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Exosomes in Diabetic Kidney Disease

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Abstract
Background: Diabetic kidney disease (DKD) is a major complication of diabetes mellitus and a common cause of end-stage kidney disease. The incidence of DKD is rising worldwide and associated with increased morbidity and premature mortality, indicating an urgent need to further explore the underlying pathogenesis and potential biomarkers. Exosomes are nanoscale vesicles secreted by all cell types that play an essential role in cellular homeostasis and intercellular communications by transferring molecular cargoes between different cells. ***Summary:*** Emerging evidence indicates that exosomes are both a crucial signaling mediator and a potential biomarker of DKD. On the one hand, exosomes released by various kidney resident cells facilitate the cell-cell crosstalk as a contributing factor in DKD; on the other hand, exosomes can be detected from urine and blood and have emerged as promising noninvasive biomarkers for DKD. ***Key Messages:*** Herein, we highlight the recent advances in research on the role of exosomes from different kidney resident cells in DKD. We further discuss the potential use of urine exosomes as biomarkers and therapeutic agents.

Introduction

Diabetic kidney disease (DKD) is a progressive kidney disease that occurs as a principal long-term complication of diabetes mellitus (DM). It is one of the most common causes of chronic kidney disease (CKD) and end-stage kidney disease worldwide, accounting for 44.5% of patients requiring dialysis or renal transplantation [1]. Approximately half of the patients with type 2 DM and one-third with type 1 DM develop DKD. In conjunction with the rising incidence of DM, the prevalence of DKD is rapidly increasing, which imposes a huge public health and economic burden on a global level [2, 3]. The natural history of DKD is characterized by microalbuminuria and progressive loss of renal function. Microalbuminuria is currently the best noninvasive marker available for assessing the development and progression of DKD. However, 15–65% of patients with microalbuminuria regress to normoalbuminuria and do not progress to macroalbuminuria or CKD. Moreover, 20–60% of DM patients develop DKD with a decreased glomerular filtration rate but without microalbuminuria [4]. Furthermore, the current prevention and intervention strategies which mainly focus on controlling the hyperglycemia, hypertension, dyslipidemia, and suppressing the renin-angiotensin-aldosterone (RAAS) system do not succeed for all DKD patients since the prevalence of DKD and CKD continues to increase

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progressively, thus highlighting the urgent need to elucidate the pathogenesis further and thereby identify diagnostic and predictive biomarker and new therapeutic options of DKD.

Thickening of the glomerular basement membrane and tubular basement membrane is the first pathological sign of DKD, accompanied by the proliferation of mesangial cells, podocyte loss, and inflammatory cell infiltration. Kidney resident cells are involved in the whole pathophysiological process of DKD. The cell-cell interactions among kidney resident cells are based on complex intercellular signaling networks. Through cellular communication by secreted factors, exosomes have been recognized as professional information mediators involved in the pathogenesis of DKD. However, the understanding of the role of exosomes in DKD remains limited [5, 6] since the function of exosomes is mainly driven by the transmission of exosomal cargos and the type of cargos depends on the origin cells where exosomes are secreted. In this review, we focus on the role of exosomes derived from different origins, including renal resident cells and other resources such as mesenchymal stromal cells (MSCs), serum and exogenous exosomes by intramuscular injection, and further address how functional urine exosomes are used to diagnose and predict the progression of DKD.

Exosome

Exosome Biogenesis

Exosome is a group of nano-sized extracellular vesicles with a size range of approximately 30–150 nm secreted by cells. It was initially considered as an unconventional way to get rid of the cellular waste that resulted from the maturation of reticulocytes into erythrocytes in 1983 [7] and was subsequently named "exosomes" by Johnstone in 1987 [8]. More than a decade later, Raposo et al. [9] reported that exosomes isolated from Epstein-Barr virus-transformed B lymphocytes could induce T-cell responses and be essential for the adaptive immune response. Research interest in exosomes has grown substantially since then as exosomes were shown to participate in cellular processes, especially in the immune system. Over the years, exosomes have emerged as an essential player in cellular homeostasis and intercellular communication, involved in numerous cellular physiological and pathological processes [10, 11].

Exosome biogenesis is a protein quality control mechanism and can be generated from both the plasma membrane and the endosome membrane [12]. The generation of exosomes begins from endocytosis to form early endosomes by inward budding of the plasma membrane triggered by external stimuli or microbial attacks. Through the endosomal protein sorting and transport device-dependent machinery or endosomal protein sorting and transport device-independent machinery, the early endosomes sprout inward and mature to multivesicular bodies (also called late endosomes) containing interluminal vesicles. Regulated by Rab GTPases, multivesicular bodies are then secreted to the extracellular space through direct budding with the plasma membrane or receptor-ligand binding, or phagocytosis, and the released vesicles are called exosomes [13–16]. After being released from cells, exosomes are then shed into various body fluids and widely distributed in almost all kinds of body fluids such as blood, cerebrospinal fluid, bile, breast milk, synovial fluid, saliva, bile, ascites, amniotic fluid, and pleural effusion, suggesting an irreplaceable role of exosomes in physiological and pathological conditions [17].

Exosomal Cargo and Function

During exosome biogenesis and release, selective cargo loading occurs, and particular cellular constituents are shuttled into exosomes containing various proteins, microRNAs (miRNAs), mRNAs, DNA, lipids, metabolites, cell surface proteins, and many other cellular components [18]. In essence, an exosome contains a mixture of cargos rather than one specific molecule. Thus, the function of exosomes is highly heterogeneous as their various cargos faithfully reflect the cellular states of their origin cells, and the type, quality, and quantity of cargo depend on cell types and the cellular milieu [19]. Due to exosomes' lipid bilayer membrane structure, cargo within exosomes is protected from degradation and can be successfully transferred to specific recipient cells, mediating cell-cell communication between origin and recipient target cells [11, 20]. Accordingly, exosomes packaged with functional cargo play a vital role in diverse cellular processes and diseases, including DKD [21–24].

The Effect of Exosomes Secreted by Different Cells in DKD

Exosomes transfer autocrine or paracrine signals by cell-cell crosstalk between different kidney resident cells. These involved cells include glomerular endothelial cells (GECs), podocytes, glomerular mesangial cells (GMCs), macrophages, MSCs, and tubular epithelial cells (TECs) and fibroblasts (shown in Fig. 1). The complex functions of exosomes in DKD are summarized as follows and shown in Table 1.

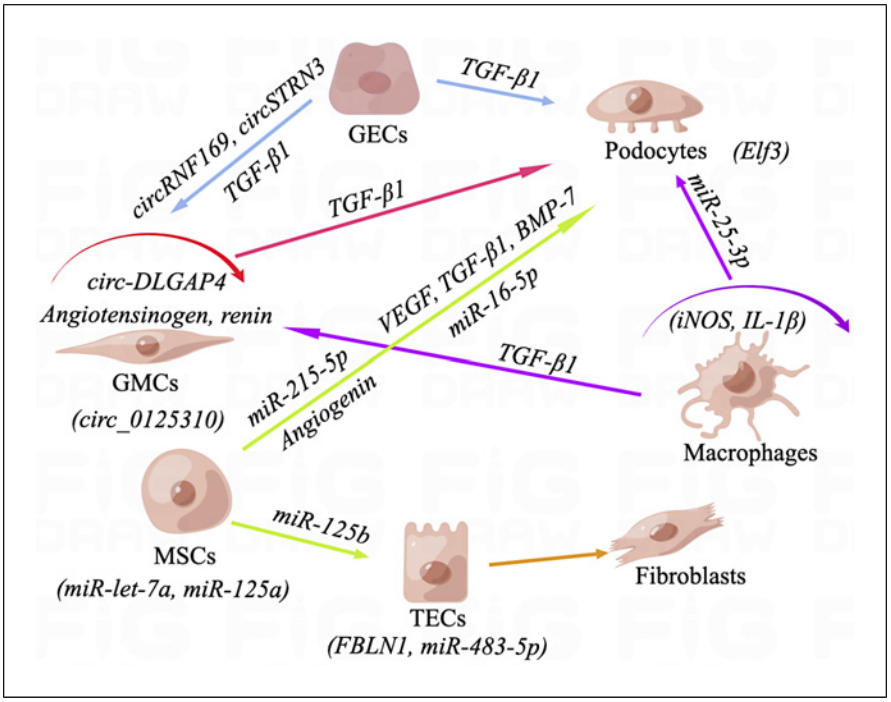

Fig. 1. Exosome-mediated intercellular communications between renal resident cells in DKD. **a** Blue arrow: exosomes derived from glomerular endothelial cells (GECs) transferred paracrine signaling to podocytes by transforming growth factor beta 1 (TGF-β1) and glomerular mesangial cells (GMCs) by circRNF169, circSTRN3, and TGF-β1. **b** Red arrow: exosomes derived from GMCs transferred paracrine signaling to podocytes by TGF-β1 and autocrine signaling to themselves by circ-DLGAP4, angiotensinogen, renin, or circ_0125310. **c** Green arrow: exosomes derived from mesenchymal stromal cells (MSCs) transferred paracrine signaling to tubular epithelial cells (TECs) by miR-125b and podocytes by miR-215-5p, vascular endothelial growth factor (VEGF), TGF-β1, bone morphogenetic protein 7 (BMP-7), angiogenin, miR-16-5p, and autocrine signaling by miR-let-7a, miR-125a. **d** Orange arrow: exosomes derived from TECs transferred paracrine signaling to fibroblasts and autocrine signaling by fibulin 1 (FBLN1), miR-483-5p. **e** Purple arrow: exosomes derived from macrophages transferred paracrine signaling to podocytes by miR-25-3p, GMCs by TGF-β1, and autocrine signaling by inducible nitric oxide synthase (iNOS), interleukin-1β (IL-1β). **f** Podocytes-derived exosomes transferred autocrine signaling by E74-like ETS transcription factor 3 (Elf3).

GEC-Derived Exosomes

GECs are specialized vascular endothelial cells that form the walls of glomerular tuft capillaries. In DKD, GEC dysfunction, including increased permeability, is a key component of diabetes induced organ damage responding with mitochondrial dysfunction and increased reactive oxygen species production [25]. The permeability may allow GEC-derived exosomes to penetrate the glomerular basement membrane to reach podocytes and induce the epithelial-mesenchymal transition (EMT) and dysfunction of podocytes by releasing exosomes containing TGF-β1 [26]. Wu et al. [27] also found that high glucose (HG)-treated GECs secrete more exosomes highly enriched in TGF-β1 mRNA than normal-glucose-treated GECs. They further demonstrated that exosomes derived from HG-treated GECs could promote α-smooth muscle actin (α-SMA) expression and proliferation and extracellular matrix (ECM) protein overproduction in GMCs through the TGF-β1/Smad3 signaling pathway. In contrast, Ling et al. showed that the exosomes derived from HG-treated GECs inhibited GMC proliferation and promoted EMT due to downregulated exosomal circRNF169 and circSTRN3 [28]. The reason underlying the discrepancy between these two studies remains unknown. It is noteworthy that exosomes contain different cargoes under different secretion environments. In addition, the isolation methods affect the quality and quantity of exosomes. Regardless, exosomes are regarded as one of the mechanisms of Tongxinluo in DKD treatment since Tongxinluo was shown to inhibit renal fibrosis by preventing the transfer of TGF-β1 from GECs to GMCs via exosomes in DKD [29].

Table 1. Exosomes involved intercellular communications in DKD

Origin	Recipient	Involved molecules	Functions	Relevant mechanism	PMID
GECs	Podocytes	TGF-β1	Fibrosis and EMT	Stimulates the EMT and dysfunction of podocytes by releasing exosomes containing TGF-β1	28839221
	GMCs	circRNF169, circSTRN3	Inhibited proliferation and promoted EMT	Promoted α-SMA expression	31497190
	GMCs	TGF-β1	Promote renal fibrosis	TGF-β1/Smad3 pathway	27010029, 28659030
GMCs	GMCs	circ-DLGAP4	Proliferation and fibrosis	Sponging miR-143 and modulating ERBB3/NF-κB/MMP-2	33230102
	HMCs	Angiotensinogen, renin	Fibrosis, proliferation	RAS activity	31000742
	Podocytes	TGF-β1	Adhesion capacity, apoptosis	TGF-β1/PI3K-Akt signaling pathway	29378192
	GMCs	circ_0125310	Promoted cell proliferation and fibrosis	Sponging miR-422a and targeting the IGF1R/p38 axis	34854210
PTECs	Fibroblast	–	Fibrosis, proliferation	–	32715764
	PTECs	FBLN1	EMT and ECM accumulation	Modulated by miR-1269b	34977033
	PTECs	miR-483-5p	Promoted fibrosis, ECM deposition	TIMP2 and MAPK1	33692334
Podocytes	–	Elf3	–	TGF-β1/Smad3 pathway	31150422
Macrophages	GMCs	TGF-β1 mRNA	Proliferation, fibrosis, inflammation	TGF-β1/Smad3 pathway	31162940
	Macrophages	iNOS and IL-1β	Proliferation, inflammation	NF-κB/p65 signaling pathway	32388490
	Podocytes	miR-25-3p	Autophagy	Inhibit DUSP	33107695
BM-MSCs	Rat TECs		Inflammation, fibrosis, EMT, apoptosis, degeneration	Regulates TGF-β, c-Src/p38-MAPK, ZO-1, TNF-α, ICAM-1, etc.	27721418
	Human TECs	miR-125b	Autophagy, apoptosis	TRAF6, Akt pathway	34024846
	Rats	-	Ameliorated fibrosis, autophagy	mTOR pathway	30467302
	Rats	miR-let-7a	Apoptosis	Downregulation of USP22	33347877
	Rats	-	Lower the blood glucose, improve renal function	Inhibits JAK2/STAT3	34306382
adMSCs	Podocytes	miR-215-5p	Metastasis, EMT	Inhibit ZEB2	32149094
	GMCs	miR-125a	Suppressed fibrosis	Directly bound to HDAC1 and downregulated ET-1	33790607
hUSCs	Podocytes	VEGF, TGF-β1, angiogenin, BMP-7	Apoptosis, proliferation	Reduces podocytic apoptosis and promotes vascular regeneration and cell survival	26852014
	Podocytes	miR-16-5p	Apoptosis	Suppresses VEGFA expression and podocytic apoptosis	31568645
	Rats	–	Inflammation, fibrosis	Inhibits inflammation and fibrosis	32746936
Muscle	Kidney	miR-23a/27a	Muscle atrophy and renal fibrosis	Akt, FoxO1, PTEN	29582582

Table 1 (continued)

Origin	Recipient	Involved molecules	Functions	Relevant mechanism	PMID
Serum of DKD patients	-	miR-4449	Regulate the expression of pro-inflammatory cytokines, ROS levels, and pyroptosis	Pyroptosis and oxidative stress	34732690

DKD, diabetic kidney disease; GECs, glomerular endothelial cells; TGF-β1, transforming growth factor beta 1; EMT, epithelial-mesenchymal transition; GMCs, glomerular mesangial cells; RNF169, ring finger protein 169; STRN3, striatin 3; α-SMA, alpha smooth muscle actin; Smad3, SMAD family member 3; DLGAP4, DLG-associated protein 4; ERBB3, Erb-B2 receptor tyrosine kinase 3; NF-κB, nuclear factor kappa-B; MMP-2, matrix metallopeptidase 2; HMCs, human mesangial cells; RAS, reticular activating system; PI3K, phosphatidylinositol-3-kinase; IGF1R, insulin-like growth factor 1 receptor; PTECs, proximal tubular epithelial cells; FBLN1, fibulin 1; ECM, extracellular matrix; TIMP2, TIMP metallopeptidase inhibitor 2; MAPK1, mitogen-activated protein kinase 1; Elf3, E74-like ETS transcription factor 3; iNOS, inducible nitric oxide synthase; IL-1β, interleukin-1β; DUSP, dual-specificity phosphatase; BM-MSCs, bone marrow mesenchymal stem cells; ZO-1, zonula occludens-1; TNF-α, tumor necrosis factor α; ICAM-1, intercellular cell adhesion molecule-1; TRAF6, TNF receptor-associated factor 6; USP22, ubiquitin specific peptidase 22; JAK2/STAT3, Janus kinase 2/signal transducer and activator of transcription 3; adMCS, adipose-derived mesenchymal stem cells; ZEB2, zinc finger E-box binding homeobox 2; HDAC1, histone deacetylase 1; ET-1, endothelin-1; hUSCs, human urine-derived stem cells; BMP-7, bone morphogenetic protein 7; VEGFA, vascular endothelial growth factor A; FoxO1, forkhead box O1; PTEN, phosphatase and tensin homologue; ROS, reactive oxygen species.

GMC-Derived Exosomes

GMC is a kind of vital indigenous source of kidney exosomes that might affect the function of the glomerulus in DKD conditions. Exosomes mediate communication between GMCs and podocytes and also GMCs themselves. HG-treated GMCs derived exosomes induced podocyte injury through the TGF-β1-PI3K/AKT pathway. Berberine may be a good candidate to protect against the damage of podocytes by reducing exosomal TGF-β1 and apoptosis and increasing adhesion [30]. In the autocrine pattern, three different studies consistently revealed that exosomes derived from DKD patients, DKD rat models, or HG-treated GMCs could promote proliferation and fibrosis of GMCs by different mechanisms such as an increased level of circ_DLGAP4 sponged miR-143 and regulated the ERBB3/NF-κB/MMP-2 axis [31], higher amounts of RAS components such as angiotensinogen and renin in exosomes [32], and exo-circ_0125310 sponged miR-422a and targeted the IGF1R/p38 axis [33]. These studies indicate that GMC-derived exosomes may promote DKD by inducing podocyte injury, proliferation, and fibrosis of GMCs.

TEC-Derived Exosomes

TECs are the main cells in the kidney and are vulnerable to injury. Accumulated evidence has suggested that tubular injury may contribute to the pathological onset of DKD. We demonstrated a profibrotic effect of HG-treated TEC-derived exosomes on fibroblasts, suggesting a pathogenic role of exosomes in DKD by mediating tubulointerstitial communication and promoting renal fibrosis. Our previous study proved that exosomes derived from TECs could mediate tubulointerstitial communication between tubular cells and fibroblasts, which regulate the proliferation and activation of fibroblasts, contributing to the paracrine signaling mechanism responsible for renal fibrosis in DKD [34]. Liu et al. [35] confirmed the effect of TEC-derived exosomes in promoting renal interstitial fibrosis. They suggested that HNRNPA1-mediated exosomal sorting transported cellular miR-483-5p out of TECs into the urine, thus lessening the restraint of cellular miR-483-5p on MAPK1 and TIMP2 mRNAs and ultimately boosting ECM deposition and the progression of renal interstitial fibrosis in DKD. Moreover, exosomes derived from HG-treated TECs induced EMT in TECs by delivering Fibulin-1, which was modulated by miR-1269b [36]. Together, these studies indicate that TEC-derived exosomes have profibrotic functions in DKD, which could be a novel mechanism in renal fibrosis of DKD.

Podocyte-Derived Exosomes

Podocytes are highly specialized epithelial cells wrapping the glomeruli capillaries to prevent the leakage of proteins into urine [37]. Since these cells normally cannot proliferate, podocyte injury and malfunction lead to proteinuria, accumulation of ECM components, and eventually irreversible glomerulosclerosis. Sakurai et al. [38] reported that AGE induced the secretion of Elf3-

containing exosomes from podocytes via the TGF-β-Smad3 signaling pathway. Further study in DKD patients demonstrated that urinary exosomal Elf3 might be an early noninvasive marker for podocyte injury in DKD since its appearance correlated with the existence of irreversible podocyte injury. However, it remains unknown whether podocyte-derived exosomes induce cell-cell communication with other renal resident cells.

Macrophage-Derived Exosomes

Macrophages have essential roles in immune surveillance and the maintenance of kidney homeostasis. During kidney injury, macrophages adapt rapidly to the dynamics of the renal microenvironment [39]. Current evidence suggests a vital communicator role for exosomes derived from macrophages which mediate signaling from macrophages to GMCs or podocytes in DKD. For GMCs, TGF-β1 containing exosomes from macrophages could induce proliferation, fibrosis, and inflammation of GMCs by the TGF-β1/Smad3 pathway [40]. For podocytes, exosomal miR-25-3p from macrophages improved HG-induced podocyte injury by inhibiting DUSP1 expression with consequent autophagy activation [41]. Furthermore, HG-treated-macrophage-derived exosomes contained higher concentrations of IL-1β and iNOS, which activate macrophages by inducing the production of intracellular iNOS and α-SMA and accelerate kidney injury via the NF-κB signaling pathway [42].

MSC-Derived Exosomes

MSCs are a heterogeneous subset of stromal stem cells with multipotent and self-renewing properties and can differentiate into cells of the mesodermal lineage [43]. Recently, MSC-derived exosomes have attracted much attention for their ability to facilitate the renal repair and their potential for DKD therapy [44, 45]. On TECs, MSC-derived exosomes exerted multiple effects, such as anti-inflammation, anti-fibrosis, anti-apoptotic effect, activation of autophagy, and protection of tight junction structure in DKD [46, 47]. On podocytes, MSC-derived exosomes suppressed apoptosis, fibrosis, and proliferation [48–50]. In addition, MSC-derived exosomes play a renal-protective role in DKD by ameliorating fibrosis and inflammation [51–53], inducing autophagy [53], inhibiting apoptosis [54], and lowering blood [55]. The mechanisms by which MSC-derived exosomes exert their therapeutic effects are multifaceted, but in general, mainly by regulating the Akt, mTOR, P38-MAPK, JAK2/STAT3, and TGF-β signaling. Due to the multipotent and self-renewing characteristics of MSCs, their derived exosomes hold an excellent promise for DKD.

Other Resources of Exosomes in Kidneys

Exosomes can be secreted from various cell types into circulation and transported to target cells throughout the body. Gao et al. [56] isolated serum exosomes from DKD patients and found that such exosomes could promote pyroptosis and oxidative stress through the miR-4449/HIC1 pathway. Interestingly, Zhang et al. [57] injected miR-23a/27a in the muscle of DKD mice that could prevent diabetes-induced muscle cachexia and attenuate renal fibrosis lesions via exosome-mediated muscle-kidney crosstalk.

Implication of Urinary Exosomes as Biomarkers of DKD

Current biomarkers for detecting DKD, such as albuminuria and eGFR, are of insufficient sensitivity to detect the early-stage DKD, highlighting the necessity for discovering novel biomarkers. Exosomes can be easily obtained from different body fluids by noninvasive methods and have emerged as a promising tool for analyzing DKD biomarkers, especially for urinary exosomes [58]. In general, plasma exosomes may not pass through the glomerular filtration barrier, and as a result, urinary exosomes mainly come from urogenital system cells. Moreover, the exosomes are protected by their bilayer membrane structure. Thus, urinary exosomes mirror the physiopathological state of the kidney other than serum or circulation [59]. Herein, we summarized the implication of urinary exosomes as biomarkers in DKD (shown in Table 2).

Urinary Exosomal Proteins

Several urinary exosomal proteins have been implicated in DKD detection. The first proteomic study of human urinary exosomes for DKD patients suggested increased AMBP and MLL3 and decreased VDAC1 levels as biomarkers for diagnosing and pathological follow-up in DKD [60]. However, urinary exosomal WT1 was the first protein detected in DKD patients. It was associated with a significant increase in urine protein-to-creatinine ratio, albumin-to-creatinine ratio, serum creatinine, and a decline in eGFR [61]. WT1, shed by renal epithelial cells, has generally been regarded as a biomarker for podocyte injury. Another study showed that increased plasmin, prostasin, and urokinase directly correlated with proteinuria levels [62]. Elf3, a predictor of irreversible podocyte injuries, was also suggested as a biomarker of DKD with high specificity which is only detectable in urinary exosomes of DKD patients rather than DM patients or healthy controls [38]. Increased FBLN1 was associated with the severity of tubular injury [36], while

Table 2. Urinary exosomal biomarkers for DKD

Exosomal	Molecules	Species	Change	Actions	PMID
Proteins	WT1	Human	↑	Predict proteinuria and decreased renal function, especially for GFR <60 mL/min/1.73 m^2	23544132
	Plasmin, prostasin, and urokinase	Human	↑	Correlated directly with urine albumin	25609736
	Elf3	Human	↑	Elf3 was detectable only in DKD patients. Irreversible injuries in podocytes were predicted	31150422
	glcUMOD	Human	↑	Forms AGEs	28592554
	AMBP, MLL3	Human	↑	-	24211404
	FBLN1	Human	↑	Correlated with the severity of tubular injury	34977033
	AQP5, AQP2	Human	↑	Positive correlation between uAQP5 and the histological class of DN	28246612
	CD63	Human	↑	Therapeutic indicator	28116765
	Regucalcin, SMP30	Rat	↓	-	26072307
	VDAC1	Human	↓	-	24211404
miRNAs	let-7c-5p	Human	↑	Correlated with renal function, level of eGFR, and disease progression	29739042
	miR-19b-3p	Mice	↑	Correlated with the severity of albuminuria, tubulointerstitial inflammation, and SOCS-1 level	31097789
	miR-188-5p	Human	↑	Regulates HG-induced EMT in HK-2 cells via the PTEN/PI3K/Akt pathway	32340338
	miR-150-3p, miR-760, miR-3677-3p, miR-548ah-3p, miR-548p, miR-320e, miR-23c	Human	↑	-	32340338
	miR-21-5p, let-7e-5p, miR-23b-3p	Human	↑	Associated with renal function, renal sclerosis, fibrosis, detection of DKD, and poor renal function	31358876
	miR-4534	Human	↑	Involved in the FoxO/BNIP3 signaling pathway, correlated with proteinuria levels in DKD.	32982978
	miR-516b-5p	Human	↑	Correlated with proteinuria levels	32982978
	miR-1269b	Human	↑	Correlated with tubular injury	34977033
	miR-451	Human, Rat	↑	Correlated with renal failure	32322216
	miR-451-5p, miR-16	Rat	↑	Predict albuminuria at an earlier time point	27101382
	miR-320c, miR-6068	Human	↑	Graded correlation with UACR.	26930277
	miR-1234-5p, miR-6133, miR-4270, miR-4739, miR-371b-5p, miR-638, miR-572, miR-1227-5p, miR-6126, miR-1915-5p, miR-4778-5p, miR-2861	Human	↑	-	26930277
	miR-133b, miR-342, miR-30a	Human	↑	Early marker before the onset of albuminuria; associated with renal function and proteinuria levels	27470555
	miR-362-3p, miR-877-3p, miR-150-5p, miR-638, miR-133b	Human	↑	Early biomarker for DKD.	29038788
	miR-145, miR-130a	Human	↑	Early biomarker for DKD.	24223694
	miR-15b, miR-34a, and miR-636	Human	↑	Positive correlated with renal function and proteinuria levels	27475263
	miR-15b-5p	Human, mice	↑	Predicted kidney injury in DKD	31991106
	miR-15a-5p	Human	↓	Mediated fibrosis	29038788
	miR29c-5p, miR-15b-5p	Human	↓	Predicted the development of DKD	29739042

Table 2 (continued)

Exosomal	Molecules	Species	Change	Actions	PMID
	miR-30b-5p, miR-125b-5p	Human	↓	Detected DKD and poor renal function	31358876
	miR-133a-3p, miR-153-3p	Human	↓	Early biomarker for DKD	32340338
	miR-155, miR-424	Human	↓	Early biomarker for DKD	24223694
mRNA	WT1	Human	↑	Reflected damage of diabetic glomeruli, predicted the decline in eGFR	30282862

WT1, Wilms tumor 1; GFR, glomerular filtration rate; Elf3, E74-like ETS transcription factor 3; DKD, diabetic kidney disease; glcUMOD, glycated uromodulin; AGEs, advanced glycation end products; AMBP, α-microglobulin/bikunin precursor; MLL3, histone-lysine N-methyltransferase; FBLN1, Fibulin-1; AQP, aquaporins; uAQP5, urine aquaporins 5; SMP30, senescence marker protein-30; VDAC1, voltage-dependent anion-selective channel protein 1; SOCS1, suppressor of cytokine signaling 1; EMT, epithelial-mesenchymal transition; PTEN, phosphatase and tensin homologue; PI3K, phosphatidylinositol-3-kinase; FoxO, forkhead box O; BNIP3, BCL2 interacting protein 3; UACR, urine albumin-to-creatinine ratio.

AQP5 and AQP2 were associated with the histological class or degree of DKD [63]. Urinary glcUMOD concentration can predict DKD status, particularly in patients with CKD1-3a stages aged <65 years and with a urine glcUMOD concentration ≥9,000 arbitrary units [64]. In another study, urinary exosomal CD63 was higher in normoalbuminuria patients than microalbuminuria patients, and it might be a therapeutic indicator in DKD, especially after α-lipoic acid treatment [65]. By contrast, regucalcin and SMP30 were strongly downregulated in both DKD kidney tissue and urinary exosomes, suggesting their potential for early diagnosis and progression monitoring of DKD [66].

Urinary Exosomal miRNAs

By modulating gene expression, miRNAs influence various cellular and physiological processes, including proliferation, differentiation, cell death, and organ development. There is an increasing body of evidence that miRNAs are crucial regulators and biomarkers of DKD. Li et al. [67] showed an upregulation of let-7c-5p in urinary exosomes of type II diabetes, which was associated with the decline of renal function and DKD progression. In contrast, miR29c-5p and miR-15b-5p were downregulated, which could also predict the progression of DKD. Lv et al. [68] revealed that a high level of miR-19b-3p was associated with the severity of albuminuria, tubulointerstitial inflammation, and SOCS-1 levels. Both miR-4534 and miR-516b-5p were upregulated and correlated with proteinuria levels in DKD, an observation probably related to the involvement of miR-4534 in the FoxO/BNIP3 signaling pathway [69]. Increased miR-21-5p, let-7e-5p, miR-23b-3 and decreased miR-30b-5p, miR-125b-5p were associated with renal function, renal sclerosis, fibrosis, which could detect DKD and reduced renal function [70]. Furthermore, miR-1269b was correlated with tubular injury, and miR-451 was correlated with renal failure, while an increased level of miR-451-5p or miR-16 might predict albuminuria at an earlier time point [36, 71, 72].

Lee et al. conducted the next-generation sequencing to analyze miRNAs in urinary exosomes of DKD and found that miR-188-5p, miR-150-3p, miR-760, miR-3677-3p, miR-548ah-3p, miR-548p, miR-320e, and miR-23c were significantly upregulated in DKD patients, while miR-133a-3p and miR-153-3p were downregulated. Functionally, they demonstrated that miR-188-5p promoted EMT in HG-treated HK-2 cells through the PTEN/PI3K/Akt pathway [73]. Delic and colleagues [74] analyzed urinary exosomes of type II DKD patients by Agilent's miRNA microarrays, showing the upregulation of 14 miRNAs (miR-320c, miR-6068, miR-1234-5p, miR-6133, miR-4270, miR-4739, miR-371b-5p, miR-638, miR-572, miR-1227-5p, miR-6126, miR-1915-5p, miR-4778-5p, and miR-2861) and downregulation of miR-30d-5p and miR-30e-5p. Further investigation showed the correlation of miR-320c and miR-6068 with UACR levels in these patients, but the function of these miRNAs was not explored.

Other studies also suggested specific urinary exosomal miRNAs as early biomarkers for DKD, including upregulated miR-133b, miR-342, miR-30a, miR-362-3p, miR-877-3p, miR-150-5p, miR-638, miR-133b, miR-145, miR-130a and downregulated miR-155, miR-424, miR-133a-3p, miR-153-3p [73, 75–77]. Moreover, some miRNAs were revealed to be correlated with renal function and proteinuria levels, such as miR-15b, miR-34a, miR-636, miR-15b-5p, miR-30b-5p, and miR-125b-5p [70, 78, 79].

Besides the protein level of WT1, its mRNA could also predict renal functional decline and damage of diabetic glomeruli in DKD patients [80]. Recently, one study

compared the differences of mRNAs, miRNAs, lncRNAs, and circRNAs profiles in exosomes derived from HG-treated human TECs for 48 h and revealed differential expression with 169 lncRNAs, 885 mRNAs, 3 circRNAs, and 152 miRNAs; however, no validation experiments were proceeded [81]. Therefore, urinary exosomal proteins and miRNAs may be novel noninvasive biomarkers for DKD. Further clinical trials should investigate their biomarker efficacy and test the possible combination use of multiple biomarkers for early detection of DKD.

Biogenesis of Exosomes in DKD

Compared with the research on the cargo and function of exosomes, very little is known about the biogenesis or particle number of exosomes in DKD. Nonetheless, we recently reported a significant decrease in exosome secretion in DKD models, including both Akita diabetic mice and HG-treated renal tubular cells [34]. Mechanistically, FOXO1 was shown to be phosphorylated and inactivated in DKD, leading to the downregulation of RAB27B, a key regulator of exosome secretion [82]. Consistently, Hassanpour et al. [83] showed that chronic exposure of human endothelial progenitor cells to diabetic sera suppressed exosome secretion or biogenesis. However, there are studies showing a higher particle number of exosomes in DKD models. For example, the numbers of exosomes released from GECs treated with HG for 24 h were 1.5–3 times higher than the normal-glucose-treated group [26–28]. The cause of the discrepancy between these studies is unclear, but it may be related to the experimental conditions, including the cell and tissue types and the duration of diabetic exposure. In this regard, longer than 1-week exposure of cells to the diabetic condition [83] yielded results that are consistent with in vivo model of weeks of diabetes [34, 82]. Of note, DKD is a chronic progressive disease. Thus, an in vitro model with a longer diabetic exposure time may better reflect the natural course of DKD in human patients.

Conclusion

Exosomes mediate communication by autocrine or paracrine mechanisms between different kidney resident cells. Recent studies have demonstrated the changes of exosomes in DKD, including both exosomal cargos and numbers. Functionally, exosomes have been implicated in the pathogenesis and development of DKD. Despite these progresses, important questions mainly open in this field. For example, since an exosome contains multiple cargo molecules, it is interesting and meaningful to clarify the role of specific exosomal cargos in DKD, which may lead to the identification of novel therapeutic targets for DKD. In addition, how do cells control the content and number of cargos inside exosomes and what influences the binding of exosomes with specific kidney cell types in DKD? Addressing this question is critical to understanding the function of exosomes in DKD. Also, we have just begun to understand how kidney cells control the number of exosomes or exosome secretion in DKD. As eluded above, there may be a biphasic change in exosome secretion after diabetic exposure. In the early time, the exosome section may be increased, but chronic exposure to diabetic conditions leads to a significant decrease in exosome secretion. Certainly, exosome secretion can be regulated by RAB27B and FOXO1 [82], but it also depends on cell types and cellular conditions.

Strikingly, urinary exosomes showed a particular advantage as biomarkers for DKD. miRNAs and proteins in urinary exosomes are the most studied molecules that are indicative of renal function and disease progression in DKD. However, it is noteworthy that some of them might not be specific for DKD but general CKD. Furthermore, most of these studies did not compare the efficacy of biomarker candidates with albuminuria except for WT1 protein [61]. Further studies need to validate the clinical applicability of these potential biomarkers. Moreover, further research is encouraged to test the diagnostic potential or prognostic role of other exosomal components in DKD, besides miRNAs and proteins.

Conflict of Interest Statement

The authors have no conflicts of interest to declare.

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Author Contributions

Conceptualization: J.W. and Z.D.; original draft preparation: J.W.; review and editing: J.W. M.Z., Y.Y., Y.L., P.F., and Z.D.

References

1 CfDCa Prevention. Chronic kidney disease in the United States, 2021. Atlanta, GA: US Department of Health and Human Services; 2021 March 4. p. 2021.
2 GBD Chronic Kidney Disease Collaboration. Global, regional, and national burden of chronic kidney disease, 1990-2017: a systematic analysis for the Global Burden of Disease Study 2017. Lancet. 2020;395(10225):709–33.
3 Yang C, Gao B, Zhao X, Su Z, Sun X, Wang HY, et al. Executive summary for China kidney disease network (CK-NET) 2016 annual data report. Kidney Int. 2020;98(6):1419–23.
4 Reutens AT. Epidemiology of diabetic kidney disease. Med Clin North Am. 2013;97(1):1–18.
5 Lu Y, Liu D, Feng Q, Liu Z. Diabetic nephropathy: perspective on extracellular vesicles. Front Immunol. 2020;11:943.
6 Chen J, Zhang Q, Liu D, Liu Z. Exosomes: advances, development and potential therapeutic strategies in diabetic nephropathy. Metabolism. 2021;122:154834.
7 Pan BT, Johnstone RM. Fate of the transferrin receptor during maturation of sheep reticulocytes in vitro: selective externalization of the receptor. Cell. 1983;33(3):967–78.
8 Johnstone RM, Adam M, Hammond JR, Orr L, Turbide C. Vesicle formation during reticulocyte maturation. Association of plasma membrane activities with released vesicles (exosomes). J Biol Chem. 1987;262(19):9412–20.
9 Raposo G, Nijman HW, Stoorvogel W, Liejendekker R, Harding CV, Melief CJ, et al. B lymphocytes secrete antigen-presenting vesicles. J Exp Med. 1996;183(3):1161–72.
10 Kalluri R, LeBleu VS. The biology, function, and biomedical applications of exosomes. Science. 2020;367(6478):eaau6977.
11 Tkach M, Thery C. Communication by extracellular vesicles: where we are and where we need to go. Cell. 2016;164(6):1226–32.
12 Pegtel DM, Gould SJ Annu Rev Biochem. 2019;88:487–514.
13 Kowal J, Tkach M, Thery C. Biogenesis and secretion of exosomes. Curr Opin Cell Biol. 2014;29:116–25.
14 Thery C, Zitvogel L, Amigorena S. Exosomes: composition, biogenesis and function. Nat Rev Immunol. 2002;2(8):569–79.
15 Colombo M, Moita C, van Niel G, Kowal J, Vigneron J, Benaroch P, et al. Analysis of ESCRT functions in exosome biogenesis, composition and secretion highlights the heterogeneity of extracellular vesicles. J Cell Sci. 2013;126(Pt 24):5553–65.
16 Ostrowski M, Carmo NB, Krumeich S, Fanget I, Raposo G, Savina A, et al. Rab27a and Rab27b control different steps of the exosome secretion pathway. Nat Cell Biol. 2010;12(1):19–30; sup pp 1-13.
17 Lin J, Li J, Huang B, Liu J, Chen X, Chen XM, et al. Exosomes: novel biomarkers for clinical diagnosis. ScientificWorldJournal. 2015;2015:657086.
18 Willms E, Johansson HJ, Mager I, Lee Y, Blomberg KEM, Sadik M, et al. Cells release subpopulations of exosomes with distinct molecular and biological properties. Sci Rep. 2016;6:22519.
19 Hessvik NP, Llorente A. Current knowledge on exosome biogenesis and release. Cell Mol Life Sci. 2018;75(2):193–208.
20 Kowal J, Arras G, Colombo M, Jouve M, Morath JP, Primdal-Bengtson B, et al. Proteomic comparison defines novel markers to characterize heterogeneous populations of extracellular vesicle subtypes. Proc Natl Acad Sci U S A. 2016;113(8):E968–77.
21 Simons M, Raposo G. Exosomes – vesicular carriers for intercellular communication. Curr Opin Cell Biol. 2009;21(4):575–81.
22 Mathieu M, Martin-Jaular L, Lavieu G, Thery C. Specificities of secretion and uptake of exosomes and other extracellular vesicles for cell-to-cell communication. Nat Cell Biol. 2019;21(1):9–17.
23 Lv LL, Feng Y, Tang TT, Liu BC. New insight into the role of extracellular vesicles in kidney disease. J Cell Mol Med. 2019;23(2):731–9.
24 Zhang W, Zhou X, Zhang H, Yao Q, Liu Y, Dong Z. Extracellular vesicles in diagnosis and therapy of kidney diseases. Am J Physiol Renal Physiol. 2016;311(5):F844–51.
25 Ramnath RD, Satchell SC. Glomerular endothelial cells: assessment of barrier properties in vitro. Methods Mol Biol. 2020;2067:145–51.
26 Wu X, Gao Y, Xu L, Dang W, Yan H, Zou D, et al. Exosomes from high glucose-treated glomerular endothelial cells trigger the epithelial-mesenchymal transition and dysfunction of podocytes. Sci Rep. 2017;7(1):9371.
27 Wu XM, Gao YB, Cui FQ, Zhang N. Exosomes from high glucose-treated glomerular endothelial cells activate mesangial cells to promote renal fibrosis. Biol Open. 2016;5(4):484–91.
28 Ling L, Tan Z, Zhang C, Gui S, Cui Y, Hu Y, et al. CircRNAs in exosomes from high glucose-treated glomerular endothelial cells activate mesangial cells. Am J Transl Res. 2019;11(8):4667–82.
29 Wu XM, Gao YB, Xu LP, Zou DW, Zhu ZY, Wang XL, et al. Tongxinluo inhibits renal fibrosis in diabetic nephropathy: involvement of the suppression of intercellular transfer of TGF-[formula: see text]1-containing exosomes from GECs to GMCs. Am J Chin Med. 2017;45(5):1075–92.
30 Wang YY, Tang LQ, Wei W. Berberine attenuates podocytes injury caused by exosomes derived from high glucose-induced mesangial cells through TGFβ1-PI3K/AKT pathway. Eur J Pharmacol. 2018;824:185–92.
31 Bai S, Xiong X, Tang B, Ji T, Li X, Qu X, et al. Exosomal circ_DLGAP4 promotes diabetic kidney disease progression by sponging miR-143 and targeting ERBB3/NF-κB/MMP-2 axis. Cell Death Dis. 2020;11(11):1008.
32 da Silva Novaes A, Borges FT, Maquigussa E, Varela VA, Dias MVS, Boim MA. Influence of high glucose on mesangial cell-derived exosome composition, secretion and cell communication. Sci Rep. 2019;9(1):6270.
33 Zhu Y, Zha F, Tang B, Ji TT, Li XY, Feng L, et al. Exosomal hsa_circ_0125310 promotes cell proliferation and fibrosis in diabetic nephropathy via sponging miR-422a and targeting the IGF1R/p38 axis. J Cell Mol Med. 2022;26(1):151–62.
34 Wen J, Ma Z, Livingston MJ, Zhang W, Yuan Y, Guo C, et al. Decreased secretion and profibrotic activity of tubular exosomes in diabetic kidney disease. Am J Physiol Renal Physiol. 2020;319(4):F664–73.
35 Liu D, Liu F, Li Z, Pan S, Xie J, Zhao Z, et al. HNRNPA1-mediated exosomal sorting of miR-483-5p out of renal tubular epithelial cells promotes the progression of diabetic nephropathy-induced renal interstitial fibrosis. Cell Death Dis. 2021;12(3):255.
36 Tsai Y-C, Hung W-W, Chang W-A, Wu P-H, Wu L-Y, Lee S-C, et al. Autocrine exosomal fibulin-1 as a target of MiR-1269b induces epithelial-mesenchymal transition in proximal tubule in diabetic nephropathy.Front Cell Dev Biol. 2021;9:789716.
37 Scott RP, Quaggin SE. Review series: the cell biology of renal filtration. J Cell Biol. 2015;209(2):199–210.
38 Sakurai A, Ono H, Ochi A, Matsuura M, Yoshimoto S, Kishi S, et al. Involvement of Elf3 on Smad3 activation-dependent injuries in podocytes and excretion of urinary exosome in diabetic nephropathy. PLoS One. 2019;14(5):e0216788.
39 Tang PMK, Nikolic-Paterson DJ, Lan HY. Macrophages: versatile players in renal inflammation and fibrosis. Nat Rev Nephrol. 2019;15(3):144–58.
40 Zhu Q-J, Zhu M, Xu X-X, Meng X-M, Wu Y-G. Exosomes from high glucose-treated macrophages activate glomerular mesangial cells *via* TGF-β1/Smad3 pathway *in vivo* and *in vitro*. FASEB J. 2019;33(8):9279–90.
41 Huang H, Liu H, Tang J, Xu W, Gan H, Fan Q, et al. M2 macrophage-derived exosomal miR-25-3p improves high glucose-induced podocytes injury through activation autophagy via inhibiting DUSP1 expression. IUBMB Life. 2020;72(12):2651–62.
42 Zhu M, Sun X, Qi X, Xia L, Wu Y. Exosomes from high glucose-treated macrophages activate macrophages andinduce inflammatory responses via NF-κB signaling pathway in vitro and in vivo. Int Immunopharmacol. 2020;84:106551.
43 Uccelli A, Moretta L, Pistoia V. Mesenchymal stem cells in health and disease. Nat Rev Immunol. 2008;8(9):726–36.

44 Hickson LJ, Eirin A, Conley SM, Taner T, Bian X, Saad A, et al. Diabetic kidney disease alters the transcriptome and function of human adipose-derived mesenchymal stromal cells but maintains immunomodulatory and paracrine activities important for renal repair. Diabetes. 2021;70(7):1561–74.
45 Abdel Aziz MT, Wassef MAA, Ahmed HH, Rashed L, Mahfouz S, Aly MI, et al. The role of bone marrow derived-mesenchymal stem cells in attenuation of kidney function in rats with diabetic nephropathy. Diabetol Metab Syndr. 2014;6(1):34.
46 Nagaishi K, Mizue Y, Chikenji T, Otani M, Nakano M, Konari N, et al. Mesenchymal stem cell therapy ameliorates diabetic nephropathy via the paracrine effect of renal trophic factors including exosomes. Sci Rep. 2016;6:34842.
47 Cai X, Zou F, Xuan R, Lai XY. Exosomes from mesenchymal stem cells expressing microribonucleic acid-125b inhibit the progression of diabetic nephropathy via the tumour necrosis factor receptor-associated factor 6/Akt axis. Endocr J. 2021;68(7):817–28.
48 Jin J, Wang Y, Zhao L, Zou W, Tan M, He Q. Exosomal miRNA-215-5p derived from adipose-derived stem cells attenuates epithelial-mesenchymal transition of podocytes by inhibiting ZEB2. Biomed Res Int. 2020;2020:2685305.
49 Jiang ZZ, Liu YM, Niu X, Yin JY, Hu B, Guo SC, et al. Exosomes secreted by human urine-derived stem cells could prevent kidney complications from type I diabetes in rats. Stem Cell Res Ther. 2016;7:24.
50 Duan YR, Chen BP, Chen F, Yang SX, Zhu CY, Ma YL, et al. Exosomal microRNA-16-5p from human urine-derived stem cells ameliorates diabetic nephropathy through protection of podocyte. J Cell Mol Med. 2021;25(23):10798–813.
51 Xiang E, Han B, Zhang Q, Rao W, Wang Z, Chang C, et al. Human umbilical cord-derived mesenchymal stem cells prevent the progression of early diabetic nephropathy through inhibiting inflammation and fibrosis. Stem Cell Res Ther. 2020;11(1):336.
52 Hao Y, Miao J, Liu W, Cai K, Huang X, Peng L. Mesenchymal stem cell-derived exosomes carry MicroRNA-125a to protect against diabetic nephropathy by targeting histone deacetylase 1 and downregulating endothelin-1. Diabetes Metab Syndr Obes. 2021;14:1405–18.
53 Ebrahim N, Ahmed IA, Hussien NI, Dessouky AA, Farid AS, Elshazly AM, et al. Mesenchymal stem cell-derived exosomes ameliorated diabetic nephropathy by autophagy induction through the mTOR signaling pathway. Cells. 2018;7(12):226.
54 Mao R, Shen J, Hu X. BMSCs-derived exosomal microRNA-let-7a plays a protective role in diabetic nephropathy via inhibition of USP22 expression. Life Sci. 2021;268:118937.
55 Wang S, Bao L, Fu W, Deng L, Ran J. Protective effect of exosomes derived from bone marrow mesenchymal stem cells on rats with diabetic nephropathy and its possible mechanism. Am J Transl Res. 2021;13(6):6423–30.
56 Gao C, Wang B, Chen Q, Wang M, Fei X, Zhao N. Serum exosomes from diabetic kidney disease patients promote pyroptosis and oxidative stress through the miR-4449/HIC1 pathway. Nutr Diabetes. 2021;11(1):33.
57 Zhang A, Li M, Wang B, Klein JD, Price SR, Wang XH. miRNA-23a/27a attenuates muscle atrophy and renal fibrosis through muscle-kidney crosstalk. J Cachexia Sarcopenia Muscle. 2018;9(4):755–70.
58 Lin CH, Chang YC, Chuang LM. Early detection of diabetic kidney disease: present limitations and future perspectives. World J Diabetes. 2016;7(14):290–301.
59 Gudehithlu KP, Garcia-Gomez I, Vernik J, Brecklin C, Kraus M, Cimbaluk DJ, et al. In diabetic kidney disease urinary exosomes better represent kidney specific protein alterations than whole urine. Am J Nephrol. 2015;42(6):418–24.
60 Zubiri I, Posada-Ayala M, Sanz-Maroto A, Calvo E, Martin-Lorenzo M, Gonzalez-Calero L, et al. Diabetic nephropathy induces changes in the proteome of human urinary exosomes as revealed by label-free comparative analysis. J Proteomics. 2014;96:92–102.
61 Kalani A, Mohan A, Godbole MM, Bhatia E, Gupta A, Sharma RK, et al. Wilm's tumor-1 protein levels in urinary exosomes from diabetic patients with or without proteinuria. PLoS One. 2013;8(3):e60177.
62 Andersen H, Friis UG, Hansen PBL, Svenningsen P, Henriksen JE, Jensen BL. Diabetic nephropathy is associated with increased urine excretion of proteases plasmin, prostasin and urokinase and activation of amiloride-sensitive current in collecting duct cells. Nephrol Dial Transplant. 2015;30(5):781–9.
63 Rossi L, Nicoletti MC, Carmosino M, Mastrofrancesco L, Di Franco A, Indrio F, et al. Urinary excretion of kidney aquaporins as possible diagnostic biomarker of diabetic nephropathy. J Diabetes Res. 2017;2017:4360357.
64 Chang CC, Chen CY, Huang CH, Wu CL, Wu HM, Chiu PF, et al. Urinary glycated uromodulin in diabetic kidney disease. Clin Sci. 2017;131(15):1815–29.
65 Sun H, Yao W, Tang Y, Zhuang W, Wu D, Huang S, et al. Urinary exosomes as a novel biomarker for evaluation of alpha-lipoic acid's protective effect in early diabetic nephropathy. J Clin Lab Anal. 2017;31(6):e22129.
66 Zubiri I, Posada-Ayala M, Benito-Martin A, Maroto AS, Martin-Lorenzo M, Cannata-Ortiz P, et al. Kidney tissue proteomics reveals regucalcin downregulation in response to diabetic nephropathy with reflection in urinary exosomes. Transl Res. 2015;166(5):474–84.e4.
67 Li W, Yang S, Qiao R, Zhang J. Potential value of urinary exosome-derived let-7c-5p in the diagnosis and progression of type II diabetic nephropathy. Clin Lab. 2018;64(5):709–18.
68 Lv LL, Feng Y, Wu M, Wang B, Li ZL, Zhong X, et al. Exosomal miRNA-19b-3p of tubular epithelial cells promotes M1 macrophage activation in kidney injury. Cell Death Differ. 2020;27(1):210–26.
69 Zhao Y, Shen A, Guo F, Song Y, Jing N, Ding X, et al. Urinary exosomal MiRNA-4534 as a novel diagnostic biomarker for diabetic kidney disease. Front Endocrinol. 2020;11:590.
70 Zang J, Maxwell AP, Simpson DA, McKay GJ. Differential expression of urinary exosomal MicroRNAs miR-21-5p and miR-30b-5p in individuals with diabetic kidney disease. Sci Rep. 2019;9(1):10900.
71 Mohan A, Singh RS, Kumari M, Garg D, Upadhyay A, Ecelbarger CM, et al. Urinary exosomal microRNA-451-5p is a potential early biomarker of diabetic nephropathy in rats. PLoS One. 2016;11(4):e0154055.
72 Kumari M, Mohan A, Ecelbarger CM, Gupta A, Prasad N, Tiwari S. miR-451 loaded exosomes are released by the renal cells in response to injury and associated with reduced kidney function in human. Front Physiol. 2020;11:234.
73 Lee WC, Li LC, Ng HY, Lin PT, Chiou TTY, Kuo WH, et al. Urinary exosomal MicroRNA signatures in nephrotic, biopsy-proven diabetic nephropathy. J Clin Med. 2020;9(4):1220.
74 Delić D, Eisele C, Schmid R, Baum P, Wiech F, Gerl M, et al. Urinary exosomal miRNA signature in type II diabetic nephropathy patients. PLoS One. 2016;11(3):e0150154.
75 Eissa S, Matboli M, Bekhet MM. Clinical verification of a novel urinary microRNA panal: 133b, -342 and -30 as biomarkers for diabetic nephropathy identified by bioinformatics analysis. Biomed Pharmacother. 2016;83:92–9.
76 Xie Y, Jia Y, Cuihua X, Hu F, Xue M, Xue Y. Urinary exosomal MicroRNA profiling in incipient type 2 diabetic kidney disease. J Diabetes Res. 2017;2017:6978984.
77 Barutta F, Tricarico M, Corbelli A, Annaratone L, Pinach S, Grimaldi S, et al. Urinary exosomal microRNAs in incipient diabetic nephropathy. PLoS One. 2013;8(11):e73798.
78 Tsai YC, Kuo MC, Hung WW, Wu LY, Wu PH, Chang WA, et al. High glucose induces mesangial cell apoptosis through miR-15b-5p and promotes diabetic nephropathy by extracellular vesicle delivery. Mol Ther. 2020;28(3):963–74.

79 Eissa S, Matboli M, Aboushahba R, Bekhet MM, Soliman Y. Urinary exosomal microRNA panel unravels novel biomarkers for diagnosis of type 2 diabetic kidney disease. J Diabetes Complications. 2016;30(8):1585–92.
80 Abe H, Sakurai A, Ono H, Hayashi S, Yoshimoto S, Ochi A, et al. Urinary exosomal mRNA of WT1 as diagnostic and prognostic biomarker for diabetic nephropathy. J Med Invest. 2018;65(3.44):208–15.
81 Zhou S, Fang J, Hu M, Pan S, Liu D, Xing G, et al. Determining the influence of high glucose on exosomal lncRNAs, mRNAs, circRNAs and miRNAs derived from human renal tubular epithelial cells. Aging. 2021;13(6):8467–80.
82 Zeng M, Wen J, Ma Z, Xiao L, Liu Y, Kwon S, et al. FOXO1-Mediated downregulation of RAB27B leads to decreased exosome secretion in diabetic kidneys. Diabetes. 2021;70(7):1536–48.
83 Hassanpour M, Cheraghi O, Brazvan B, Hiradfar A, Aghamohammadzadeh N, Rahbarghazi R, et al. Chronic exposure of human endothelial progenitor cells to diabetic condition abolished the regulated kinetics activity of exosomes. Iran J Pharm Res. 2018;17(3):1068–80.