1q in patients with subacute cutaneous lupus erythematosus. *Lupus*. 2003;12(2):124-132. doi:1191/0961203303lu329oa
45. Kassianos AJ, Wang X, Sampangi S, Muczynski K, Healy H, Wilkinson R. Increased tubulointerstitial recruitment of human CD141(hi) CLEC9A(+) and CD1c(+) myeloid dendritic cell subsets in renal fibrosis and chronic kidney disease. *Am J Physiol Renal Physiol*. 2013;305(10):F1391-F1401. doi:1152/ajprenal.00318.2013
46. Moreth K, Brodbeck R, Babelova A, Gretz N, Spieker T, Zeng-Brouwers J, Pfeilschifter J, Young MF, Schaefer RM, Schaefer L. The proteoglycan biglycan regulates expression of the B cell chemoattractant CXCL13 and aggravates murine lupus nephritis. *J Clin Invest*. 2010;120(12):4251-4272. doi:1172/JCI42213
47. Halvorsen B, Santilli F, Scholz H, Sahraoui A, Gulseth HL, Wium C, Lattanzio S, Formoso G, Di Fulvio P, Otterdal K, et al. LIGHT/TNFSF14 is increased in patients with type 2 diabetes mellitus and promotes islet cell dysfunction and endothelial cell inflammation in vitro. *Diabetologia*. 2016;59(10):2134-2144. doi:1007/s00125-016-4036-y
48. Glawe JD, Patrick DR, Huang M, Sharp CD, Barlow SC, Kevil CG. Genetic deficiency of Itgb2 or ItgaL prevents autoimmune diabetes through distinctly different mechanisms in NOD/LtJ mice. *Diabetes*. 2009;58(6):1292-1301. doi:2337/db08-0804
49. Blue EK, Sheehan BM, Nuss ZV, Boyle FA, Hocutt CM, Gohn CR, Varberg KM, McClintick JN, Haneline LS. Epigenetic Regulation of Placenta-Specific 8 Contributes to Altered Function of Endothelial Colony-Forming Cells Exposed to Intrauterine Gestational Diabetes Mellitus. *Diabetes*. 2015;64(7):2664-2675. doi:2337/db14-1709
50. Totomoch-Serra A, Muñoz ML, Burgueño J, Revilla-Monsalve MC, Perez-Muñoz A, Diaz-Badillo Á. The ADRA2A rs553668 variant is associated with type 2 diabetes and five variants were associated at nominal significance levels in a population-based case-control study from Mexico City. *Gene*. 2018;669:28-34. doi:1016/j.gene.2018.05.078
51. Gonzalez Badillo FE, Zisi Tegou F, Abreu MM, Masina R, Sha D, Najjar M, Wright SH, Bayer AL, Korpos É, Pugliese A, et al. CCL21 Expression in β -Cells Induces Antigen-Expressing Stromal Cell Networks in the Pancreas and Prevents Autoimmune Diabetes in Mice. *Diabetes*. 2019;68(10):1990-2003. doi:2337/db19-0239
52. Nejatian N, Häfner AK, Shoghi F, Badenhop K, Penna-Martinez M. 5-Lipoxygenase (ALOX5): Genetic susceptibility to type 2 diabetes and vitamin D effects on monocytes. *J Steroid Biochem Mol Biol*. 2019;187:52-57. doi:1016/j.jsbmb.2018.10.022

53. de Luis DA, Izaola O, Primo D, de la Fuente B, Aller R. Polymorphism rs3123554 in the cannabinoid receptor gene type 2 (CNR2) reveals effects on body weight and insulin resistance in obese subjects. *Endocrinol Diabetes Nutr.* 2017;64(8):440-445. doi:1016/j.endinu.2017.06.001
54. Tamagno G, Fedtke K, Eidenmüller M, Geks J, Hamann A, Langer K, Kann PH. The polymorphism of type 1 collagen (COL1A1) gene does not correlate with an increased risk of foot ulcers in patients with diabetes mellitus. *Exp Clin Endocrinol Diabetes.* 2015;123(4):240-245. doi:1055/s-0034-1395582
55. Wang W, Yan X, Lin Y, Ge H, Tan Q. Wnt7a promotes wound healing by regulation of angiogenesis and inflammation: Issues on diabetes and obesity. *J Dermatol Sci.* 2018;S0923-1811(18)30103-8. doi:1016/j.jdermsci.2018.02.007
56. Tabassum R, Mahajan A, Dwivedi OP, Chauhan G, Spurgeon CJ, Kumar MV, Ghosh S, Madhu SV, Mathur SK, Chandak GR, et al. Common variants of SLAMF1 and ITLN1 on 1q21 are associated with type 2 diabetes in Indian population. *J Hum Genet.* 2012;57(3):184-190. doi:1038/jhg.2011.150
57. Aparicio JM, Wakisaka A, Takada A, Matsuura N, Yoshiki T. Non-HLA genetic factors and insulin dependent diabetes mellitus in the Japanese: TCRA, TCRB and TCRG, INS, THY1, CD3D and ETS1. *Dis Markers.* 1990;8(5):283-294.
58. Mohamed WA, Schaalán MF. Antidiabetic efficacy of lactoferrin in type 2 diabetic pediatrics; controlling impact on PPAR- γ , SIRT-1, and TLR4 downstream signaling pathway. *Diabetol Metab Syndr.* 2018;10:89. doi:1186/s13098-018-0390-x
59. Li H, Liu J, Wang Y, Fu Z, Hüttemann M, Monks TJ, Chen AF, Wang JM. MiR-27b augments bone marrow progenitor cell survival via suppressing the mitochondrial apoptotic pathway in Type 2 diabetes. *Am J Physiol Endocrinol Metab.* 2017;313(4):E391-E401. doi:1152/ajpendo.00073.2017
60. Kim YI, Lee FN, Choi WS, Lee S, Youn JH. Insulin regulation of skeletal muscle PDK4 mRNA expression is impaired in acute insulin-resistant states. *Diabetes.* 2006;55(8):2311-2317. doi:2337/db05-1606
61. Alarslan P, Unal Kocabas G, Demir I, Guler A, Bozkaya G, Aslanipour B, Calan M. Increased urocortin 3 levels are associated with the risk of having type 2 diabetes mellitus. *J Diabetes.* 2020;12(6):474-482. doi:1111/1753-0407.13020
62. Rees SD, Britten AC, Bellary S, O'Hare JP, Kumar S, Barnett AH, Kelly MA. The promoter polymorphism -232C/G of the PCK1 gene is associated with type 2 diabetes in a UK-resident South Asian population. *BMC Med Genet.* 2009;10:83. doi:1186/1471-2350-10-83
63. Torsvik J, Johansson BB, Dalva M, Marie M, Fjeld K, Johansson S, Bjørkøy G, Saraste J, Njølstad PR, Molven A. Endocytosis of secreted carboxyl ester lipase in a syndrome of diabetes and pancreatic exocrine dysfunction. *J Biol Chem.* 2014;289(42):29097-29111. doi:1074/jbc.M114.574244
64. Bouras H, Roig SR, Kurstjens S, Tack CJJ, Kebieche M, de Baaij JHF, Hoenderop JGJ. Metformin regulates TRPM6, a potential explanation for magnesium imbalance in type 2 diabetes patients. *Can J Physiol Pharmacol.* 2020;98(6):400-411. doi:1139/cjpp-2019-0570

65. Lally S, Tan CY, Owens D, Tomkin GH. Messenger RNA levels of genes involved in dysregulation of postprandial lipoproteins in type 2 diabetes: the role of Niemann-Pick C1-like 1, ATP-binding cassette, transporters G5 and G8, and of microsomal triglyceride transfer protein. *Diabetologia*. 2006;49(5):1008-1016. doi:1007/s00125-006-0177-8
66. Dawed AY, Donnelly L, Tavendale R, Carr F, Leese G, Palmer CN, Pearson ER, Zhou K. CYP2C8 and SLC01B1 Variants and Therapeutic Response to Thiazolidinediones in Patients With Type 2 Diabetes. *Diabetes Care*. 2016;39(11):1902-1908. doi:2337/dc15-2464
67. Jamwal R, de la Monte SM, Ogasawara K, Adusumalli S, Barlock BB, Akhlaghi F. Nonalcoholic Fatty Liver Disease and Diabetes Are Associated with Decreased CYP3A4 Protein Expression and Activity in Human Liver. *Mol Pharm*. 2018;15(7):2621-2632. doi:1021/acs.molpharmaceut.8b00159
68. Zhang L, Wang YN, Ju JM, Shabanova A, Li Y, Fang RN, Sun JB, Guo YY, Jin TZ, Liu YY, et al. Mzb1 protects against myocardial infarction injury in mice via modulating mitochondrial function and alleviating inflammation. *Acta Pharmacol Sin*. 2020;10.1038/s41401-020-0489-0. doi:1038/s41401-020-0489-0
69. Ellenbroek GHJM, de Haan JJ, van Klarenbosch BR, Brans MAD, van de Weg SM, Smeets MB, de Jong S, Arslan F, Timmers L, Goumans MTH, et al. Leukocyte-Associated Immunoglobulin-like Receptor-1 is regulated in human myocardial infarction but its absence does not affect infarct size in mice. *Sci Rep*. 2017;7(1):18039. doi:1038/s41598-017-13678-5
70. Guo X, Chen Y, Lu Y, Li P, Yu H, Diao FR, Tang WD, Hou P, Zhao XX, et al. High level of circulating microRNA-142 is associated with acute myocardial infarction and reduced survival. *Ir J Med Sci*. 2020;189(3):933-937. doi:1007/s11845-020-02196-5
71. Tillmanns J, Hoffmann D, Habbaba Y, Schmitto JD, Sedding D, Fraccarollo D, Galuppo P, Bauersachs J. Fibroblast activation protein alpha expression identifies activated fibroblasts after myocardial infarction. *J Mol Cell Cardiol*. 2015;87:194-203. doi:1016/j.yjmcc.2015.08.016
72. Cavallari JF, Fullerton MD, Duggan BM, Foley KP, Denou E, Smith BK, Desjardins EM, Henriksbo BD, Kim KJ, Tuinema BR, et al. Muramyl Dipeptide-Based Postbiotics Mitigate Obesity-Induced Insulin Resistance via IRF4. *Cell Metab*. 2017;25(5):1063-1074.e3. doi:1016/j.cmet.2017.03.021
73. Kosugi T, Yuzawa Y, Sato W, Arata-Kawai H, Suzuki N, Kato N, Matsuo S, Kadomatsu K. Midkine is involved in tubulointerstitial inflammation associated with diabetic nephropathy. *Lab Invest*. 2007;87(9):903-913. doi:1038/labinvest.3700599
74. Gale JD, Gilbert S, Blumenthal S, Elliott T, Pergola PE, Goteti K, Scheele W, Perros-Huguet C. Effect of PF-04634817, an Oral CCR2/5 Chemokine Receptor Antagonist, on Albuminuria in Adults with Overt Diabetic Nephropathy. *Kidney Int Rep*. 2018;3(6):1316-1327. doi:1016/j.ekir.2018.07.010
75. Kelly KJ, Zhang J, Han L, Wang M, Zhang S, Dominguez JH. Intravenous renal cell transplantation with SAA1-positive cells prevents the progression of chronic renal failure in rats with ischemic-diabetic nephropathy. *Am J Physiol Renal Physiol*. 2013;305(12):F1804-F1812. doi:1152/ajprenal.00097.2013

76. Tang S, Wang X, Deng T, Ge H, Xiao X. Identification of C3 as a therapeutic target for diabetic nephropathy by bioinformatics analysis. *Sci Rep.* 2020;10(1):13468. doi:1038/s41598-020-70540-x
77. Li T, Yu Z, Qu Z, Zhang N, Crew R, Jiang Y. Decreased number of CD19+CD24hiCD38hi regulatory B cells in Diabetic nephropathy. *Mol Immunol.* 2019;112:233-239. doi:1016/j.molimm.2019.05.014
78. Yahya MJ, Ismail PB, Nordin NB, Akim ABM, Yusuf WSBM, Adam NLB, Yusoff MJ. Association of CCL2, CCR5, ELM01, and IL8 Polymorphism with Diabetic Nephropathy in Malaysian Type 2 Diabetic Patients. *Int J Chronic Dis.* 2019;2019:2053015. doi:1155/2019/2053015
79. Li MX, Zhao YF, Qiao HX, Zhang YP, Li XJ, Ren WD, Yu P. CXCR3 knockdown protects against high glucose-induced podocyte apoptosis and inflammatory cytokine production at the onset of diabetic nephropathy. *Int J Clin Exp Pathol.* 2017;10(8):8829-8838.
80. Ni X, Gu Y, Yu H, Wang S, Chen Y, Wang X, Yuan X, Jia W. Serum Adipocyte Fatty Acid-Binding Protein 4 Levels Are Independently Associated with Radioisotope Glomerular Filtration Rate in Type 2 Diabetic Patients with Early Diabetic Nephropathy. *Biomed Res Int.* 2018;2018:4578140. doi:1155/2018/4578140
81. Carlsson AC, Nowak C, Lind L, Östgren CJ, Nyström FH, Sundström J, Carrero JJ, Riserus U, Ingelsson E, Fall T, et al. Growth differentiation factor 15 (GDF-15) is a potential biomarker of both diabetic kidney disease and future cardiovascular events in cohorts of individuals with type 2 diabetes: a proteomics approach. *Ups J Med Sci.* 2020;125(1):37-43. doi:1080/03009734.2019.1696430
82. Jing F, Zhao J, Jing X, Lei G. Long noncoding RNA Airn protects podocytes from diabetic nephropathy lesions via binding to Igf2bp2 and facilitating translation of Igf2 and Lamb2. *Cell Biol Int.* 2020;44(9):1860-1869. doi:1002/cbin.11392
83. Gu T, Falhammar H, Gu HF, Brismar K. Epigenetic analyses of the insulin-like growth factor binding protein 1 gene in type 1 diabetes and diabetic nephropathy. *Clin Epigenetics.* 2014;6(1):10. doi:1186/1868-7083-6-10
84. Senthilkumar GP, Anithalekshmi MS, Yasir M, Parameswaran S, Packirisamy RM, Bobby Z. Role of omentin 1 and IL-6 in type 2 diabetes mellitus patients with diabetic nephropathy. *Diabetes Metab Syndr.* 2018;12(1):23-26. doi:1016/j.dsx.2017.08.005
85. Chen YG, Cieccko AE, Khaja S, Grzybowski M, Geurts AM, Lieberman SM. UBASH3A deficiency accelerates type 1 diabetes development and enhances salivary gland inflammation in NOD mice. *Sci Rep.* 2020;10(1):12019. doi:1038/s41598-020-68956-6
86. Sinha S, Renavikar PS, Crawford MP, Steward-Tharp SM, Brate A, Tsalikian E, Tansey M, Shivapour ET, Cho T, Kamholz J, et al. Altered expression of SIRPy on the T-cells of relapsing remitting multiple sclerosis and type 1 diabetes patients could potentiate effector responses from T-cells. *PLoS One.* 2020;15(8):e0238070. doi:1371/journal.pone.0238070
87. Burren OS, Guo H, Wallace C. VSEAMS: a pipeline for variant set enrichment analysis using summary GWAS data identifies IKZF3, BATF and ESRRA as key transcription factors in type 1 diabetes. *Bioinformatics.* 2014;30(23):3342-3348. doi:1093/bioinformatics/btu571

88. Hussain S, Wagner M, Ly D, Delovitch TL. Role of regulatory invariant CD1d-restricted natural killer T-cells in protection against type 1 diabetes. *Immunol Res.* 2005;31(3):177-188. doi:1385/IR:31:3:177
89. Fraser HI, Howlett S, Clark J, Rainbow DB, Stanford SM, Wu DJ, Hsieh YW, Maine CJ, Christensen M, Kuchroo V, et al. Ptpn22 and Cd2 Variations Are Associated with Altered Protein Expression and Susceptibility to Type 1 Diabetes in Nonobese Diabetic Mice. *J Immunol.* 2015;195(10):4841-4852. doi:4049/jimmunol.1402654
90. Ramos-Lopez E, Ghebru S, Van Autreve J, Aminkeng F, Herwig J, Seifried E, Seidl C, Van der Auwera B, Badenhoop K. Neither an intronic CA repeat within the CD48 gene nor the HERV-K18 polymorphisms are associated with type 1 diabetes. *Tissue Antigens.* 2006;68(2):147-152. doi:1111/j.1399-0039.2006.00637.x
91. Holmberg D, Ruikka K, Lindgren P, Eliasson M, Mayans S. Association of CD247 (CD3 ζ) gene polymorphisms with T1D and AITD in the population of northern Sweden. *BMC Med Genet.* 2016;17(1):70. doi:1186/s12881-016-0333-z
92. Hussein AG, Mohamed RH, Alghobashy AA. Synergism of CYP2R1 and CYP27B1 polymorphisms and susceptibility to type 1 diabetes in Egyptian children. *Cell Immunol.* 2012;279(1):42-45. doi:1016/j.cellimm.2012.08.006
93. Sun Z, Ma Y, Chen F, Wang S, Chen B, Shi J. miR-133b and miR-199b knockdown attenuate TGF- β 1-induced epithelial to mesenchymal transition and renal fibrosis by targeting SIRT1 in diabetic nephropathy. *Eur J Pharmacol.* 2018;837:96-104. doi:1016/j.ejphar.2018.08.022
94. Eftekhari A, Vahed SZ, Kavetsky T, Rameshrad M, Jafari S, Chodari L, Hosseiniyan SM, Derakhshankhah H, Ahmadian E, Ardalan M. Cell junction proteins: Crossing the glomerular filtration barrier in diabetic nephropathy. *Int J Biol Macromol.* 2020;148:475-482. doi:1016/j.ijbiomac.2020.01.168
95. Wang J, Gao Y, Ma M, Li M, Zou D, Yang J, Zhu Z, Zhao X. Effect of miR-21 on renal fibrosis by regulating MMP-9 and TIMP1 in kk-ay diabetic nephropathy mice. *Cell Biochem Biophys.* 2013;67(2):537-546. doi:1007/s12013-013-9539-2
96. Langham RG, Kelly DJ, Gow RM, Zhang Y, Cox AJ, Qi W, Thai K, Pollock CA, Christensen PK, Parving HH, et al. Increased renal gene transcription of protein kinase C-beta in human diabetic nephropathy: relationship to long-term glycaemic control. *Diabetologia.* 2008;51(4):668-674. doi:1007/s00125-008-0927-x
97. Ban CR, Twigg SM, Franjic B, Brooks BA, Celermajer D, Yue DK, McLennan SV. Serum MMP-7 is increased in diabetic renal disease and diabetic diastolic dysfunction. *Diabetes Res Clin Pract.* 2010;87(3):335-341. doi:1016/j.diabres.2010.01.004
98. McKay GJ, Kavanagh DH, Crean JK, Maxwell AP. Bioinformatic Evaluation of Transcriptional Regulation of WNT Pathway Genes with reference to Diabetic Nephropathy. *J Diabetes Res.* 2016;2016:7684038. doi:1155/2016/7684038
99. Zhong JM, Lu YC, Zhang J. Dexmedetomidine Reduces Diabetic Neuropathy Pain in Rats through the Wnt 10a/ β -Catenin Signaling Pathway. *Biomed Res Int.* 2018;2018:9043628.

100. Sheng J, Li H, Dai Q, Lu C, Xu M, Zhang J, Feng J. DUSP1 recuses diabetic nephropathy via repressing JNK-Mff-mitochondrial fission pathways. *J Cell Physiol.* 2019;234(3):3043-3057. doi:1002/jcp.27124
101. Abe H, Sakurai A, Ono H, Hayashi S, Yoshimoto S, Ochi A, Ueda S, Nishimura K, Shibata E, Tamaki M, et al. Urinary Exosomal mRNA of WT1 as Diagnostic and Prognostic Biomarker for Diabetic Nephropathy. *J Med Invest.* 2018;65(3.4):208-215. doi:2152/jmi.65.208
102. Ng MC, Baum L, So WY, Lam VK, Wang Y, Poon E, Tomlinson B, Cheng S, Lindpaintner K, Chan JC. Association of lipoprotein lipase S447X, apolipoprotein E exon 4, and apoC3 -455T>C polymorphisms on the susceptibility to diabetic nephropathy. *Clin Genet.* 2006;70(1):20-28. doi:1111/j.1399-0004.2006.00628.x
103. Asgarbeik S, Mohammad Amoli M, Enayati S, Bandarian F, Nasli-Esfahani E, Forouzanfar K, Razi F, Angaji SA. The Role of ERFF1+808T/G Polymorphism in Diabetic Nephropathy. *Int J Mol Cell Med.* 2019;8(Suppl1):49-55. doi:22088/IJMCM.BUMS.8.2.49
104. Tsantoulas C, Laínez S, Wong S, Mehta I, Vilar B, McNaughton PA. Hyperpolarization-activated cyclic nucleotide-gated 2 (HCN2) ion channels drive pain in mouse models of diabetic neuropathy. *Sci Transl Med.* 2017;9(409):eaam6072. doi:1126/scitranslmed.aam6072
105. Zhang D, Gu T, Forsberg E, Efendic S, Brismar K, Gu HF. Genetic and functional effects of membrane metalloendopeptidase on diabetic nephropathy development. *Am J Nephrol.* 2011;34(5):483-490. doi:1159/000333006
106. Chen CH, Lin KD, Ke LY, Liang CJ, Kuo WC, Lee MY, Lee YL, Hsiao PJ, Hsu CC, Shin SJ. O-GlcNAcylation disrupts STRA6-retinol signals in kidneys of diabetes. *Biochim Biophys Acta Gen Subj.* 2019;1863(6):1059-1069. doi:1016/j.bbagen.2019.03.014
107. De la Cruz-Cano E, Jiménez-González CDC, Morales-García V, Pineda-Pérez C, Tejas-Juárez JG, Rendón-Gandarilla FJ, Jiménez-Morales S, Díaz-Gandarilla JA. Arg913Gln variation of SLC12A3 gene is associated with diabetic nephropathy in type 2 diabetes and Gitelman syndrome: a systematic review. *BMC Nephrol.* 2019;20(1):393. doi:1186/s12882-019-1590-9
108. Fawzy MS, Abu AlSel BT. Assessment of Vitamin D-Binding Protein and Early Prediction of Nephropathy in Type 2 Saudi Diabetic Patients. *J Diabetes Res.* 2018;2018:8517929. doi:1155/2018/8517929
109. Mathews JA, Wurmbrand AP, Ribeiro L, Neto FL, Shore SA. Induction of IL-17A Precedes Development of Airway Hyperresponsiveness during Diet-Induced Obesity and Correlates with Complement Factor D. *Front Immunol.* 2014;5:440. doi:3389/fimmu.2014.00440
110. Guo X, Li F, Xu Z, Yin A, Yin H, Li C, Chen SY. DOCK2 deficiency mitigates HFD-induced obesity by reducing adipose tissue inflammation and increasing energy expenditure. *J Lipid Res.* 2017;58(9):1777-1784. doi:1194/jlr.M073049
111. Moreno-Navarrete JM, Latorre J, Lluch A, Ortega FJ, Comas F, Arnoriaga-Rodríguez M, Ricart W, Fernández-Real JM. Lysozyme is a component of the innate immune system linked to obesity

- associated-chronic low-grade inflammation and altered glucose tolerance. *Clin Nutr.* 2020;S0261-5614(20)30452-0. doi:1016/j.clnu.2020.08.036
112. Wang L, Wang Y, Zhang C, Li J, Meng Y, Dou M, Noguchi CT, Di L. Inhibiting Glycogen Synthase Kinase 3 Reverses Obesity-Induced White Adipose Tissue Inflammation by Regulating Apoptosis Inhibitor of Macrophage/CD5L-Mediated Macrophage Migration. *Arterioscler Thromb Vasc Biol.* 2018;38(9):2103-2116. doi:1161/ATVBAHA.118.311363
 113. Lee H, Lee YJ, Choi H, Seok JW, Yoon BK, Kim D, Han JY, Lee Y, Kim HJ, Kim JW. SCARA5 plays a critical role in the commitment of mesenchymal stem cells to adipogenesis. *Sci Rep.* 2017;7(1):14833. doi:1038/s41598-017-12512-2
 114. Han CY, Kang I, Harten IA, Gebe JA, Chan CK, Omer M, Alonge KM, den Hartigh LJ, Gomes Kjerulf D, Goodspeed L, et al. Adipocyte-Derived Versican and Macrophage-Derived Biglycan Control Adipose Tissue Inflammation in Obesity. *Cell Rep.* 2020;31(13):107818. doi:1016/j.celrep.2020.107818
 115. Zhang W, Wu X, Pei Z, Kiess W, Yang Y, Xu Y, Chang Z, Wu J, Sun C, Luo F. GDF5 Promotes White Adipose Tissue Thermogenesis via p38 MAPK Signaling Pathway. *DNA Cell Biol.* 2019;38(11):1303-1312. doi:1089/dna.2019.4724
 116. Crowley RK, O'Reilly MW, Bujalska IJ, Hassan-Smith ZK, Hazlehurst JM, Foucault DR, Stewart PM, Tomlinson JW. SFRP2 Is Associated with Increased Adiposity and VEGF Expression. *PLoS One.* 2016;11(9):e0163777. doi:1371/journal.pone.0163777
 117. Gan M, Shen L, Wang S, Guo Z, Zheng T, Tan Y, Fan Y, Liu L, Chen L, Jiang A, et al. Genistein inhibits high fat diet-induced obesity through miR-222 by targeting BTG2 and adipor1. *Food Funct.* 2020;11(3):2418-2426. doi:1039/c9fo00861f
 118. Caracciolo V, Young J, Gonzales D, Ni Y, Flowers SJ, Summer R, Waldman SA, Kim JK, Jung DY, Noh HL, et al. Myeloid-specific deletion of Zfp36 protects against insulin resistance and fatty liver in diet-induced obese mice. *Am J Physiol Endocrinol Metab.* 2018;315(4):E676-E693. doi:1152/ajpendo.00224.2017
 119. Godlewski G, Jourdan T, Szanda G, Tam J, Cinar R, Harvey-White J, Liu J, Mukhopadhyay B, Pacher P, Ming Mo F, et al. Mice lacking GPR3 receptors display late-onset obese phenotype due to impaired thermogenic function in brown adipose tissue. *Sci Rep.* 2015;5:14953. doi:1038/srep14953
 120. Khaidakov M, Mitra S, Kang BY, Wang X, Kadlubar S, Novelli G, Raj V, Winters M, Carter WC, Mehta JL. Oxidized LDL receptor 1 (OLR1) as a possible link between obesity, dyslipidemia and cancer. *PLoS One.* 2011;6(5):e20277. doi:1371/journal.pone.0020277
 121. Benson KK, Hu W, Weller AH, Bennett AH, Chen ER, Khetarpal SA, Yoshino S, Bone WP, Wang L, Rabinowitz JD, et al. Natural human genetic variation determines basal and inducible expression of PM20D1, an obesity-associated gene. *Proc Natl Acad Sci U S A.* 2019;116(46):23232-23242. doi:1073/pnas.1913199116
 122. Lloret-Linares C, Miyauchi E, Luo H, Labat L, Bouillot JL, Poitou C, Oppert JM, Laplanche JL, Mouly S, Scherrmann JM, et al. Oral Morphine Pharmacokinetic in Obesity: The Role of P-Glycoprotein, MRP2,

- MRP3, UGT2B7, and CYP3A4 Jejunal Contents and Obesity-Associated Biomarkers. *Mol Pharm.* 2016;13(3):766-773. doi:1021/acs.molpharmaceut.5b0065
123. Zhang H, He Y, He X, Wang L, Jin T, Yuan D. Three SNPs of FCRL3 and one SNP of MTMR3 are associated with immunoglobulin A nephropathy risk. *Immunobiology.* 2020;225(1):151869. doi:1016/j.imbio.2019.11.004
124. Zhou XJ, Cheng FJ, Qi YY, Zhao YF, Hou P, Zhu L, Lv JC, Zhang H. FCGR2B and FCRLB gene polymorphisms associated with IgA nephropathy. *PLoS One.* 2013;8(4):e61208. doi:1371/journal.pone.0061208
125. Kosacka J, Nowicki M, Klötting N, Kern M, Stumvoll M, Bechmann I, Serke H, Blüher M. COMP-angiopoietin-1 recovers molecular biomarkers of neuropathy and improves vascularisation in sciatic nerve of ob/ob mice. *PLoS One.* 2012;7(3):e32881. doi:1371/journal.pone.0032881
126. Hanudel MR, Rappaport M, Chua K, Gabayan V, Qiao B, Jung G, Salusky IB, Ganz T, Nemeth E. Levels of the erythropoietin-responsive hormone erythroferrone in mice and humans with chronic kidney disease. *Haematologica.* 2018;103(4):e141-e142. doi:3324/haematol.2017.181743
127. Bonomo JA, Ng MC, Palmer ND, Keaton JM, Larsen CP, Hicks PJ; T2D-GENES Consortium, Langefeld CD, Freedman BI, Bowden DW. Coding variants in nephrin (NPHS1) and susceptibility to nephropathy in African Americans. *Clin J Am Soc Nephrol.* 2014;9(8):1434-1440. doi:2215/CJN.00290114
128. Zou J, Liu KC, Wang WP, Xu Y. Circular RNA COL1A2 promotes angiogenesis via regulating miR-29b/VEGF axis in diabetic retinopathy. *Life Sci.* 2020;256:117888. doi:1016/j.lfs.2020.117888
129. Sergeys J, Van Hove I, Hu TT, Temps C, Carragher NO, Unciti-Broceta A, Feyen JHM, Moons L, Porcu M. The retinal tyrosine kinome of diabetic Akimba mice highlights potential for specific Src family kinase inhibition in retinal vascular disease. *Exp Eye Res.* 2020;197:108108. doi:1016/j.exer.2020.108108
130. Wang H, Lou H, Li Y, Ji F, Chen W, Lu Q, Xu G. Elevated vitreous Lipocalin-2 levels of patients with proliferative diabetic retinopathy. *BMC Ophthalmol.* 2020;20(1):260. doi:1186/s12886-020-01462-5
131. Ankit BS, Mathur G, Agrawal RP, Mathur KC. Stronger relationship of serum apolipoprotein A-1 and B with diabetic retinopathy than traditional lipids. *Indian J Endocrinol Metab.* 2017;21(1):102-105. doi:4103/2230-8210.196030
132. Samokhin AO, Stephens T, Wertheim BM, Wang RS, Vargas SO, Yung LM, Cao M, Brown M, Arons E, Dieffenbach PB, et al. NEDD9 targets COL3A1 to promote endothelial fibrosis and pulmonary arterial hypertension. *Sci Transl Med.* 2018;10(445):eaap7294. doi:1126/scitranslmed.aap7294
133. Douma LG, Solocinski K, Holzworth MR, Crislip GR, Masten SH, Miller AH, Cheng KY, Lynch IJ, Cain BD, Wingo CS, et al. Female C57BL/6J mice lacking the circadian clock protein PER1 are protected from nondipping hypertension. *Am J Physiol Regul Integr Comp Physiol.* 2019;316(1):R50-R58. doi:1152/ajpregu.00381.2017
134. Altura BM, Kostellow AB, Zhang A, Li W, Morrill GA, Gupta RK, Altura BT. Expression of the nuclear factor-kappaB and proto-oncogenes c-fos and c-jun are induced by low extracellular Mg²⁺ in aortic

- and cerebral vascular smooth muscle cells: possible links to hypertension, atherogenesis, and stroke. *Am J Hypertens*. 2003;16(9 Pt 1):701-707. doi:10.1016/s0895-7061(03)00987-7
135. Kim BG, Yoo TH, Yoo JE, Seo YJ, Jung J, Choi JY. Resistance to hypertension and high Cl⁻ excretion in humans with SLC26A4 mutations. *Clin Genet*. 2017;91(3):448-452. doi:10.1111/cge.12789
 136. Gao BF, Shen ZC, Bian WS, Wu SX, Kang ZX, Gao Y. Correlation of hypertension and F2RL3 gene methylation with Prognosis of coronary heart disease. *J Biol Regul Homeost Agents*. 2018;32(6):1539-1544.
 137. Williams JS, Hopkins PN, Jeunemaitre X, Brown NJ. CYP4A11 T8590C polymorphism, salt-sensitive hypertension, and renal blood flow. *J Hypertens*. 2011;29(10):1913-1918. doi:10.1097/HJH.0b013e32834aa786
 138. Geng H, Li B, Wang Y, Wang L. Association Between the CYP4F2 Gene rs1558139 and rs2108622 Polymorphisms and Hypertension: A Meta-Analysis. *Genet Test Mol Biomarkers*. 2019;23(5):342-347. doi:10.89/gtmb.2018.0202
 139. Onions KL, Gamez M, Buckner NR, Baker SL, Betteridge KB, Desideri S, Dallyn BP, Ramnath RD, Neal CR, Farmer LK, et al. VEGFC Reduces Glomerular Albumin Permeability and Protects Against Alterations in VEGF Receptor Expression in Diabetic Nephropathy. *Diabetes*. 2019;68(1):172-187. doi:10.2337/db18-0045
 140. Rafatian G, Kamkar M, Parent S, Michie C, Risha Y, Molgat ASD, Seymour R, Suuronen EJ, Davis DR. Mybl2 rejuvenates heart explant-derived cells from aged donors after myocardial infarction. *Aging Cell*. 2020;19(7):e13174. doi:10.1111/acer.13174
 141. Cengiz M, Karatas OF, Koparir E, Yavuzer S, Ali C, Yavuzer H, Kirat E, Karter Y, et al. Differential expression of hypertension-associated microRNAs in the plasma of patients with white coat hypertension. *Medicine (Baltimore)*. 2015;94(13):e693. doi:10.1097/MD.0000000000000693
 142. Hogewind BF, Micheal S, Schoenmaker-Koller FE, Hoyng CB, den Hollander AI. Analyses of Sequence Variants in the MYOC Gene and of Single Nucleotide Polymorphisms in the NR3C1 and FKBP5 Genes in Corticosteroid-Induced Ocular Hypertension. *Ophthalmic Genet*. 2015;36(4):299-302. doi:10.3109/13816810.2013.879598
 143. Wang J, Wang G, Liang Y, Zhou X. Expression Profiling and Clinical Significance of Plasma MicroRNAs in Diabetic Nephropathy. *J Diabetes Res*. 2019;2019:5204394. doi:10.1155/2019/5204394
 144. Li Y, He XN, Li C, Gong L, Liu M. Identification of Candidate Genes and MicroRNAs for Acute Myocardial Infarction by Weighted Gene Coexpression Network Analysis. *Biomed Res Int*. 2019;2019:5742608. doi:10.1155/2019/5742608
 145. Keller MP, Paul PK, Rabaglia ME, Stapleton DS, Schueler KL, Broman AT, Ye SI, Leng N, Brandon CJ, Neto EC, et al. The Transcription Factor Nfatc2 Regulates β -Cell Proliferation and Genes Associated with Type 2 Diabetes in Mouse and Human Islets. *PLoS Genet*. 2016;12(12):e1006466. doi:10.1371/journal.pgen.1006466
 146. Fujimoto K, Chen Y, Polonsky KS, Dorn GW 2nd. Targeting cyclophilin D and the mitochondrial permeability transition enhances beta-cell survival and prevents diabetes in Pdx1 deficiency. *Proc*

147. Xu Y, Song R, Long W, Guo H, Shi W, Yuan S, Xu G, Zhang T. CREB1 functional polymorphisms modulating promoter transcriptional activity are associated with type 2 diabetes mellitus risk in Chinese population. *Gene*. 2018;665:133-140. doi:1016/j.gene.2018.05.002
148. Du L, Qian X, Li Y, Li XZ, He LL, Xu L, Liu YQ, Li CC, Ma P, Shu FL, et al. Sirt1 inhibits renal tubular cell epithelial-mesenchymal transition through YY1 deacetylation in diabetic nephropathy. *Acta Pharmacol Sin*. 2020;10.1038/s41401-020-0450-2. doi:1038/s41401-020-0450-2
149. Zhang SZ, Qiu XJ, Dong SS, et al. MicroRNA-770-5p is involved in the development of diabetic nephropathy through regulating podocyte apoptosis by targeting TP53 regulated inhibitor of apoptosis 1. *Eur Rev Med Pharmacol Sci*. 2019;23(3):1248-1256. doi:26355/eurev_201902_17018
150. Zhao L, Chi L, Zhao J, Wang X, Chen Z, Meng L, Liu G, Guan G, Wang F. Serum response factor provokes epithelial-mesenchymal transition in renal tubular epithelial cells of diabetic nephropathy. *Physiol Genomics*. 2016;48(8):580-588. doi:1152/physiolgenomics.00058.2016

Tables

Due to technical limitations, Tables 1-6 are only available as a download in the supplemental files section.

Figures

Volcano plot of differentially expressed genes. Genes with a significant change of more than two-fold were selected. Green dot represented up regulated significant genes and red dot represented down regulated significant genes.

Heat map of differentially expressed genes. Legend on the top left indicate log fold change of genes. (A1 – A28 = DN samples; B1 – B8 = normal control samples)

Figure 3

PPI network and the most significant modules of DEGs. (A) The PPI network of DEGs was constructed using Cytoscape (B) The most significant module was obtained from PPI network with 15 nodes and 36 edges for up regulated genes (C) The most significant module was obtained from PPI network with 7 nodes and 12 edges for up regulated genes. Up regulated genes are marked in green; down regulated genes are marked in red

Figure 4

(A) Target gene - miRNA regulatory network between target genes and miRNAs (B) Target gene - TF regulatory network between target genes and TFs. Up regulated genes are marked in green; down regulated genes are marked in red; The blue color diamond nodes represent the key miRNAs; the yellow color triangle nodes represent the key TFs

Figure 5

Figure 6

Structures of designed molecules

Figure 7

3D Binding of Molecule SALISO 2 and SALPYR 12 with 5UTZ & 5T5T

Figure 8

2D Binding of Molecule SALISO 2 and SALPYR 12 with 5UTZ & 5T5T

Supplementary Files

This is a list of supplementary files associated with this preprint. Click to download.

- [Tables.docx](#)