

CLINICAL STUDY



Mechanism of *Rhizoma Chuanxiong* for the treatment of diabetic kidney disease based on network pharmacology

Yuhe Yan^{a,b,c*}, Honghong Shi^{a,b,c*}, Yue Li^{a,b,c}, Xing Wan^{a,b,c}, Jinxin Li^{a,b,c} and Lihua Wang^{a,b,c} 

^aDivision of Nephrology, The Second Hospital of Shanxi Medical University, Taiyuan, China; ^bShanxi Kidney Disease Institute, Taiyuan, China; ^cInstitute of Nephrology, Shanxi Medical University, Taiyuan, China

ABSTRACT

Background: Although *Rhizoma Chuanxiong* has been used for the treatment of diabetic kidney disease (DKD), the relevant mechanisms remain unclear. The purpose of this study was to investigate the potential targets and mechanisms of *Rhizoma Chuanxiong* in treating DKD, utilizing network pharmacology.

Methods: Active compounds of *Rhizoma Chuanxiong* were obtained from the Traditional Chinese Medicine System Pharmacology Database and Analysis Platform database. SwissTargetPrediction was used to obtain the potential targets of active ingredients. DKD-associated targets were gathered from the GeneCards, DisGeNET, and OMIM databases. The STRING database and Cytoscape 3.7.2 were used for investigating core targets and interactions among targets. Gene Ontology and Kyoto Encyclopedia of Gene Genomes enrichment were performed using DAVID database. Molecular docking was performed using AutoDock-1.5.7 based on the crystal structures of the targets as deposited in the Research Collaboratory for Structural Bioinformatics (RCSB) Protein Data Bank.

Results: The top 10 core targets were identified, namely PPARG, AKT1, EGFR, STAT3, CASP3, PPARA, ICAM1, PTGS2, SRC, and MMP9. Enrichment analysis revealed that the primary pathways involving these targets including prolactin signaling pathway, AGE-RAGE signaling pathway in diabetic complications, TNF signaling pathway, relaxin signaling pathway, VEGF signaling pathway, and FoxO signaling pathway. Molecular docking demonstrated that mandenol exhibited a strong binding affinity toward EGFR domain, and wallichilide displayed pronounced binding affinity toward AKT1, EGFR, STAT3, and PTGS2 domains. Additionally, myricanone and senkyunone also showed strong binding affinity for AKT1, EGFR, CASP3, STAT3, and PTGS2 domains.

Conclusions: This study revealed the potential multi-component and multi-target mechanisms of *Rhizoma Chuanxiong* in treating DKD through network pharmacology. Supplementary experiments are required to further verify these findings.

ARTICLE HISTORY

Received 14 January 2025

Revised 20 March 2025

Accepted 18 June 2025

KEYWORDS

Rhizoma Chuanxiong;
DKD; network
pharmacology;
mechanism

Introduction

Diabetes, a group of metabolic diseases, has become a significant global public health concern and a leading cause of death and disability worldwide. According to data from the Global Burden of Disease, 529 million people worldwide were living with diabetes in 2021, and this number is expected to increase to 1.31 billion by 2050 [1]. Diabetic kidney disease (DKD) is one of the common microvascular complications in patients with diabetes, and the main cause of chronic kidney disease and end-stage renal disease worldwide, significantly increasing the risk of morbidity and mortality in patients with diabetes [2]. DKD is a complex disease process that involves several pathophysiologic mechanisms, such as metabolic dysregulation (hyperglycemia, insulin resistance, and dyslipidemia), hemodynamic change, oxidative stress,

inflammation, and fibrosis. Metabolic dysregulation leads to the activation of polyol pathway, advanced glycation end-products formation, protein kinase C activation, and hexosamine pathway flux, and these pathways contribute to oxidative stress, inflammation, and fibrosis, creating a vicious cycle [3,4]. Hyperglycemia-induced activation of the renin-angiotensin-aldosterone system (RAS) results in increased intraglomerular pressure and glomerular hyperfiltration, which leads to glomerular capillary hypertension, endothelial dysfunction, and podocyte injury [5]. Current treatments for DKD include RAAS inhibition, sodium-glucose cotransporter 2 inhibitors, glucagon-like peptide-1 receptor agonists, mineralocorticoid receptor antagonists [3]. These treatments are not universally applicable to patients across all stages of DKD. And they are focus on managing risk

CONTACT Lihua Wang  lihuawang236@126.com  Division of Nephrology, The Second Hospital of Shanxi Medical University, Taiyuan, China

*These authors contributed equally to this work and are co-first authors.

 Supplemental data for this article is available online at <https://doi.org/10.1080/0886022X.2025.2524528>

© 2025 The Author(s). Published by Informa UK Limited, trading as Taylor & Francis Group.

This is an Open Access article distributed under the terms of the Creative Commons Attribution-NonCommercial License (<http://creativecommons.org/licenses/by-nc/4.0/>), which permits unrestricted non-commercial use, distribution, and reproduction in any medium, provided the original work is properly cited. The terms on which this article has been published allow the posting of the Accepted Manuscript in a repository by the author(s) or with their consent.

factors, such as hypertension, hyperglycemia, and proteinuria, rather than directly targeting the underlying pathogenic mechanisms of DKD. The absence of targeted therapeutic strategies significantly restricts capacity to delay the progression of the disease. Therefore, identifying effective medications is critical in the treatment of DKD.

Traditional Chinese medicine has shown positive effects on the treatment of DKD. *Rhizoma Chuanxiong*, a well-known traditional Chinese medicine dried from rhizome of *Rhizoma chuanxiong* Hort, has been demonstrated to increase blood circulation, reduce inflammation, and alleviate pain [6]. Several studies have revealed that *Rhizoma Chuanxiong* exerts anti-thrombotic effects and prevent liver and kidney fibrosis. Furthermore, studies have indicated that *Rhizoma Chuanxiong* exhibits protective effects against acute renal injury [7–10]. Recent clinical and experimental studies suggested that the active ingredients of *Rhizoma Chuanxiong* may have a positive effect on DKD [11–13]. However, the related mechanism remains unclear.

Network pharmacology, an emerging interdisciplinary approach rooted in systems biology and network analysis of biological systems, employs advanced informatics technologies and biomedical databases to predict drug-disease interactions and elucidate underlying therapeutic mechanisms [14]. This study aimed to investigate the possible molecular mechanism underlying the effects of *Rhizoma Chuanxiong* on DKD using network pharmacology approach.

Materials and methods

Acquisition of active compounds and targets for *Rhizoma Chuanxiong* and therapeutic targets for DKD

The active components of *Rhizoma Chuanxiong* were systematically retrieved from the Traditional Chinese Medicine System Pharmacology Database and Analysis Platform (TCMSP) (<http://tcmsp.com>, Version 2.3) with screening criteria based on absorption, distribution, metabolism, excretion (ADME) [15,16]. The ADME parameters were achieved by satisfying the following standards: drug-likeness (DL) ≥ 0.18 , oral bioavailability (OB) $\geq 30\%$, hydrogen bond donors (Hdon) < 5 , and hydrogen bond acceptors (Hacc) < 10 [17,18]. Canonical SMILES obtained from PubChem database (<https://pubchem.ncbi.nlm.nih.gov/>) were imported into SwissTargetPrediction (<http://www.swisstargetprediction.ch/>) for potential targets prediction of *Rhizoma Chuanxiong* [19]. GeneCards (<https://www.genecards.org/>, version 5.23), DisGeNET (<https://www.disgenet.org/home/>, version 24.2), and OMIM (<https://omim.org/>) were utilized to collect DKD-associated targets, with all target data restricted to 'Homo sapiens'. In the Genecards database, the targets with a score greater than the median were identified as a potential target for DKD [20,21]. In the DisGeNET database, the targets with a score of gda > 0.1 were identified as potential targets for DKD [22,23]. The disease targets from OMIM database were obtained from established scientific studies and experimental results. Therefore, all the DKD-associated targets were included in the analysis.

Venny2.1 was utilized to identify and visualize the overlapping genes between active compounds of *Rhizoma Chuanxiong* and DKD.

Visualization of drug-disease target protein-protein interaction networks

To elucidate the interplay between *Rhizoma Chuanxiong* and DKD, we conducted protein-protein interaction (PPI) network analysis using overlapping genes identified between *Rhizoma Chuanxiong* compounds and DKD. The gene set was uploaded to the STRING database (<https://string-db.org/cgi/input.pl>, Version 12.0) for PPI network construction [24]. The software Cytoscape 3.10.1 was used to further visualize the protein-protein interaction (PPI) network. The further visualize of PPI network was performed using the CentiScaPe 2.2 plugin in Cytoscape 3.10.1. Key topological parameters, including closeness centrality, betweenness centrality, and degree centrality, were calculated for each node. Nodes were filtered based on the average values of these parameters to identify biologically significant hubs. Specifically, nodes with values above the average for closeness centrality, betweenness centrality, and degree centrality were identified as core targets for further analysis [25]. The size of the node is directly related to the degree of the core target, reflecting core target topological significance in the pharmacological-disease network [26].

Pathway analysis of *Rhizoma Chuanxiong* for DKD and the *Rhizoma Chuanxiong*-component-target signaling pathway network

To investigate the biological functions of potential targets in DKD, the David database (version 6.8) was utilized to collect information on Gene Ontology (GO) and Kyoto Encyclopedia of Gene Genomes (KEGG) analysis data, and GO enrichment analysis was carried out at three different levels: biological process (BP), molecular function (MF), cellular component (CC) [27]. The threshold was set at $p < 0.05$, FDR < 0.05 for GO enrichment and pathway screening. Considering the p -value reflects only the statistical significance of target enrichment in the pathway, not the biological correlation, we excluded the non-relevant pathways in our results. A dot plot was used to visually represent the findings of the KEGG pathway enrichment study. The gene ratio is illustrated on the horizontal axis, while the names of the enriched pathways are shown on the vertical axis. The color scale represents specific thresholds of the p -value, and the size of each dot correlates with the number of genes associated with each pathway. The dot plot and bar plot were constructed using the web-based platform for data analysis and visualization, <https://www.bioinformatics.com.cn> (last accessed on 20 Feb 2024) [28]. The KEGG pathway maps were generated using the KEGG database (<https://www.kegg.jp/kegg/>, Release 111.1, September 1, 2024). We utilized software Cytoscape 3.10.1 to construct and visualize the drug-compound-target-signaling pathway network.

Molecular docking and visualization of active ingredients and target proteins

The core targets in the PPI were ranked by degree value in descending order, and the top five core targets overlapping with those in the drug-component-target-signaling pathway network were selected for molecular docking. The molecular docking technology was executed using AutoDock (Version 1.5.6 Sep_17_14) based on the crystal structures of the targets obtained from the RCSB Protein Data Bank (<http://www.pdb.org/>). The primary objective gene was searched in the RCSB Protein Data Bank, and the selection criteria were confined to (homo species with X-ray resolution $\leq 2\text{\AA}$). The active components of *Rhizoma Chuanxiong* were analyzed using AutoDock Vina software (Version 1.5.6 Sep_17_14) to evaluate their binding energy with specific proteins. To ensure the reliability of the results, we conducted 50 times independent molecular docking experiments for the same ligand-receptor complex, and based on the principle of energy minimization selected the lowest binding energy value as our result. A binding energy threshold of lower than -5 kcal/mol indicated stable affinity between the components and proteins, the affinity between components and proteins improved. The components with the highest binding affinity were combined with proteins and visualized using PyMOL 3.1.

Table 1. Active components and ADME parameters of *Rhizoma Chuanxiong*.

Mol ID	Molecule name	MW	OB	DL	Hdon	Hacc
MOL002135	Myricanone	356.45	40.6	0.51	2	5
MOL001494	Mandenol	308.56	42	0.19	0	2
MOL002157	wallichilide	412.57	42.31	0.71	0	5
MOL002151	senkyunone	326.52	47.66	0.24	0	2

Results

Acquisition of active compounds and targets for *Rhizoma Chuanxiong* and therapeutic targets for DKD

The active components of *Rhizoma Chuanxiong* were screened based on ADME parameters from the TCMSP database, including Myricanone, Mandenol, wallichilide, and senkyunone (Table 1). After removing duplicates, a total of 248 potential compound-related targets were identified using the SwissTargetPrediction platform. There were 1780 therapeutic targets of DKD that were acquired from GeneCards, DisGeNET, and OMIM databases after deleting duplicates. The potential targets of *Rhizoma Chuanxiong* and DKD were imported into the Venny 2.1 online tool to generate a Venn diagram. Finally, 95 overlapping targets were identified for further analysis (Figure 1 and Table 2).

Visualization of drug-disease target protein-protein interaction networks

Figure 2 illustrates the PPI network of overlapping targets constructed using the STRING database. The subsequent analysis was conducted with Cytoscape3.10.1 software, and the network consisted of 95 nodes and 780 edges was constructed without any filtering. Following a screening process with the parameters: betweenness > 95.65263158 , closeness > 0.00536038 , and degree > 16.42105263 , we refined the network to 21 nodes and 157 edges (Figure 3). The 21 core targets were hierarchically ranked by degree value in descending order: PPARG, AKT1, EGFR, STAT3, CASP3, PPARA, ICAM1, PTGS2, SRC, MMP9, CCND1, SCARB1, MAPK14, ACE, NR3C1, KDR, MAPK8, APOB, CYP19A1, FASN, and SLC2A2. The size and color of each node in Figure 3 correspond to its degree (larger node and redder color indicate higher degree).

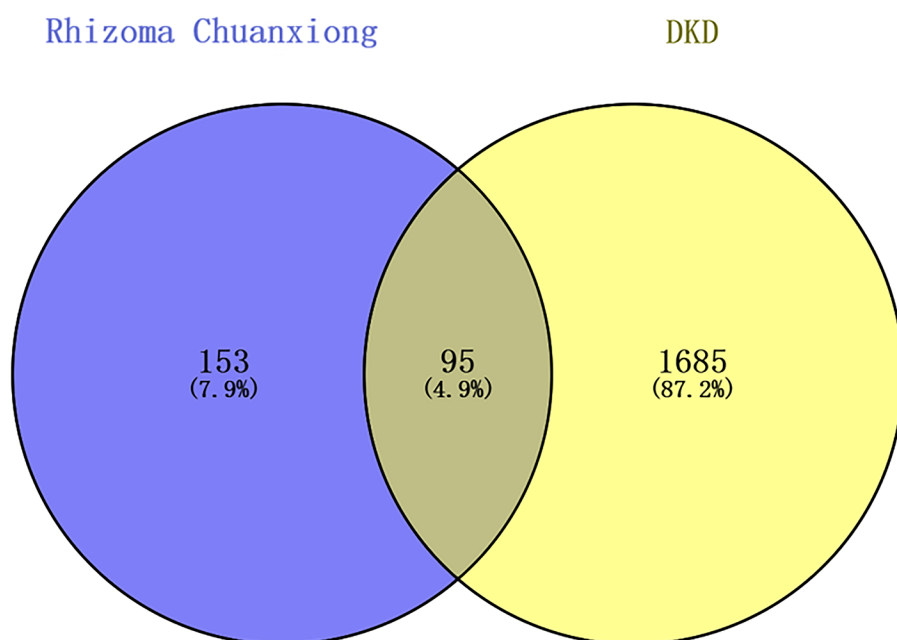


Figure 1. The overlapping targets between *Rhizoma Chuanxiong* and DKD.

Table 2. Overlapping genes between the DKD and *Rhizoma Chuanxiong*.

HSD11B1	CNR1	NR1H3	PTPN6	EPAS1	ICAM1	CPT1B
CFTR	FABP3	SCD	ROCK2	CYP24A1	SELE	PYGM
KCNH2	PTGS1	FABP1	ROCK1	CYP27B1	TGM2	EPHX2
ACE	CYP19A1	MTTP	F2R	CCR5	DRD2	HTR2A
FABP4	PTGS2	APOB	NLRP3	PARP1	HPGD5	PPIA
JAK1	PTPN1	ALOX5AP	CCND1	CPT1A	TRPV1	DRD3
SLC2A1	PPARG	AR	EGFR	CPT2	MMP9	MME
SLC5A1	HMGCR	CCNE1	CALCLRL	F10	KHK	SCARB1
NR3C2	PPARA	ACACB	NAMPT	CTSB	MAPK10	BDKRB2
FLT1	PPARD	AVPR2	C3AR1	SRC	PTK2	UTS2R
KDR	NR3C1	CASR	MDM2	SLC2A3	P2RX7	FASN
CASP3	KCNJ11	HSD11B2	SOAT1	MAPK14	KCNJ5	TEK
STAT3	SLC2A2	SYK	DKK1	MAPK8	AKT1	MTNR1B
PTK2B	CCR1	GLI2	IKKBK			

The DAVID database was used for enrichment analysis of 21 core targets, including the BP, MF, and CC of GO, as well as the KEGG pathway. The top 20 entries were selected based on the *p*-value. The results implied that the main BP involved in the treatment of *Rhizoma Chuanxiong* for DKD included response to UV-A, xenobiotic stimulus, cellular response to cadmium ion, signal transduction, and so on (Figure 4(a)). Regarding MF, the results indicated that the targets were predominantly enriched in enzyme binding, protein phosphatase binding, nuclear receptor activity, integrin binding, and transmembrane receptor protein tyrosine kinase activity, and so on (Figure 4(b)). The enrichment results of CC involved were mainly integral component of cytoplasm, membrane raft, receptor complex, caveola, plasma membrane, and so on (Figure 4(c)). The KEGG pathway was primarily enriched in the Prolactin signaling pathway, the AGE-RAGE signaling pathway in diabetic complications, the TNF signaling pathway, the Relaxin signaling pathway, the VEGF signaling

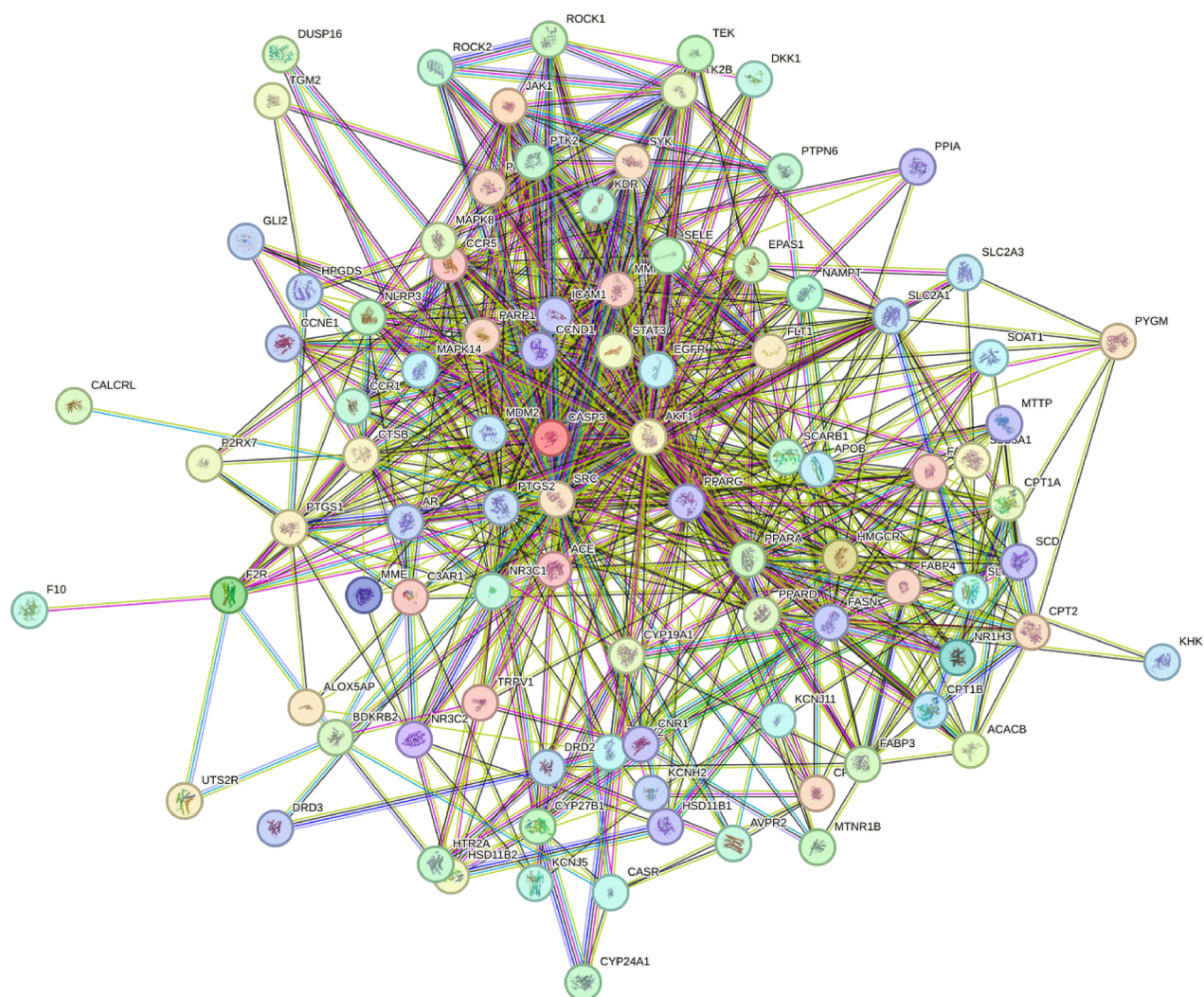


Figure 2. PPI network of overlapping targets.

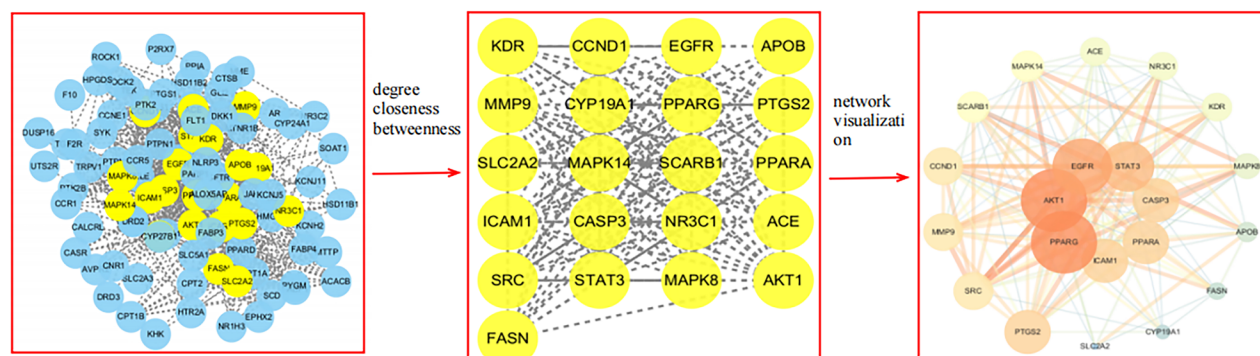


Figure 3. Screening and visualization of core targets.

Table 3. Degree value of core targets.

AKT1	PPARG	EGFR	STAT3	CASP3	PTGS2	PPARA
20	20	19	18	17	17	17
ICAM1	SRC	MMP9	CCND1	SCARB1	APK14	NR3C1
17	16	16	16	15	15	14
ACE	KDR	MAPK8	APOB	FASN	CYP19A1	SLC2A2
14	14	13	11	10	9	6

pathway, and the FoxO signaling pathway (Figure 5, Figures S1–S6, and Table S1).

Drug-component-target-signaling pathway network

A drug–component–target–signaling pathway network for the treatment of DKD with *Rhizoma Chuanxiong* was constructed (Figure 6). The integrative network revealed four principal bioactive active components with distinct pharmacological profiles. Mandenol acted on five targets (EGFR, PTGS2, CCND1, and KDR), regulating six key pathways, including the Prolactin signaling pathway, the AGE-RAGE signaling pathway, the TNF signaling pathway, the Relaxin signaling pathway, the VEGF signaling pathway, and the FoxO signaling pathway. Myricanone acted on two targets (EGFR and STAT3), regulating four key pathways, including the Prolactin signaling pathway, the AGE-RAGE signaling pathway, the Relaxin signaling pathway, and the FoxO signaling pathway. Wallichilde acted on eight targets (AKT1, STAT3, CASP3, SRC, CCND1, MAPK14, MAPK8, and SLC2A2), influencing six key pathways, including the Prolactin signaling pathway, the AGE-RAGE signaling pathway, the TNF signaling pathway, the Relaxin signaling pathway, the VEGF signaling pathway, and FoxO signaling pathway. And senkyunone interacted with five targets (ICAM1, SRC, MMP9, KDR, and MAPK8), affecting the same six key pathways.

Molecular docking between active ingredients and key targets

Molecular docking was performed for the top five core targets (AKT1, EGFR, CASP3, STAT3, and PTGS2) overlapping with the targets in the drug-component-target-signaling pathway network and their corresponding active ingredients of *Rhizoma Chuanxiong* (Myricanone, Mandenol, wallichilde, and senkyunone) (Table 4). Mandenol exhibited stable

binding efficacy to EGFR. The binding affinity between Myricanone, senkyunone, and five protein targets (AKT1, EGFR, CASP3, STAT3, and PTGS2) were less than -5.0 kcal/mol, which indicating the stable binding patterns between the active compounds and protein targets. Additionally, wallichilde demonstrated stable binding affinity to AKT1, EGFR, STAT3, and PTGS2. Among them, the lowest binding affinity of 2RGP protein was -7.85 kcal/mol between EGFR and Myricanone. The molecular docking results visualized using PyMOL indicate that Myricanone binds more strongly to GLY796, MET793, ASP855, and LYS745 in 2RGP, with a total of six hydrogen bonds with lengths of 2.7, 2.2, 1.9, 3.5, 2.9, and 1.8 angstroms (Figure 7).

Discussion

DKD is a chronic microvascular complication caused by diabetes, which occurs in about 20–40% of diabetic patients, and causes a significant economic and social burden [29]. Although current therapeutic strategies, including intensive control of glucose, lipid, urinary protein, and aggressive blood pressure, can delay the progression speed of DKD to end-stage renal disease, there is still no effective therapy to completely halt its progression to end-stage renal disease. Therefore, there is an urgent need to explore novel therapeutic alternatives. Clinical and experimental studies have shown that *Rhizoma Chuanxiong* exhibits promising nephroprotective effects in DKD patients, with a favorable safety profile [11–13]. However, the related mechanisms remain unclear. In the present study, we used a network pharmacological approach to uncover the major constituents, potential therapeutic targets, and significant pathways of *Rhizoma Chuanxiong* in the treatment of DKD.

We discovered that the primary active components of *Rhizoma Chuanxiong*, including Mandenol, Myricanone, senkyunone, and wallichilde, may play significant roles in the treatment of DKD. Although we selected active components of *Rhizoma Chuanxiong* based on the parameters ADME, which cannot fully predict the metabolic fate of the components *in vivo*. After metabolism in the *vivo*, active components may undergo structural modifications or lose critical functional groups, which can impair their ability to bind with targets. In addition, the metabolism of active components *in*

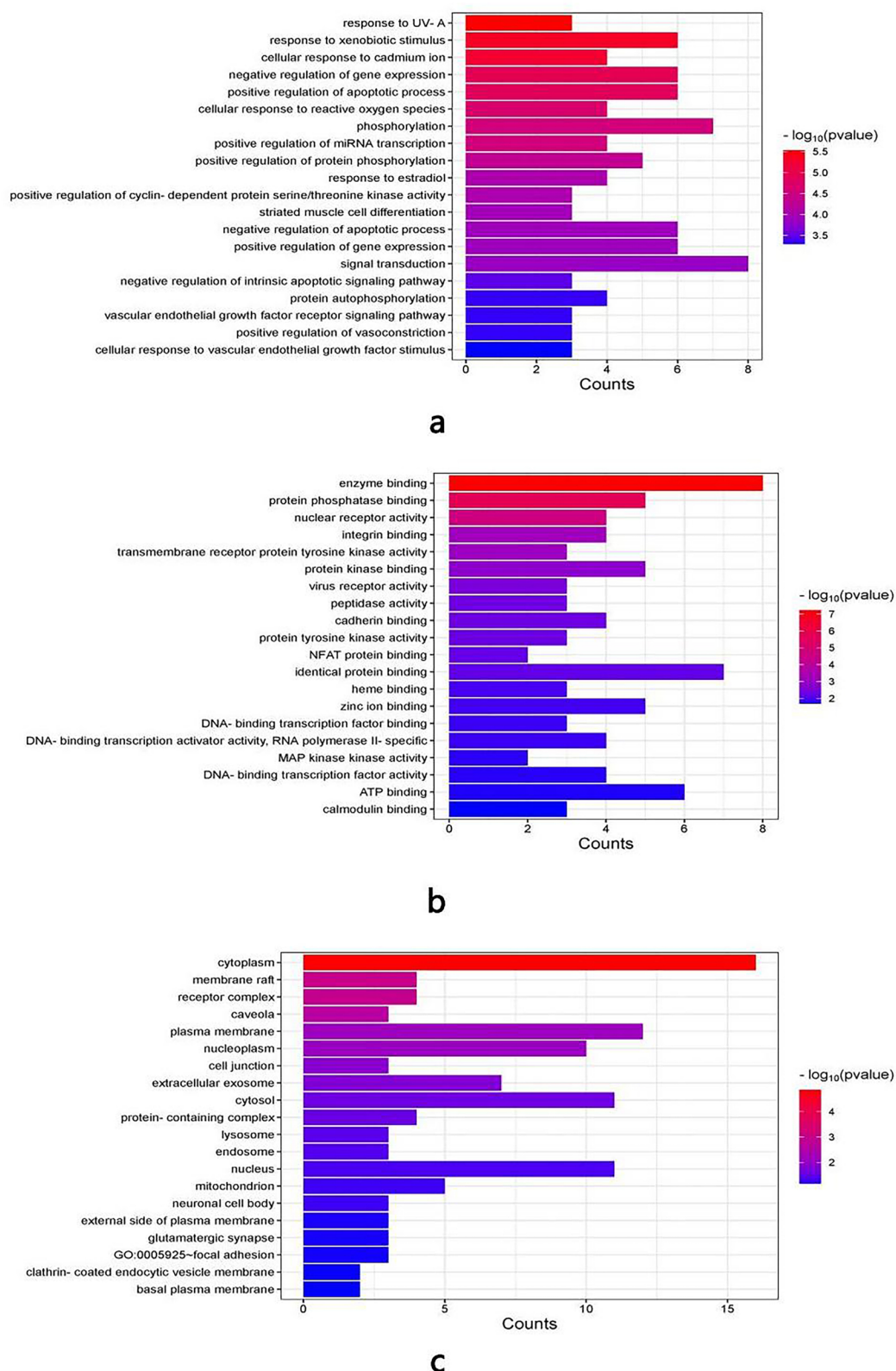


Figure 4. (a–c) GO (BP, MF, CC) analyses the therapeutic target genes of *Rhizoma Chuanxiong* for treatment of DKD.

in vivo may reduce the amount of effective components entering systemic circulation, resulting in insufficient concentrations at target tissues and ultimately leading to reduced drug bioavailability. Therefore, we further reviewed the relevant

literature and found that Mandenol, Myricanone, senkyunone still retain the therapeutic efficacy even after undergoing metabolism *in vivo* [30–35]. No relevant literature reported the therapeutic efficacy of wallichilide *in vivo* experiment. It

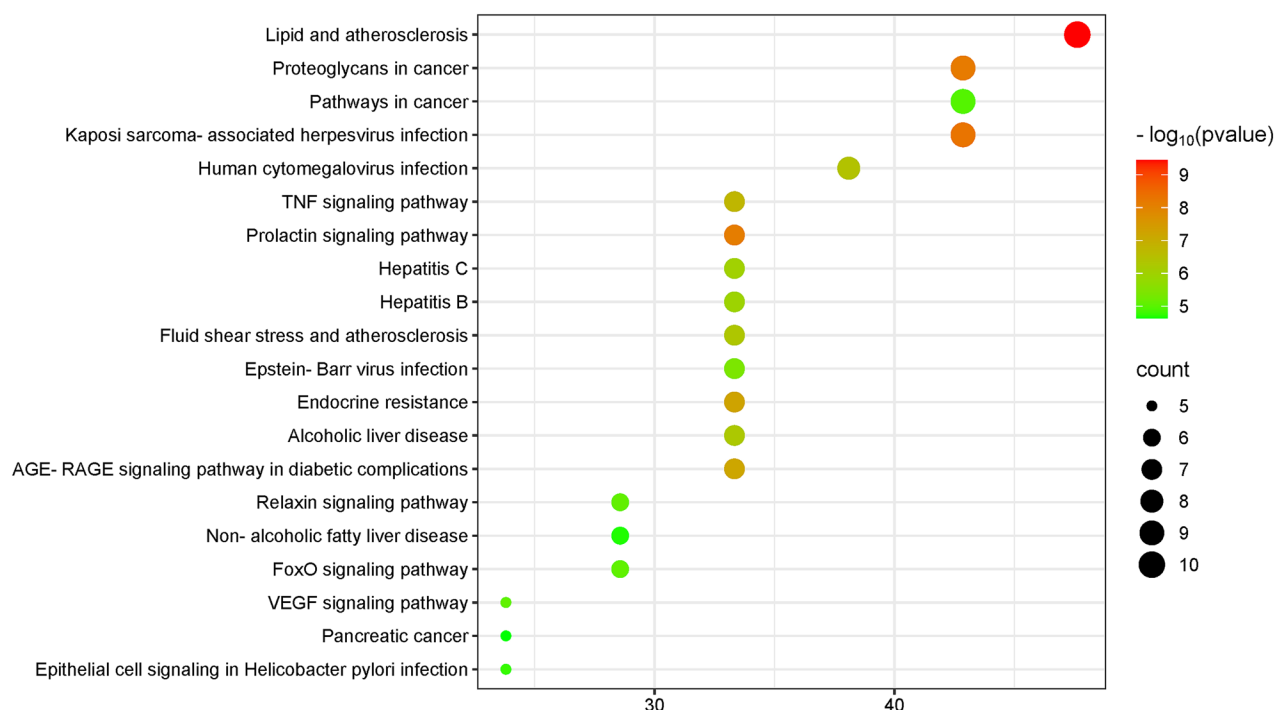


Figure 5. KEGG enrichment analysis of the *Rhizoma Chuanxiong* for the treatment of DKD.

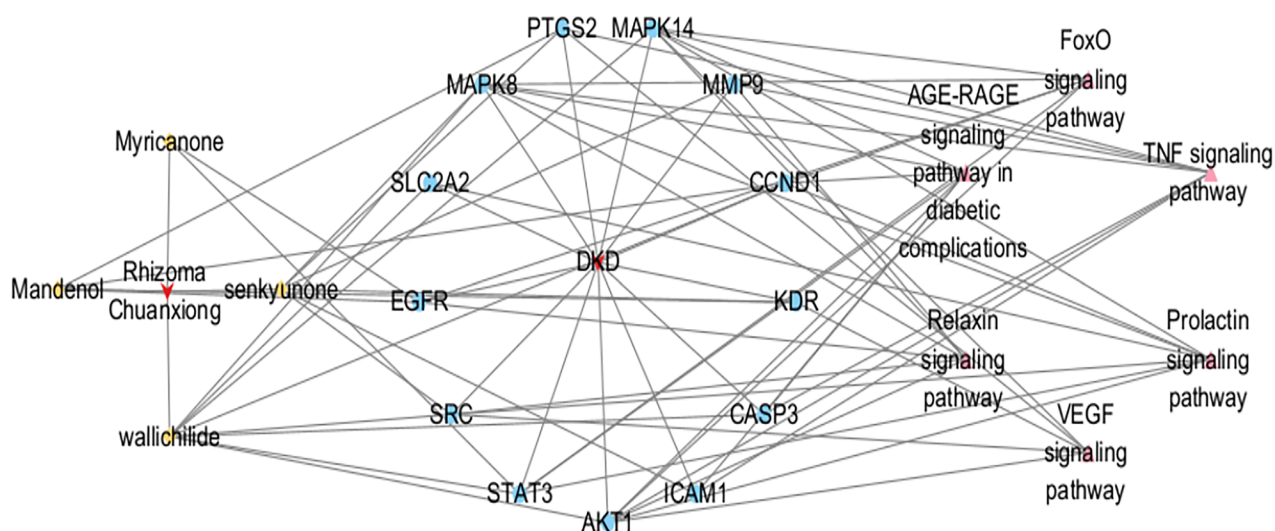


Figure 6. Relationships between KEGG pathways and active compounds, targets of *Rhizoma Chuanxiong* for the treatment of DKD.

Table 4. The binding energy of main compounds and key targets.

Active ingredients	Binding energy (kcal/mol)				
	AKT1	EGFR	CASP3	STAT3	PTGS2
Mandenol	-3.39	-5.1	-2.19	-3.46	-3.85
Myricanone	-6.94	-7.85	-5.3	-5.64	-6.66
Senkyunone	-6.22	-6.58	-5.21	-5.46	-6.18
Wallichilde	-5.34	-7.23	-4.33	-5.91	-7.19

is noteworthy that Mandenol has a DL value of 0.19, which is close to the lower threshold, and its low polarity ($H_{don} = 0$, $H_{acc} = 2$) may influence its bioavailability and target binding capacity. However, the small molecular size and lipophilic nature of Mandenol could facilitate its

absorption through passive diffusion, compensating for its low DL value and polarity. In addition, several fundamental research have demonstrated the therapeutic effect of Mandenol for disease (such as erectile dysfunction, atherosclerosis, xanthomatous biliary cirrhosis) *in vivo* [30–32].

The active components of *Rhizoma Chuanxiong* are associated with multiple proteins and signaling pathways, indicating their potential for further investigation. Based on the KEGG pathway enrichment analysis, drug-component-target-signaling pathway network revealed that numerous targets in DKD are regulated by various compounds. These targets include, but are not limited to AKT1, EGFR, STAT3, CASP3, ICAM1, PTGS2, SRC, and MMP9. AKT1, also known

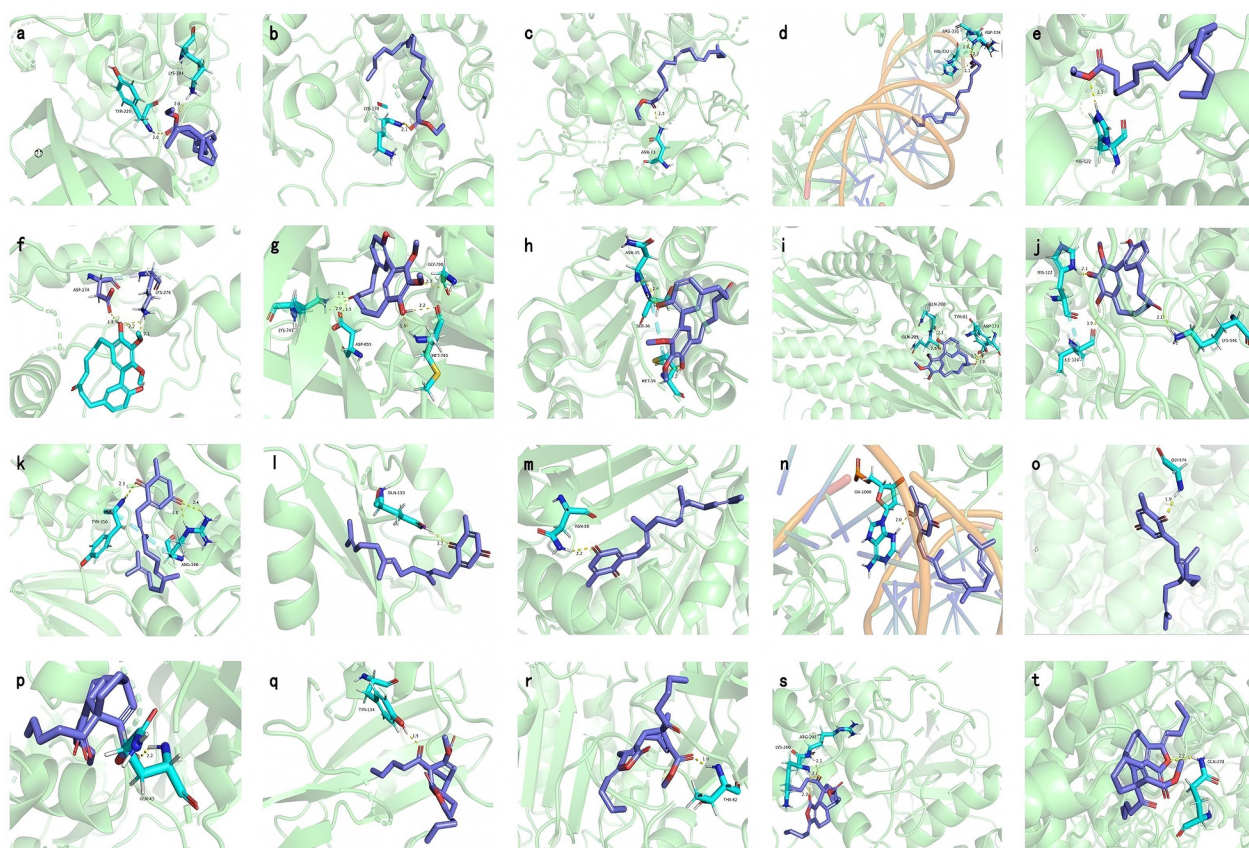


Figure 7. Molecular docking between active ingredients and key targets. (a) Mandenol with AKT1 (PDB code: 4GV1); (b) Mandenol with EGFR (PDB code: 4RJ3); (c) Mandenol with CASP3 (PDB code: 5IAE); (d) Mandenol with STAT3 (PDB code: 6QHD); (e) Mandenol with PTGS2 (PDB code: 5F1A); (f) Myricanone with AKT1 (PDB code: 4GV1); (g) Myricanone with EGFR (PDB code: 2RGP); (h) Myricanone with CASP3 (PDB code: 3PD1); (i) Myricanone with STAT3 (PDB code: 6TLC); (j) Myricanone with PTGS2 (PDB code: 5IKV); (k) Senkyunone with AKT1 (PDB code: 3QKL); (l) Senkyunone with EGFR (PDB code: 3IN8); (m) Senkyunone with CASP3 (PDB code: 4EHL); (n) Senkyunone with STAT3 (PDB code: 6QHD); (o) Senkyunone with PTGS2 (PDB code: 5IKR); (p) Wallichilide with AKT1 (PDB code: 7NH5); (q) Wallichilide with EGFR (PDB code: 3WA4); (r) Wallichilide with CASP3 (PDB code: 5IAB); (s) Wallichilide with STAT3 (PDB code: 5AX3); (t) Wallichilide with PTGS2 (PDB code: 5IKR).

as protein kinase B, participates in multiple signaling pathways related to DKD. These complex signaling pathways underscore the diverse regulatory roles of AKT1 in kidney cell processes affected by DKD and contribute to renal protection [36]. EGFR, a member of the receptor tyrosine kinase ErbB family, is widely expressed in the mammalian kidney, including the podocyte, and plays a critical role in regulating and maintaining the filtration function of glomeruli [37]. Podocyte-specific deletion of EGFR significantly ameliorates the progression of glomerular injury and tubulointerstitial fibrosis in DKD, and this protection was associated with reducing reactive oxygen species, increased podocyte integrity, and the maintenance of autophagy through the inhibition of rubicon expression [37,38]. STAT3, a member of the STAT family, plays a crucial role in the early inflammatory response in DKD, and contributes to proteinuria, glomerular cell proliferation, renal fibrosis of in diabetic mice [39,40]. Inhibition of STAT3 attenuates progression of DKD. CASP3, a protease involved in apoptosis, triggers cell apoptosis upon activation [41]. Reduced CASP3 activity in db/db mice has been shown to inhibit the progression of DKD [42]. ICAM1, an acute-phase protein marker of inflammation, is overexpressed in the glomeruli and

tubular epithelial cells of diabetic animal models, and it participates in the development of DKD [43]. PTGS2 is a key enzyme in the conversion of arachidonic acid to the inflammatory mediator prostaglandins and key markers of ferroptosis [44]. PTGS2 influences renin release and regulation of the RAAS, as well as renal blood flow and hemodynamics [45]. The activation of RASS system and ferroptosis are significant contributors to the pathogenesis and progression of DKD [46,47]. In this study, we discovered that EGFR and PTGS2 exhibit stronger binding affinities with Mandenol, Myricanone, senkyunone, and wallichilide than with other targets, suggesting that EGFR and PTGS2 may play crucial roles in the therapeutic effects of *Rhizoma Chuanxiong* on DKD.

According to KEGG enrichment analysis, the pharmacological effects of *Rhizoma Chuanxiong* on DKD are closely associated with six signaling pathways: Prolactin signaling pathway, AGE-RAGE signaling pathway, TNF signaling pathway, Relaxin signaling pathway, VEGF signaling pathway, and FoxO signaling pathway. Previous studies have demonstrated that urine albumin excretion was significantly and positively correlated with serum prolactin levels and with mRNA expressions of the prolactin receptor, suggesting a potential

role of prolactin in the development of renal injury in DKD [48,49]. The AGE-RAGE signaling pathway is associated with chronic inflammation, oxidative stress, and apoptosis, ultimately leading to kidney damage in animal models of diabetic kidney disease (DKD). Potential treatment options for DKD through the inhibition of AGE-RAGE have been reviewed [50,51]. The TNF pathway is closely associated with systemic inflammation and may mediate the progression of DKD pathogenesis [52]. Elevated TNF levels are associated with increased epithelial damage and death *in vitro* and *vivo*, and TNF has been recognized as a potential therapeutic target for DKD [53]. Studies have indicated that TNF- α is highly expressed in DKD, and elevated levels of TNF- α promote renal lipid accumulation and tubular injury [54]. Relaxin, a kind of peptide hormone, mediates its actions through relaxin/insulin-like family peptide receptor 1 (RXFP1). Relaxin treatment can increase the production of bile acids in the kidney cortex of streptozocin-induced diabetic mice, and ameliorate the increase of markers of oxidative stress, inflammatory cytokines, and renal fibrosis [55]. VEGF is a critical regulator in endothelial cell survival, proliferation, and angiogenesis, and upregulation of VEGF in glomerular is related to the pathogenesis of DKD [56]. Inhibition of VEGF signal pathway attenuate DKD by reducing oxidative stress, inflammation, and apoptosis [56,57]. The FoxO pathway primarily plays a role in the inflammatory response, oxidative stress, mitophagy, mitochondrial dysfunction, and apoptosis in DKD [58–61]. Research has shown that FOXO1 activity is significantly diminished in diabetic kidneys, and the upregulation of FoxO1 markedly alleviates renal functional impairment, delays renal tubulointerstitial fibrosis, and reduces apoptosis in diabetic mice [62–64]. In our study, we found that Prolactin signaling pathway, AGE-RAGE signaling pathway, Relaxin signaling pathway, and FoxO signaling pathway are all related to the core targets (AKT1, EGFR, CASP3, STAT3, and PTGS2).

Previous studies primarily focused on the general nephro-protective effects of *Rhizoma Chuanxiong* our research provides a more comprehensive and mechanistic perspective by identifying specific active compounds (Mandenol, Myricanone, senkyunone, wallichilide) and their multi-target interactions with key proteins and signaling pathways implicated in DKD pathogenesis. The targets and pathways related to therapeutic efficacy of *Rhizoma Chuanxiong* for DKD in our study are known to play pivotal roles in key pathological processes of DKD, such as chronic inflammatory response, oxidative stress, apoptosis, autophagy, mitochondrial dysfunction, which reminds us *Rhizoma Chuanxiong* may treat DKD by improving these key pathological processes of DKD. The ethanol extract of *Rhizoma Chuanxiong Hort.* has been demonstrated to attenuate both structural and functional renal damage in a streptozotocin-induced DKD model *in vivo* through inhibiting oxidative stress and inflammatory responses. These findings are consistent with our findings [12]. The clinical implication of our findings is particularly noteworthy. The identification of specific active compounds of *Rhizoma Chuanxiong* and their associated targets provides a solid foundation for future drug

development and precision medicine approaches in DKD treatment. However, our study has a notable limitation that should be acknowledged. We relied exclusively on network pharmacology and molecular docking approaches to elucidate the potential mechanisms of *Rhizoma Chuanxiong* for the treatment of DKD, without experimental validation. Therefore, our findings provide only preliminary mechanistic insights rather than definitive conclusions, and they must be confirmed by real experimental data.

Conclusion

In conclusion, our study provides a comprehensive analysis and discussion of the potential mechanisms of *Rhizoma Chuanxiong* in treating DKD, based on network pharmacology and molecular docking analysis. The results suggest that the therapeutic efficacy of *Rhizoma Chuanxiong* for DKD may be associated with multiple pathways, such as Prolactin signaling pathway, AGE-RAGE signaling pathway, TNF signaling pathway, Relaxin signaling pathway, VEGF signaling pathway, and FoxO signaling pathway through targets, such as AKT1, EGFR, STAT3, CASP3, ICAM1, PTGS2, SRC, and MMP9. This study establishes a theoretical foundation for further research on the mechanisms of *Rhizoma Chuanxiong* for the treatment of DKD, and additional experimental validation is necessary.

Authors contributions

Y.Y. wrote the manuscript and analyzed the data. Y.L., X.W., and J.L. participated in data analysis. L.W. and H.S. participated in article topic selection, paper writing instruction, and revision. All authors have read and approved the final manuscript for publication.

Disclosure statement

No potential conflict of interest was reported by the author(s).

Data and materials availability statement

The datasets analyzed during the current study are available from the corresponding author on reasonable request.

Funding

This work was supported by the National Natural Science Foundation of China under Grant [No. 82170746]; Basic Research Program of Science and Technology Department of Shanxi Province under Grant [No. 202303021222331]; Doctoral Foundation of the Second Hospital of Shanxi Medical University under Grant [No. 202301-20].

ORCID

Lihua Wang  <http://orcid.org/0000-0003-2741-7765>

References

- [1] GBD 2021 Diabetes Collaborators. Global, regional, and national burden of diabetes from 1990 to 2021, with projections of prevalence to 2050: a systematic analysis for the Global Burden of Disease Study 2021. *Lancet*. 2023;402(10397):203–234. doi: [10.1016/S0140-6736\(23\)01301-6](https://doi.org/10.1016/S0140-6736(23)01301-6).
- [2] Gansevoort RT, Correa-Rotter R, Hemmelgarn BR, et al. Chronic kidney disease and cardiovascular risk: epidemiology, mechanisms, and prevention. *Lancet*. 2013;382(9889):339–352. doi: [10.1016/S0140-6736\(13\)60595-4](https://doi.org/10.1016/S0140-6736(13)60595-4).
- [3] Naaman SC, Bakris GL. Diabetic nephropathy: update on pillars of therapy slowing progression. *Diabetes Care*. 2023;46(9):1574–1586. doi: [10.2337/dci23-0030](https://doi.org/10.2337/dci23-0030).
- [4] Jung CY, Yoo TH. Pathophysiologic mechanisms and potential biomarkers in diabetic kidney disease. *Diabetes Metab J*. 2022;46(2):181–197. doi: [10.4093/dmj.2021.0329](https://doi.org/10.4093/dmj.2021.0329).
- [5] Tuttle KR, Agarwal R, Alpers CE, et al. Molecular mechanisms and therapeutic targets for diabetic kidney disease. *Kidney Int*. 2022;102(2):248–260. doi: [10.1016/j.kint.2022.05.012](https://doi.org/10.1016/j.kint.2022.05.012).
- [6] Lim S, Lim C, Cho S. Methanol extract of *Ligusticum chuanxiong* Hort. Rhizome ameliorates bilateral common carotid artery stenosis-induced cognitive deficit in mice by altering microglia and astrocyte activation. *Front Pharmacol*. 2024;15:1329895. doi: [10.3389/fphar.2024.1329895](https://doi.org/10.3389/fphar.2024.1329895).
- [7] He S, He X, Pan S, et al. Exploring the mechanism of *Chuanxiong Rhizoma* against thrombosis based on network pharmacology, molecular docking and experimental verification. *Molecules*. 2023;28(18):6702. doi: [10.3390/molecules28186702](https://doi.org/10.3390/molecules28186702).
- [8] Li Y, Li F, Ding M, et al. *Chuanxiong Rhizoma* extracts prevent liver fibrosis via targeting CTCF-c-MYC-H19 pathway. *Chin Herb Med*. 2024;16(1):82–93. doi: [10.1016/j.chmed.2023.07.003](https://doi.org/10.1016/j.chmed.2023.07.003).
- [9] Li D, Long Y, Yu S, et al. Research advances in cardio-cerebrovascular diseases of *Ligusticum chuanxiong* Hort. *Front Pharmacol*. 2021;12:832673. doi: [10.3389/fphar.2021.832673](https://doi.org/10.3389/fphar.2021.832673).
- [10] Li J, Li T, Li Z, et al. Nephroprotective mechanisms of *Rhizoma Chuanxiong* and *Radix et Rhizoma Rhei* against acute renal injury and renal fibrosis based on network pharmacology and experimental validation. *Front Pharmacol*. 2023;14:1154743. doi: [10.3389/fphar.2023.1154743](https://doi.org/10.3389/fphar.2023.1154743).
- [11] Wang B, Ni Q, Wang X, et al. Meta-analysis of the clinical effect of ligustrazine on diabetic nephropathy. *Am J Chin Med*. 2012;40(1):25–37. doi: [10.1142/S0192415X12500036](https://doi.org/10.1142/S0192415X12500036).
- [12] Yang WJ, Li YR, Gao H, et al. Protective effect of the ethanol extract from *Ligusticum chuanxiong* rhizome against streptozotocin-induced diabetic nephropathy in mice. *J Ethnopharmacol*. 2018;227:166–175. doi: [10.1016/j.jep.2018.08.037](https://doi.org/10.1016/j.jep.2018.08.037).
- [13] Qi XY, Peng GC, Han QT, et al. Phthalides from the rhizome of *Ligusticum chuanxiong* Hort. attenuate diabetic nephropathy in mice. *J Ethnopharmacol*. 2024;319(Pt 2):117247. doi: [10.1016/j.jep.2023.117247](https://doi.org/10.1016/j.jep.2023.117247).
- [14] Zhao L, Zhang H, Li N, et al. Network pharmacology, a promising approach to reveal the pharmacology mechanism of Chinese medicine formula. *J Ethnopharmacol*. 2023;309:116306. doi: [10.1016/j.jep.2023.116306](https://doi.org/10.1016/j.jep.2023.116306).
- [15] Liu H, Wang J, Zhou W, et al. Systems approaches and polypharmacology for drug discovery from herbal medicines: an example using licorice. *J Ethnopharmacol*. 2013;146(3):773–793. doi: [10.1016/j.jep.2013.02.004](https://doi.org/10.1016/j.jep.2013.02.004).
- [16] Xu X, Zhang W, Huang C, et al. A novel chemometric method for the prediction of human oral bioavailability. *Int J Mol Sci*. 2012;13(6):6964–6982. doi: [10.3390/ijms13066964](https://doi.org/10.3390/ijms13066964).
- [17] Shi H, Hou Y, Su X, et al. Mechanism of action of *Tripterygium wilfordii* for treatment of idiopathic membranous nephropathy based on network pharmacology. *Ren Fail*. 2022;44(1):116–125. doi: [10.1080/0886022X.2021.2024850](https://doi.org/10.1080/0886022X.2021.2024850).
- [18] Lipinski CA, Lombardo F, Dominy BW, et al. Experimental and computational approaches to estimate solubility and permeability in drug discovery and development settings. *Adv Drug Deliv Rev*. 2001;46(1–3):3–26. doi: [10.1016/S0169-409X\(00\)00129-0](https://doi.org/10.1016/S0169-409X(00)00129-0).
- [19] Gfeller D, Grosdidier A, Wirth M, et al. SwissTargetPrediction: a web server for target prediction of bioactive small molecules. *Nucleic Acids Res*. 2014;42(Web Server issue):W32–W38. doi: [10.1093/nar/gku293](https://doi.org/10.1093/nar/gku293).
- [20] Cheung S, Zhong Y, Wu L, et al. Mechanism interpretation of Guhan Yangshengjing for protection against Alzheimer's disease by network pharmacology and molecular docking. *J Ethnopharmacol*. 2024;328:117976. doi: [10.1016/j.jep.2024.117976](https://doi.org/10.1016/j.jep.2024.117976).
- [21] Cheng M, Li M, Zhang Y, et al. Exploring the mechanism of PPCPs on human metabolic diseases based on network toxicology and molecular docking. *Environ Int*. 2025;196:109324. doi: [10.1016/j.envint.2025.109324](https://doi.org/10.1016/j.envint.2025.109324).
- [22] Tuo Y, Lu X, Tao F, et al. The potential mechanisms of catechins in tea for anti-hypertension: an integration of network pharmacology, molecular docking, and molecular dynamics simulation. *Foods*. 2024;13(17):2685. doi: [10.3390/foods13172685](https://doi.org/10.3390/foods13172685).
- [23] Gao X, Li S, Cong C, et al. A network pharmacology approach to estimate potential targets of the active ingredients of epimedium for alleviating mild cognitive impairment and treating Alzheimer's disease. *Evid Based Complement Alternat Med*. 2021;2021:2302680. doi: [10.1155/2021/2302680](https://doi.org/10.1155/2021/2302680).
- [24] Szklarczyk D, Kirsch R, Koutrouli M, et al. The STRING database in 2023: protein-protein association networks and functional enrichment analyses for any sequenced genome of interest. *Nucleic Acids Res*. 2023;51(D1):D638–D646. doi: [10.1093/nar/gkac1000](https://doi.org/10.1093/nar/gkac1000).
- [25] Scardoni G, Tosadori G, Faizan M, et al. Biological network analysis with CentiScaPe: centralities and experimental dataset integration. *F1000Research*. 2014;3:139. doi: [10.12688/f1000research.4477.2](https://doi.org/10.12688/f1000research.4477.2).
- [26] Shannon P, Markiel A, Ozier O, et al. Cytoscape: a software environment for integrated models of biomolecular interaction networks. *Genome Res*. 2003;13(11):2498–2504. doi: [10.1101/gr.1239303](https://doi.org/10.1101/gr.1239303).
- [27] Ashburner M, Ball CA, Blake JA, et al. Gene ontology: tool for the unification of biology. The Gene Ontology Consortium. *Nat Genet*. 2000;25(1):25–29. doi: [10.1038/75556](https://doi.org/10.1038/75556).
- [28] Gao Y, Shang B, He Y, et al. The mechanism of Gejie Zhilao Pill in treating tuberculosis based on network pharmacology and molecular docking verification. *Front*

- Cell Infect Microbiol. 2024;14:1405627. doi: [10.3389/fcimb.2024.1405627](https://doi.org/10.3389/fcimb.2024.1405627).
- [29] Conserva F, Gesualdo L, Papale M. A systems biology overview on human diabetic nephropathy: from genetic susceptibility to post-transcriptional and post-translational modifications. *J Diabetes Res*. 2016;2016:7934504. doi: [10.1155/2016/7934504](https://doi.org/10.1155/2016/7934504).
- [30] Ge P, Xie H, Guo Y, et al. Linoleyl acetate and mandenol alleviate HUA-induced ED via NLRP3 inflammasome and JAK2/STAT3 signalling conduction in rats. *J Cell Mol Med*. 2024;28(17):e70075. doi: [10.1111/jcmm.70075](https://doi.org/10.1111/jcmm.70075).
- [31] Koo HJ, Park HJ, Byeon HE, et al. Chinese yam extracts containing β -sitosterol and ethyl linoleate protect against atherosclerosis in apolipoprotein E-deficient mice and inhibit muscular expression of VCAM-1 *in vitro*. *J Food Sci*. 2014;79(4):H719–H729. doi: [10.1111/1750-3841.12401](https://doi.org/10.1111/1750-3841.12401).
- [32] Fuld H, Horwich L. Xanthomatous biliary cirrhosis treated with ethyl linoleate. *Lancet*. 1959;1(7078):890–891. doi: [10.1016/s0140-6736\(59\)91979-8](https://doi.org/10.1016/s0140-6736(59)91979-8).
- [33] Datey KK, Purandare NM, Dalvi CP. Ethyl linoleate as an antiatherogenic agent. An experimental study in rabbits. *Angiology*. 1966;17(10):732–737. doi: [10.1177/000331976601701003](https://doi.org/10.1177/000331976601701003).
- [34] Yang H, Cao J, Zhou L, et al. Exploring the cardioprotective mechanisms of *Ligusticum wallichii* in myocardial infarction through network pharmacology and experimental validation. *Drug Des Devel Ther*. 2025;19:281–302. doi: [10.2147/DDDT.S481499](https://doi.org/10.2147/DDDT.S481499).
- [35] Paul A, Das J, Das S, et al. Anticancer potential of myricanone, a major bioactive component of *Myrica cerifera*: novel signaling cascade for accomplishing apoptosis. *J Acupunct Meridian Stud*. 2013;6(4):188–198. doi: [10.1016/j.jams.2013.05.003](https://doi.org/10.1016/j.jams.2013.05.003).
- [36] Heljić M, Brazil DP. Protein kinase B/Akt regulation in diabetic kidney disease. *Front Biosci*. 2011;3(1):98–104. doi: [10.2741/s135](https://doi.org/10.2741/s135).
- [37] Chen J, Chen JK, Nagai K, et al. EGFR signaling promotes TGF β -dependent renal fibrosis. *J Am Soc Nephrol*. 2012;23(2):215–224. doi: [10.1681/ASN.2011070645](https://doi.org/10.1681/ASN.2011070645).
- [38] Li Y, Pan Y, Cao S, et al. Podocyte EGFR inhibits autophagy through upregulation of rubicon in type 2 diabetic nephropathy. *Diabetes*. 2021;70(2):562–576. doi: [10.2337/db20-0660](https://doi.org/10.2337/db20-0660).
- [39] Zheng C, Huang L, Luo W, et al. Inhibition of STAT3 in tubular epithelial cells prevents kidney fibrosis and nephropathy in STZ-induced diabetic mice. *Cell Death Dis*. 2019;10(11):848. doi: [10.1038/s41419-019-2085-0](https://doi.org/10.1038/s41419-019-2085-0).
- [40] Li L, Wei T, Liu S, et al. Complement C5 activation promotes type 2 diabetic kidney disease via activating STAT3 pathway and disrupting the gut-kidney axis. *J Cell Mol Med*. 2021;25(2):960–974. doi: [10.1111/jcmm.16157](https://doi.org/10.1111/jcmm.16157).
- [41] Fuchs Y, Steller H. Live to die another way: modes of programmed cell death and the signals emanating from dying cells. *Nat Rev Mol Cell Biol*. 2015;16(6):329–344. doi: [10.1038/nrm3999](https://doi.org/10.1038/nrm3999).
- [42] Ghosh S, Khazaei M, Moien-Afshari F, et al. Moderate exercise attenuates caspase-3 activity, oxidative stress, and inhibits progression of diabetic renal disease in db/db mice. *Am J Physiol Renal Physiol*. 2009;296(4):F700–F708. doi: [10.1152/ajprenal.90548.2008](https://doi.org/10.1152/ajprenal.90548.2008).
- [43] Gu HF, Ma J, Gu KT, et al. Association of intercellular adhesion molecule 1 (ICAM1) with diabetes and diabetic nephropathy. *Front Endocrinol*. 2012;3:179. doi: [10.3389/fendo.2012.00179](https://doi.org/10.3389/fendo.2012.00179).
- [44] Xie Y, Hou W, Song X, et al. Ferroptosis: process and function. *Cell Death Differ*. 2016;23(3):369–379. doi: [10.1038/cdd.2015.158](https://doi.org/10.1038/cdd.2015.158).
- [45] Nasrallah R, Hassouneh R, Hébert RL. PGE2, kidney disease, and cardiovascular risk: beyond hypertension and diabetes. *J Am Soc Nephrol*. 2016;27(3):666–676. doi: [10.1681/ASN.2015050528](https://doi.org/10.1681/ASN.2015050528).
- [46] Rahimi Z. The role of renin angiotensin aldosterone system genes in diabetic nephropathy. *Can J Diabetes*. 2016;40(2):178–183. doi: [10.1016/j.jcjd.2015.08.016](https://doi.org/10.1016/j.jcjd.2015.08.016).
- [47] Wang H, Liu D, Zheng B, et al. Emerging role of ferroptosis in diabetic kidney disease: molecular mechanisms and therapeutic opportunities. *Int J Biol Sci*. 2023;19(9):2678–2694. doi: [10.7150/ijbs.81892](https://doi.org/10.7150/ijbs.81892).
- [48] Al-Trad B. Prolactin receptor mRNA expression in experimental diabetic nephropathy: relationship with urinary albumin excretion. *Neuro Endocrinol Lett*. 2015;36(6):552–556.
- [49] Singh U, Mathur MK, Shalini. Association of serum prolactin levels with glycemic control in type 2 diabetes mellitus & its correlation with diabetic nephropathy. *J Assoc Phys India*. 2023;71(1):1.
- [50] Sanajou D, Ghorbani Haghighi A, Argani H, et al. AGE-RAGE axis blockade in diabetic nephropathy: current status and future directions. *Eur J Pharmacol*. 2018;833:158–164. doi: [10.1016/j.ejphar.2018.06.001](https://doi.org/10.1016/j.ejphar.2018.06.001).
- [51] Ramasamy R, Yan SF, Schmidt AM. Receptor for AGE (RAGE): signaling mechanisms in the pathogenesis of diabetes and its complications. *Ann N Y Acad Sci*. 2011;1243(1):88–102. doi: [10.1111/j.1749-6632.2011.06320.x](https://doi.org/10.1111/j.1749-6632.2011.06320.x).
- [52] Cao L, Boston A, Jegede O, et al. Inflammation and kidney injury in diabetic African American men. *J Diabetes Res*. 2019;2019:5359635. doi: [10.1155/2019/5359635](https://doi.org/10.1155/2019/5359635).
- [53] Park J, Guan Y, Sheng X, et al. Functional methylome analysis of human diabetic kidney disease. *JCI Insight*. 2019;4(11):e128886. doi: [10.1172/jci.insight.128886](https://doi.org/10.1172/jci.insight.128886).
- [54] Yang W, Luo Y, Yang S, et al. Ectopic lipid accumulation: potential role in tubular injury and inflammation in diabetic kidney disease. *Clin Sci*. 2018;132(22):2407–2422. doi: [10.1042/CS20180702](https://doi.org/10.1042/CS20180702).
- [55] Leo CH, Ou JLM, Ong ES, et al. Relaxin elicits renoprotective actions accompanied by increasing bile acid levels in streptozotocin-induced diabetic mice. *Biomed Pharmacother*. 2023;162:114578. doi: [10.1016/j.biopha.2023.114578](https://doi.org/10.1016/j.biopha.2023.114578).
- [56] Yang KJ, Choi WJ, Chang YK, et al. Inhibition of xanthine oxidase protects against diabetic kidney disease through the amelioration of oxidative stress via VEGF/VEGFR axis and NOX-FoxO3a-eNOS signaling pathway. *Int J Mol Sci*. 2023;24(4):3807. doi: [10.3390/ijms24043807](https://doi.org/10.3390/ijms24043807).
- [57] Lou Z, Li Q, Wang C, et al. The effects of microRNA-126 reduced inflammation and apoptosis of diabetic nephropathy through PI3K/AKT signalling pathway by VEGF. *Arch Physiol Biochem*. 2022;128(5):1265–1274. doi: [10.1080/13813455.2020.1767146](https://doi.org/10.1080/13813455.2020.1767146).
- [58] Li W, Du M, Wang Q, et al. FoxO1 promotes mitophagy in the podocytes of diabetic male mice via the PINK1/

- Parkin pathway. *Endocrinology*. 2017;158(7):2155–2167. doi: [10.1210/en.2016-1970](https://doi.org/10.1210/en.2016-1970).
- [59] Wu L, Zhang Y, Ma X, et al. The effect of resveratrol on FoxO1 expression in kidneys of diabetic nephropathy rats. *Mol Biol Rep*. 2012;39(9):9085–9093. doi: [10.1007/s11033-012-1780-z](https://doi.org/10.1007/s11033-012-1780-z).
- [60] Wu L, Wang Q, Guo F, et al. Activation of FoxO1/PGC-1 α prevents mitochondrial dysfunction and ameliorates mesangial cell injury in diabetic rats. *Mol Cell Endocrinol*. 2015;413:1–12. doi: [10.1016/j.mce.2015.06.007](https://doi.org/10.1016/j.mce.2015.06.007).
- [61] Wang Y, He W. Improving the dysregulation of FoxO1 activity is a potential therapy for alleviating diabetic kidney disease. *Front Pharmacol*. 2021;12:630617. doi: [10.3389/fphar.2021.630617](https://doi.org/10.3389/fphar.2021.630617).
- [62] Chen P, Shi X, Xu X, et al. Liraglutide ameliorates early renal injury by the activation of renal FoxO1 in a type 2 diabetic kidney disease rat model. *Diabetes Res Clin Pract*. 2018;137:173–182. doi: [10.1016/j.diabres.2017.09.006](https://doi.org/10.1016/j.diabres.2017.09.006).
- [63] Huang F, Wang Q, Guo F, et al. FoxO1-mediated inhibition of STAT1 alleviates tubulointerstitial fibrosis and tubule apoptosis in diabetic kidney disease. *EBioMedicine*. 2019;48:491–504. doi: [10.1016/j.ebiom.2019.09.002](https://doi.org/10.1016/j.ebiom.2019.09.002).
- [64] Ren H, Shao Y, Wu C, et al. Metformin alleviates oxidative stress and enhances autophagy in diabetic kidney disease via AMPK/SIRT1-FoxO1 pathway. *Mol Cell Endocrinol*. 2020;500:110628. doi: [10.1016/j.mce.2019.110628](https://doi.org/10.1016/j.mce.2019.110628).