Kidney Diseases

Review Article

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Impact of Angiopoietin-2 on Kidney Diseases

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Abstract
Background: Angiopoietins (Ang) are essential angiogenic factors involved in angiogenesis, vascular maturation, and inflammation. The most studied angiopoietins, angiopoietin-1 (Ang-1) and angiopoietin-2 (Ang-2), behave antagonistically to each other in vivo to sustain vascular endothelium homeostasis. While Ang-1 typically acts as the endothelium-protective mediator, its context-dependent antagonist Ang-2 can promote endothelium permeability and vascular destabilization, hence contributing to a poor outcome in vascular diseases via endothelial injury, vascular dysfunction, and microinflammation. The pathogenesis of kidney diseases is associated with endothelial dysfunction and chronic inflammation in renal diseases. ***Summary:*** Several preclinical studies report overexpression of Ang-2 in renal tissues of certain kidney disease models; additionally, clinical studies show increased levels of circulating Ang-2 in the course of chronic kidney disease, implying that Ang-2 may serve as a useful biomarker in these patients. However, the exact mechanisms of Ang-2 action in renal diseases remain unclear. ***Key Messages:*** We summarized the recent findings on Ang-2 in kidney diseases, including preclinical studies and clinical studies, aiming to provide a systematic understanding of the role of Ang-2 in these diseases.

Introduction

Vascular homeostasis plays a pivotal role in many diseases such as atherosclerosis, neurodegeneration, cancer, age-related cognitive decline [1], and kidney disease [2]. This homeostasis is balanced by angiogenic factors, such as vascular endothelial growth factor (VEGF), vascular endothelial growth factor receptor 1 (VEGFR1), and angiopoietin-1 (Ang-1), and antiangiogenic factors, such as VEGFR2 and angiopoietin-2 (Ang-2).

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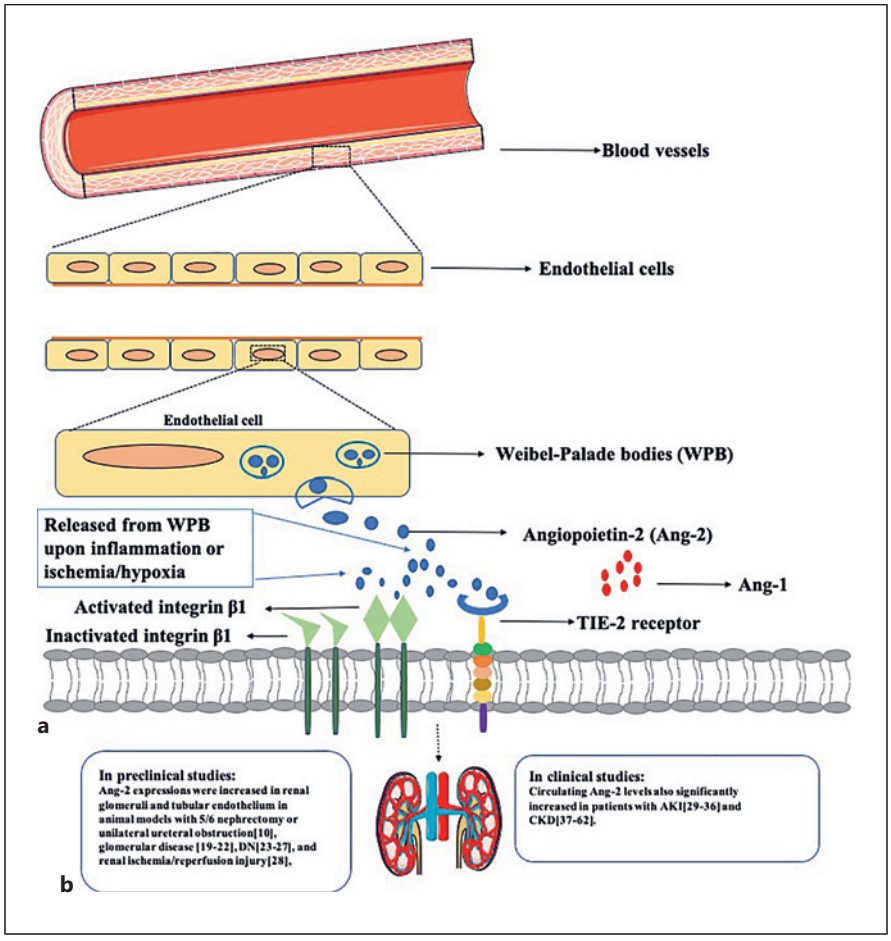

Fig. 1. a, b The process of Ang-2 from generation and function. The figure depicts that Ang-2 is partly derived from ECs of blood vessels. Ang-2 is rapidly released into circulation upon stimuli of inflammation or ischemia/hypoxia. Ang-2 competes for the TIE-2 receptor with Ang-1 and can also bind to activated integrin β1 to elicit biological function. **a** Ang-2 expression in kidney tissue was shown to be significantly upregulated in preclinical kidney disease models. **b** Circulating Ang-2 levels are markedly increased in patients with various kidney diseases based on preclinical studies and clinical studies. Ang-2, angiopoietin-2; Ang-1, angiopoietin-1; AKI, acute kidney injury; ADPKD, autosomal dominant polycystic kidney disease; GCN, chronic glomerulonephritis; CKD, chronic kidney disease; DN, diabetic nephropathy; DM, diabetes mellitus; ECs, endothelial cells; GN, glomerulonephritis; LN, lupus nephritis.

Angiopoietin growth factors participate in vascular development and repair through the endothelial Tie receptor tyrosine kinases. Ang-1 and Ang-2 are two major angiopoietin isoforms regulating vascular homeostasis. Ang-1 plays a pivotal role in eliciting an anti-inflammatory response in endothelial cells (ECs) and stabilizing the vessel wall by binding to Tie2 receptors and inducing Tie2 activation (autophosphorylation). However, Ang-2 has the opposite actions to Ang-1, acting as a proinflammatory natural antagonist for Ang-1 and its Tie2 receptor [3]. Ang-2 is mainly produced by ECs [4] and stored in endothelial Weibel-Palade bodies [5], being rapidly released into circulation upon various stimuli such as inflammation or ischemia/hypoxia [6] (Fig. 1a). Ang-2 acts as a competitive inhibitor of the Ang-1-induced phosphorylation and activation of the Tie2 receptor and also binds to activated integrin β1 to promote endothelium permeability and blood vessel wall destabilization [7]. Furthermore, Ang-2 stimulates ECs migration and proliferation, as well as promotes neovascularization in synchronous action with vascular endothelial growth factor-A (VEGF-A) [8]. In this regard, it is reported that the biological effects of Ang-2 depend on the levels of VEGFA in the microenvironment, leading to vessel regression at low levels of surrounding VEGFA, while acting proangiogenic in the presence of high VEGF-A levels [3, 9].

Many traditional risk factors, such as cardiovascular disease, diabetes mellitus (DM), hypertension, and hyperlipidemia, significantly contribute to high morbidity and mortality in chronic kidney disease (CKD). However, even when these traditional indicators are controlled, actual mortality exceeds expected mortality [10]. In this regard, much attention has been paid to nontraditional risk factors of poor renal outcomes, including endothelial dysfunction and inflammation. According to previous studies, endothelial dysfunction was observed in CKD [11] and was regarded as a key risk factor for developing cardiovascular events in CKD patients [12–14], even in predialysis CKD patients [15]. Ang-2, a key regulator of

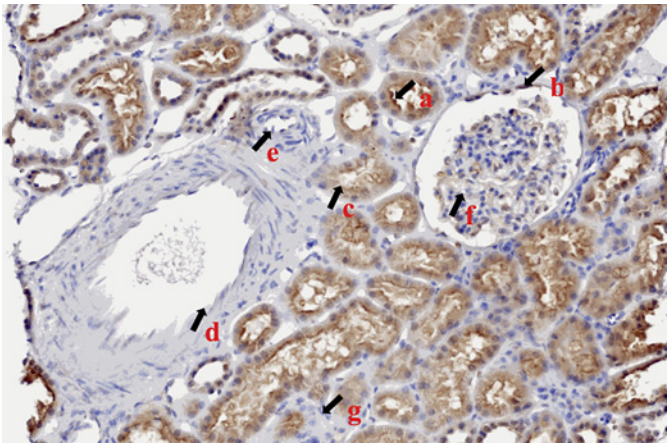

Fig. 2. Ang-2 immunostaining in normal kidney biopsy. Immunohistochemical expression of Ang-2 in normal human kidney tissue. Brown indicates Ang-2 positive. Physiological Ang-2 expression in tubular epithelial cells (arrow a shows strong Ang-2 positivity of the brush border, and arrow c shows moderate cytoplasmatic Ang-2 positivity) next to a focal weak Ang-2 positivity of glomerular parietal epithelial cells (arrow b shows Bowman's capsule) may be observed. No specific Ang-2 staining can be detected in arteries (arrows d, e), glomerular capillaries (arrow f), or interstitium (arrow g). Immunohistochemistry was performed on paraffin-embedded renal biopsy samples from a healthy biopsy in the University Medical Centre Mannheim using a primary antibody (Angiopoietin 2 Polyclonal Antibody [DF6137]). Image acquisition was done using a PreciPoint scanning microscope (using objective ×40/0.65 NA) and MicroPoint software (v.2016-02-05; PreciPoint, Freising, Germany).

angiogenesis, involving endothelial remodeling and inflammation, plays a crucial role in context-dependent effects on ECs. Previously, animal studies using transgenic mice demonstrated the lethal phenotype of Ang-2 overexpression, suggesting that increased circulating Ang-2 levels are probably a harmful sign, which is similar to the phenomenon seen in Ang1 and Tie2 knockouts [3, 16]. Generally, physiological serum levels of Ang-1 exceed those of Ang-2 [8], which maintains vessel wall stabilization. Interestingly, in both preclinical and clinical studies of various renal diseases, markedly increased levels of Ang-2 have been reported (Fig. 1b). However, the molecular mechanisms of Ang-2 are complex and its role in the pathogenesis of CKD remains unclear.

Our preliminary findings in human renal biopsies with normal histology showed strong and moderate immunohistochemical positivity for Ang-2 in the brush border and cytoplasm of proximal tubular epithelia, respectively, followed by focal weak staining in glomerular partial epithelial cells of Bowman's capsule and absence in blood vessels and interstitium (Fig. 2). In this review, we mainly focused on renal diseases and summarized the published data from animal and clinical studies, intending to provide a relatively comprehensive overview of the current knowledge on the role of Ang-2 in these disease settings.

Ang-2 in Kidney Diseases

Renal diseases, especially CKD, with their worldwide prevalence of 10–13% are on the rise, mainly due to the accelerated aging population, lifestyle changes, and increasing prevalence of obesity [17]. CKD is progressive, irreversible, and a common risk factor for cardiovascular-related disease. Several causes such as DM, hypertension, CVD, and other kidney-related diseases contribute to the onset and progression of CKD [18]. Individuals with this pathology are most of the time asymptomatic and renal complications typically occur in more advanced stages. CKD is clinically defined by classical biomarkers, including estimated glomerular filtration rate (eGFR) and albuminuria. However, given the complicated clinical settings, these biological indicators are not sufficient to predict and quantify CKD-related outcomes for an individual patient. Accordingly, it is necessary to explore more efficient biomarkers combined with eGFR and albuminuria to guide the diagnosis, treatment, and predictive assessment of CKD.

It has been reported that Ang-2 expression was upregulated in animal models of kidney disease, including glomerular disease [19–22] and diabetic nephropathy (DN) [23–27]. Also, consistent findings were observed in renal damage models, such as in rats with renal ischemia-reperfusion (I/R) injury [28] and mice with unilateral ureteral obstruction or 5/6 subtotal nephrectomy (5/6Nx) [10]. In clinical practice, Ang-2 alterations are primarily discussed in patients with acute kidney injury (AKI) [29–36] or various etiologies of CKD [37–62]. These findings from previous preclinical studies and clinical studies demonstrated that Ang-2 may be a pathogenic factor promoting the onset and progression of AKI and CKD; meanwhile, a negative prognostic factor predicts the clinical outcomes.

Animal Models in Kidney Diseases

Glomerular Disease

In the mouse model of lupus nephritis (LN), Liu et al. [19] showed that the protein expression of Ang-2 in the kidney was upregulated by the means of Western blot and immunohistochemistry, suggesting that Ang-2 may be associated with the progression of LN. Employing a daunorubicin-induced progressive glomerulosclerosis rat

Table 1. Studies of Ang-2 in animal models of renal diseases

First author	Year of publication	Species	Renal disease model	Main findings
Fan-Chi Chang et al. [10]	2014	Mice	5/6 nephrectomy or unilateral ureteral obstruction	Plasma levels of Ang-2 increased and Ang-2 was markedly expressed in the tubular endothelium of fibrotic kidneys
Liu Xue et al. [19]	2019	Mice	LN	The expression of Ang-2 protein increased in renal glomeruli
Lu Yuanhang et al. [20]	2006	Rat	Glomerulosclerosis (related to podocyte injury)	The expression of Ang-2 protein in glomeruli and Ang2 mRNA increased in the group with podocyte injury and was positively correlated with 24 h UPPER
Haitao Yuan et al. [21]	2002	Mice	GN	Ang-2 immunostaining was detectable and prominent in diseased glomeruli and related to the loss of glomerular capillaries
Belinda Davis et al. [22]	2007	Mice	DN	Ang-2 was markedly upregulated in the tubular endothelium of fibrotic kidneys
Bishoy Rizkalla et al. [23]	2005	Rat	T2DM	Renal expression of Ang-2 increased
Kunihiro Ichinose et al. [24]	2006	Mice	T2DM	Renal expression of Ang-2 increased
Yoshihiko Yamamoto et al. [25]	2004	Mice	DN	The protein expression level of Ang-2 increased in the renal cortex of DN mice compared with healthy mice
Kunihiro Ichinose et al. [26]	2005	Mouse	T1DM	Increased expression of Ang-2 protein in the kidney
Luo Changqing et al. [27]	2014	Mice	DN	The expression of Ang-2 mRNA and protein levels greatly increased in STZ-induced diabetic mice; Ang-2 was mainly expressed in glomeruli
Meriem Khairoun et al. [28]	2013	Rat	renal ischemia/reperfusion injury	Protein expression of Ang-2 increased starting at 5 h until 72 h after renal I/R; increased Ang-2 expression accompanied by a loss of ECs

DN, diabetic nephropathy; ECs, endothelial cells; GN, glomerulonephritis; LN, lupus nephritis; STZ, streptozotocin; T1DM, type 1 diabetes mellitus; T2DM, type 2 diabetes mellitus; UPPER, urinary protein excretion rate.

model, Lu et al. [20] demonstrated an increased glomerular Ang-2 mRNA and protein expression, which positively correlated with 24-h urinary protein quantitative measurements (Table 1). The authors suggested that locally boosted Ang-2 expression may be a risk factor for the progress of glomerulosclerosis [20]. In mice models of immune-mediated glomerulonephritis (GN), Ang-2 staining was barely detectable in the glomerular capillary loops in the control group, where the animals did not suffer from immune-mediated GN [21]. However, glomerular Ang-2 immunostaining was detectable and prominent in affected glomeruli and some sclerotic glomeruli [21]. Meanwhile, in sections from the control group, both Ang-1 and VEGF-A immunostaining were detectable in glomeruli but were diminished in glomerular tufts in the GN group [21]. Davis et al. showed glomerular endothelial apoptosis and proteinuria increased significantly, while VEGF-A and nephrin proteins markedly decreased in the mice models of podocyte-specific Ang-2 overexpression [22]. In vitro, the addition of exogenous Ang-2 to isolated adult rat glomeruli significantly downregulated the levels of VEGF-A protein after incubating for 24 h [22]. These findings may provide insights into the pathogenesis of glomerular disease and proteinuria [22].

Based on the above, it can be hypothesized that upregulation of Ang-2 in glomerular disease models disturbs growth factor homeostasis. This could modulate glomerular and peritubular capillary remodeling, resulting in the loss of glomerular capillaries, which may be a molecular mechanism of increased Ang-2 contributing to the pathogenesis of glomerular diseases.

Experimental Diabetic Nephropathy

In 2005, Rizkalla et al. [23] generated rat models of experimental diabetes induced by streptozotocin. In this

experiment, some groups of rats were treated with an angiotensin type 1 receptor subtype (AT1) or angiotensin type 2 receptor subtype receptor antagonist [23]. The study demonstrated that both the Ang-2 gene and protein expression significantly increased in the kidney tissues of diabetic rats, and the increased Ang-2 levels were significantly attenuated by the AT1 receptor antagonist but not by the AT2 receptor antagonist [23]. Furthermore, it was shown that Ang-2 immunostaining was predominantly located in glomerular cells, including epithelium and endothelium, whereas less staining was observed in the vasa recta and dilated tubules [23]. In addition, Ang-2 staining was increased in glomerular ECs of untreated diabetic rats at week 4 of the experiment [23]. Similarly, Ichinose et al. [24] reported that renal expression of Ang-2 significantly increased in a mice model of obese type 2 diabetes. In an animal model of DN, Yamamoto et al. [25] demonstrated that expression levels of Ang-2 increased in the renal cortex of DN mice compared with healthy mice in immunoblotting analyses. This study also demonstrated that increased renal expression of Ang-2 was suppressed by the tumstatin peptide, which is an inhibitor of angiogenesis derived from type IV collagen with the ability to inhibit the neovascularization induced by VEGF-A in vivo [25]. Subsequently, several studies obtained consistent findings [26, 27]. In a model of type 1 DN induced by low-dose streptozotocin (STZ) injection, Ichinose and colleagues found that compared with nondiabetic mice, Ang-2 protein levels increased in diabetic mice. The increased Ang-2 protein expression in the kidney was able to be inhibited by the endostatin peptide, a potent angiogenesis inhibitor derived from type XVIII collagen [26]. Moreover, Luo et al. [27] observed that both Ang-2 mRNA and protein levels greatly increased in a DN model by STZ induction, when compared with nondiabetic mice. In vitro, Ang-2 protein levels were shown to significantly increase in cultured mouse ECs under high glucose conditions [27]. Additionally, it was demonstrated that increased Ang-2 levels in vivo and in vitro can be significantly reduced by Alprostadil (prostaglandin E1) treatment [27]. In this study, urinary protein excretion, creatinine clearance rate, and kidney/body weight ratio were significantly reduced after Alprostadil treatment in STZ-injected mice [27]. These outcomes probably indicate that decreased Ang-2 levels caused by Alprostadil may serve as a mechanism of Alprostadil eliciting kidney protection and improving renal function.

Globally, the most common cause of end-stage renal disease (ESRD) is DN and the pathogenesis of DN is complicated and overlapping. It is reported that angiogenic phenomena were probably associated with the progression of DN. Angiogenesis-related growth factors such as Ang-1, Ang-2, and VEGF-A have been involved in the development of DN. Hypothetically, the chronic inflammation induced by long-standing high glucose stimulation promotes glomerular ECs to excrete more Ang-2, resulting in dysfunction of the endothelium and angiogenesis, which supports the role of Ang-2 in DN.

Other Types of Renal Damage

To investigate the impact of I/R injury on the dynamics of Ang expression, as well as the relationship between I/R, pericytes, and the development of fibrosis, Khairoun et al. [28] conducted a study using I/R models of male Lewis rats. This I/R model employed unilateral renal ischemia for 45 min at first, followed by removal of the contralateral kidney, and finally sacrificed rats at various time points after reperfusion. In this study, researchers observed that protein expression of Ang-2 increased starting at 5 h after I/R and maintained an increasing tendency until up to 72 h, resulting in a higher Ang-2/Ang-1 ratio [28]. After recovery of renal function, the Ang-2/Ang-1 ratio returned to the baseline levels 9 weeks after I/R. The entire process was accompanied by dynamic changes in pericytes and cortical ECs [28]. The authors suggested that Ang-2 was an important factor in renal microvascular changes and fibrosis development [28]. Moreover, it has been shown that circulating Ang-2 also increased in mice models with unilateral ureteral obstruction or 5/6Nx [10]. Chang et al. [10] found that the transcript of Ang-2 was upregulated in the kidney, but downregulated in the lung and aorta after 5/6Nx. Additionally, the staining of Ang-2 showed a marked expression in fibrotic renal tubular epithelium and glomeruli in 5/6Nx mice [10]. This study demonstrated that Ang-2 was probably the link between renal fibrosis and arterial stiffness [10]. These findings provided an insight that targeting Ang-2 may be a novel treatment for CVD in CKD patients based on the mechanisms of reducing inflammation levels and collagen expression.

Overall, based on the preclinical studies outlined above, increased renal Ang-2 expression levels were observed in experimental models of kidney disease. It is reported that the expression of Ang-2 is upregulated during mouse kidney development, whereas, after birth, it is rarely detected in the glomeruli of adult normal kidneys [22]. Thus, a greatly increased Ang-2 expression in the adult glomeruli, as observed in some renal disease models, might indicate that Ang-2, while playing a pivotal role in normal nephrogenesis, is harmful when it is highly reexpressed in the context of adult kidney disease. One

possible explanation is that the increase of Ang-2 disturbs the balance between antiangiogenic and proangiogenic growth factors, leading to disorders of ECs and angiogenesis in the kidney. Although current research on inhibitors directly targeting Ang-2 is still scarce, focusing on the Ang system or even the entire angiogenic system might be promising [23, 25, 26].

Clinical Studies in Kidney Diseases

Acute Kidney Injury

AKI refers to a sudden decrease in kidney function that happens quickly, generally within a few hours or days. Diagnostic criteria for AKI are based on an increase in serum creatinine level and a decrease in urine output and are limited to 7 days in duration. AKI is commonly found in patients who are suffering from critical illness, but its pathogenesis remains complex. Recent studies suggest that renal endothelial damage and microvascular dysfunction may contribute to this pathogenesis, which is supported by studies showing associations between Ang-2 and AKI [29].

Researchers observed that blood Ang-2 can serve as a potent predictor for AKI onset. In critically ill patients, Robinson-Cohen et al. [30] demonstrated that higher Ang-2 concentrations implicated a higher risk of AKI incidents. Subsequently, a prospective study investigated the relationship between blood Ang-2 and the onset of severe AKI (defined based on the Kidney Disease Improving Global Outcomes [KDIGO] stage 2/3) in critically ill patients [29]. The authors demonstrated that increased blood Ang-2 levels indicated an increased risk of AKI/death in critically ill patients after adjusting for confounding factors [29]. A similar association was reported in another cohort study of critically ill patients, but only for patients with or at risk of acute respiratory distress syndrome [31]. In patients with coronavirus disease (COVID-19), higher circulating Ang-2 levels were associated with the development of severe AKI, and the need for renal replacement therapy (RRT) [32]. However, this study lacked further elucidation regarding the specificity of Ang-2 for COVID-19-associated AKI and the role of Ang-2 in COVID-19-related endothelial disorders.

In AKI patients, Ang-2 concentrations were demonstrated to increase over time after cardiac surgery compared to control patients. In the same study, Ang-2 plasma levels correlated positively with urinary levels of N-acetyl-beta-D-glucosaminidase, a classic marker of AKI [33] (Table 2). Several limitations need to be considered when explaining the findings of this study, including underestimated AKI incidence due to only using serum creatinine levels as diagnostic criteria of AKI, incomplete dataset, and uncertain urine specimen quality [33]. Furthermore, a positive correlation between increased Ang-2 levels and the need for RRT and mortality in patients with AKI has been observed in several studies [34, 35]. However, several characteristics of participants should be of note when interpreting its results. In the study conducted by Kümpers et al. [35], the study population only included AKI patients requiring RRT, with low numbers of RIFLE-Risk and Injury subjects, and lacking non-AKI patients. In addition, it has been shown that the Ang-1/Ang-2 ratio is negatively associated with the risk of CKD progression, heart failure, and mortality in patients with AKI [36]. Although this multicenter prospective study has been well designed, it did not explore other pathways associated with EC repair that may influence results, and how the Ang pathway interacts with other pathways. Moreover, some other events that can influence the development of CKD and heart failure were not documented in this study, such as recurrent AKI attacks and rehospitalization [36].

Based on the mentioned findings, circulating Ang-2 is currently regarded as a potential biomarker to indicate the onset and progression of AKI. Consequently, measuring Ang-2 levels might assist physicians in the future in taking early intervention for AKI patients before the onset of severe AKI, CKD, and other complications.

General Chronic Kidney Disease

In clinical practice, many studies reported that circulating Ang-2 levels were increased significantly in CKD patients [37–39] and positively associated with CKD classification [38, 39]. Following these findings, researchers further explored more details on the association of Ang-2 with CKD-related markers, eGFR, and all-cause mortality in CKD. In a cohort of 416 CKD patients at stages 3–5, Chang et al. [37] reported plasma Ang-2 levels to be independently correlated with urine albumin-creatinine ratio and sensitive C-reactive protein. The results of this study warrant further exploration of the relationship between Ang-2, albuminuria, and microinflammation in patients with CKD [37]. In 2014, Tsai et al. [40] showed that higher circulating Ang-2 levels (quartile ≥3) were associated with a more rapid decline in eGFR in comparison with the lowest quartile of Ang-2 (quartile 1). This inverse correlation between GFR and Ang-2 has been observed by others as well, implying that Ang-2 increases with the progress of adult CKD [41, 42]. Moreover, after adjustment for confounding factors, Ang-2 was an independent predictor of all-cause mortality in CKD patients [39, 43]. One of our prior studies

Table 2. Clinical studies of Ang-2 in patients with kidney diseases

First author	Year of publication	Number of patients/controls	Main findings
Fernanda Macedo de Oliveira Neves et al. [29]	2019	265 patients in total: 82 AKI	Plasma Ang-2 was associated with severe AKI after adjusting for several variables; Ang-2 was a mediator between FGF23 and severe AKI
Cassianne Robinson-Cohen et al. [30]	2016	948 subjects in ICU: 506 AKI patients	Plasma concentration of Ang-2 was associated with the presence of AKI in critically ill patients
Camila Barbosa Araújo et al. [31]	2019	283 patients in total: 174 patients without ARDS (38 AKI patients) and 109 ARDS patients (51 AKI patients)	High Ang-2 concentrations at admission were more frequent in ARDS patients; among them, the Ang-2 and Ang-2/Ang-1 ratios were related to severe AKI and the need for RRT
Brandon Michael Henry et al. [32]	2021	51 COVID-19 patients in total: 12 patients with severe AKI	Blood Ang-2 levels were associated with severe AKI in critically ill patients and an independent predictor of requiring RRT in severe AKI
Rianne M Jongman et al. [33]	2015	541 patients in total: 21 AKI; 21 control group (using propensity matching)	Plasma Ang-2 levels increased 1.7-fold in AKI patients compared to the control group; plasma levels of Ang-2 correlated positively with urinary levels of NAG
Andrew S Allegretti et al. [34]	2019	191 inpatients in total: 176 with cirrhosis and AKI; 15 patients with cirrhosis and without AKI	Increased Ang-2 levels were associated with increased mortality, need for RRT, and higher AKI stage
Philipp Kümpers et al. [35]	2010	117 patients with AKI	Circulating Ang-2 levels increased significantly in severe AKI patients; elevated Ang-2 was an independent predictor of 28-day survival in ICU patients after the inception of RRT
Sherry G Mansour et al. [36]	2022	1503 participants in total: 746 with AKI and 757 without AKI	A higher Ang-1/Ang-2 ratio was related to a lower risk of CKD progression, HF, and mortality for AKI patients
Fan-Chi Chang et al. [37]	2013	416 CKD patients with stages 3–5	Plasma Ang-2 levels were independently associated with albuminuria and systemic markers of microinflammation in CKD
Roel Bijkerk et al. [38]	2022	129 patients with ESRD (before RRT)	Levels of Ang-2 in older patients were aberrant when compared with the healthy and DN patients; meanwhile, circulating Ang-2 was reported to be associated with cerebral small vessel disease, brain domains of psychomotor speed, and executive function
Sascha David et al. [39]	2012	128 CKD patients in total: 43 CKD at stage 4, 85 CKD at stage 5, 57 HD, 28 PD; 20 healthy controls with no history of CKD or CVD	Circulating Ang-2 was significantly higher in CKD patients compared to controls; Ang-2 was associated with biomarkers of vascular disease and was markedly higher in dialysis than in stage 4 CKD patients; Ang-2 was an independent predictor of mortality
Yi-Chun Tsai et al. [40]	2014	621 patients with stages 3–5 CKD	The highest quartile of Ang-2 had an increased risk of either commencing dialysis or doubling creatinine and had a more rapid decrease in eGFR than the lowest quartile of Ang-2
Sascha David et al. [41]	2010	44 patients with CKD stage 1–4; 19 patients on dialysis (CKD stage 5); 15 healthy individuals	Circulating Ang-2 was inversely correlated with GFR and increased with the degree of adult CKD

Table 2 (continued)

First author	Year of publication	Number of patients/controls	Main findings
Sazan Rasul et al. [42]	2011	80 T2DM subjects	Ang-2 levels were associated with eGFR after controlling for age and BMI
Yi-Chun Tsai et al. [43]	2015	621 predialysis CKD patients with stages 3–5	Every single higher log Ang-2 had a 5.69-fold risk for MACE or all-cause mortality
Chang Chu et al. [44]	2021	313 HD patients (206 male and 107 female)	Circulating Ang-2 levels at baseline were independently associated with all-cause mortality in male ESRD patients on HD; no association in female ESRD patients
Griet Glorieux et al. [45]	2021	523 nondialysis patients with CKD; 149 patients developed CV	Circulating Ang-2 was predictive of CV events in nondialysis patients with CKD
Yi-Chun Tsai et al. [46]	2016	270 predialysis CKD patients with stages 3–5	High-circulating Ang-2 in stages 3–5 CKD patients was positively associated with cardiac structure
Rukshana C. Shroff et al. [47]	2013	50 CKD children; 20 predialysis CKD; 30 dialysis CKD; 25 healthy controls	Circulating Ang-2 levels were increased in CKD patients on dialysis; Ang-2 was significantly related to dialysis time, carotid artery intima-media thickness, and systolic blood pressure in children with CKD on dialysis
Sascha David et al. [48]	2009	117 CKD patients; 61 HD patients, 24 PD patients; 32 renal transplant recipients; 22 healthy controls	The circulating Ang-2 level was increased in CKD dialysis patients and was related significantly to the degree of CHD and peripheral arterial disease; Ang-2 levels decreased and normalized 3 months after kidney transplantation
Xiaoxiao Yang et al. [49]	2018	324 chronic PD patients	Serum Ang-2 significantly increased with increasing severity of MIAC syndrome; High serum Ang-2 was able to independently predict cardiovascular events in PD patients
Ruth F. Dubin et al. [50]	2018	974 participants	Ang-2 was relevant to makers of HF and an indicator of the occurrence of heart failure in participants with CKD
Jack Bontekoe et al. [51]	2018	97 patients with CKD5-HD; 23–40 AF patients	Plasma levels of Ang-2 were markedly elevated in patients with the coexistence of AF and CKD5-HD in contrast to CKD5-HD alone and AF alone
Jelizaveta Sokolovska et al. [52]	2020	289 T1DM patients in total: 31 DN; 130 arterial hypertension	Increased serum Ang-2 was associated with the occurrence of DN and was an independent predictor of DN
Mohamed Abo El-Asrar et al. [53]	2016	60 T1DM patients; 30 healthy controls	Serum Ang-2 levels were higher in T1DM patients than controls, regardless of microvascular complications; higher levels of Ang-2 were observed in individuals with microalbuminuria; Ang-2 was an independent risk factor of atherosclerosis
Florian G. Scurt et al. [54]	2019	172 T2DM patients with microalbuminuria; 188 matched controls	Baseline Ang-2 was higher in diabetes patients with subsequent microalbuminuria and a significant predictor of new-onset microalbuminuria
Mohamed Salem et al. [55]	2021	40 T2DM patients with microalbuminuria	Baseline blood Ang-2 was elevated in T2DM patients with microalbuminuria and positively correlated to UACR

Table 2 (continued)

First author	Year of publication	Number of patients/controls	Main findings
Mohammad H. Aly et al. [56]	2019	180 diabetes patients; 40 healthy individuals	A significant increase of Ang-2 in patients with normal albuminuria, microalbuminuria, and macroalbuminuria was observed compared to healthy controls; Ang-2 levels were increased with the progression of albuminuria and negatively correlated with eGFR
Narisa Futrakul et al. [57]	2009	50 DN patients; 10 age-matched subjects without diabetes as controls	Plasma Ang-2 was significantly elevated in DN
Yi-Chun Tsai et al. [58]	2018	236 diabetes patients with eGFR <60 mL/min/1.73m^2	The highest quartile of Log-transformed Ang-2 had higher risks of commencing dialysis, rapid renal function decline, MACEs, and elevated all-cause mortality compared to the lowest quartile
M Yu Shvetsov et al. [59]	2015	82 CGN	The urinary excretion of Ang-2 correlated with the renal injury biomarkers NGAL and COL4, and the level of proteinuria; Ang-2 levels were high in the presence of anemia
Monika Edelbauer et al. [60]	2012	23 children and adolescents with LN; 20 healthy controls	Ang-2 levels were significantly higher in children and adolescents with active LN than in remission patients or healthy controls
Vassilios Raptis et al. [61]	2018	26 ADPKD individuals with impaired renal function; 26 ADPKD individuals with preserved renal function; 26 controls without a history of renal disease	ADPKD patients had higher Ang-2 levels than controls, especially in patients with impaired renal function
Melahat Coban et al. [62]	2018	50 patients with ADPKD; 45 healthy controls	Ang-2 was higher at all CKD stages in ADPKD patients

AKI, acute kidney injury; ARDS, acute respiratory distress syndrome; AF, atrial fibrillation; ADPKD, autosomal dominant polycystic kidney disease; BMI, body mass index; CKD, chronic kidney disease; CKD5-HD, stage 5 chronic kidney disease on hemodialysis; CGN, chronic glomerulonephritis; CVD, cardiovascular disease; COL4, type IV collagen; CHD, coronary heart disease; DN, diabetic nephropathy; ESRD, end-stage renal disease; eGFR, estimated glomerular filtration rate; EF, ejection fraction; FGF23, fibroblast growth factor 23; HD, hemodialysis; HF, heart failure; HR, hazard ratio; ICU, intensive care unit; LN, lupus nephritis; MACEs, major adverse cardiovascular events; MIAC, malnutrition, inflammation, atherosclerosis and cardiac valvular calcification; NGAL, neutrophil gelatinase-associated lipocalin; NAG, N-acetyl-beta-D-glucosaminidase; PD, peritoneal dialysis; PWV, pulse wave velocity; RRT, renal replacement therapy; T1DM, type 1 diabetes mellitus; T2DM, type 2 diabetes mellitus; UACR, urinary albumin-creatinine ratio.

also found that serum Ang-2 concentrations at admission were positively associated with all-cause mortality in male ESRD patients on hemodialysis; intriguingly, this association could only be observed in male patients and was absent in female patients. We speculated that males may be more sensitive to Ang-2 than female ESRD patients on hemodialysis (HD) [44]. Although the mechanisms of the sex-specific features of Ang-2 remain unclear, one possible assumption is that the female sex hormone that facilitates vascular ECs proliferation and survival may partially counteract the dysfunction of ECs and angiogenesis caused by increased Ang-2 levels in HD patients.

Besides, Ang-2 concentrations were also in relation to the complications of CKD. As we all know, patients with CKD manifest a significant risk of cardiovascular (CV) events. Nonetheless, available traditional cardiovascular disease (CVD) markers are insufficient to predict survival and CV burden in CKD [41]. Some nontraditional risk factors such as inflammation and endothelial dysfunction were proposed to participate in the mechanisms of CVD in CKD. Furthermore, the researcher observed that blood Ang-2 associated with inflammation and endothelial dysfunction was markedly increased in CKD patients with CVD events. In CKD patients without dialysis, blood Ang-2 concentrations were increased and associated with CV events [43, 45, 46]. Tsai et al. found that the incidence of major adverse cardiovascular events (MACEs) increased 5.69 folds with each log Ang-2 increase in patients with CKD [43]. It has been reported that cardiac structure abnormality was associated with high levels of Ang-2,

confirming the important role of angiogenesis in CVD [45, 46]. In addition to studies in CKD patients without dialysis, several studies showed that circulating Ang-2 level was also significantly increased, as well as being a potent predictor for CVD events in CKD patients on dialysis [47–49]. Shroff et al. found that Ang-2 was significantly correlated with dialysis time, carotid artery intima-media thickness, and systolic blood pressure in children on chronic dialysis [47]. David et al. [48] showed that Ang-2 levels were significantly associated with the severity of coronary heart disease (CHD) and peripheral arterial disease. Moreover, one study conducted by Yang et al. [49] reported that high serum Ang-2 was an independent predictor of CV events in dialysis patients. This study only included prevalent rather than incident peritoneal dialysis (PD) patients, which may introduce a survival bias [49]. Furthermore, serum Ang-2 was associated with markers of heart failure (HF) in CKD patients, suggesting its potential as an indicator for the occurrence of HF in CKD compared to those without CKD [50]. Moreover, in patients with the coexistence of atrial fibrillation (AF) and stage 5 CKD on HD, plasma levels of Ang-2 were markedly elevated compared to stage 5 CKD on HD alone and AF alone [51]. One explanation might be the partial overlap of CKD and CVD pathogenesis, involving microenvironmental inflammation and endothelial dysfunction.

Specific Chronic Kidney Disease

In patients with type 1 DM, Sokolovska et al. and El-Asrar et al. demonstrated that circulating Ang-2 levels were significantly raised and associated with the occurrence of DN [52, 53]. Elevated plasma Ang-2 levels have been reported in type 2 DM (T2DM) patients as well. A study conducted by Scurt et al. showed high blood Ang-2 in T2DM patients with subsequent microalbuminuria [54]. In the multivariate analysis, Ang-2 was demonstrated to be an independent predictor for the incidence of microalbuminuria in T2DM patients [54]. Although this randomized controlled study was from an international multicenter and tried to avoid confounding factors, it is important to note that standard baseline samples were not available for all patients before randomization, and samples for analysis were collected at different points after enrollment [54]. As DM progresses, DN becomes a major microvascular complication of DM. In general, the pathogenesis of DN involves a multifactorial interaction of metabolic and hemodynamic factors. Recently, angiogenesis was regarded as one of the mechanisms of the pathophysiology and progression of DN [55]. Accordingly, angiogenesis-related growth factors such as VEGF-A, Ang-1, and Ang-2 may play an important role in the pathogenesis of DN. It was reported that blood Ang-2 levels were significantly elevated [55–57] and positively correlated to the progress of albuminuria in DN patients with or without albuminuria [55, 56]. Furthermore, Tsai and colleagues demonstrated that Ang-2 can independently predict adverse clinical outcomes, including commencing dialysis, rapid renal function decline, major adverse cardiovascular events, or all-cause mortality in DN [58].

In terms of glomerular disease, Shvetsov et al. demonstrated that the urinary excretion of Ang-2 was higher in chronic glomerulonephritis (CGN) patients with nephrotic syndrome in contrast to CGN patients without nephrotic syndrome [59]. In addition, Ang-2 was increased if the patients suffered from anemia [59]. The authors also observed associations between urinary Ang-2 excretion and urinary markers of kidney injury, namely type IV collagen and neutrophil gelatinase-associated lipocalin in proteinuric CGN patients [54]. This may suggest that an elevated Ang-2 excretion in the urine together with renal injury factors contributes to glomerular damage [59]. Moreover, given the important role of EC injury in the pathogenesis of LN, Edelbauer et al. [60] conducted a study to explore the role of endothelium-related markers in children and adolescents with LN. When compared with healthy controls, circulating Ang-2 levels were markedly elevated in LN patients [60].

In autosomal dominant polycystic kidney disease (ADPKD), increased Ang-2 levels were also observed. In 2018, Raptis et al. [61] showed that circulating Ang-2 levels in ADPKD patients regardless of the state of renal function were markedly higher than in healthy controls. Moreover, ADPKD patients with impaired renal function (eGFR = 45–70 mL/min/1.73 m^2) had significantly higher levels of Ang-2 than those with preserved renal function (eGFR >70 mL/min/1.73 m^2) [61]. In the same year, Cohan et al. confirmed that serum Ang-2 levels were significantly increased in stages 1–2 CKD in comparison with healthy volunteers, but not in advanced stages of CKD, suggesting that angiogenesis may be involved in the development of early stages of ADPKD [62]. A possible explanation for these results may be that the increased degree of sclerotic glomeruli prohibited the formation of vessels in the advanced stage of ADPKD [62].

Taken together, although previous studies did not explain the specific mechanisms of Ang-2 in kidney diseases, they provide evidence that serum Ang-2 can serve as a useful diagnostic and prognostic marker for kidney disease in clinical settings. Circulating Ang-2 may

play a key role in recognizing the onset and predicting the prognosis of AKI and CKD, as well as related complications, guiding physicians to make proper early interference in the near future. Notably, some limitations should be acknowledged when interpreting the findings from these observational studies mentioned above. First, the causal association between the occurrence and development of various kidney diseases and circulating Ang-2 cannot be explained by their results due to potential residual confounding factors within the observational study. Second, these researches did not provide proof of whether circulating Ang-2 possesses biological activity in various renal diseases. Third, most data of these studies were collected from a single center and had a relatively small sample size. Therefore, these studies might have limited generalizability and wide representativeness. Lastly, circulating Ang-2 was measured once at enrollment in most of the studies. Thus, it is not clear whether dynamic Ang-2 levels have additional value in AKI, DN, CKD, and other kidney diseases.

Potential Pharmacological Intervention in the Angiopoietin-2 System

At present, angiotensin-converting enzyme inhibitors (ACEIs) or angiotensin II type 1 receptor blockers (ARBs) have been the backbone treatment for CKD patients, regardless of DM, especially for those with hypertension and proteinuria [63]. Prior studies demonstrated that ACEIs and ARBs provide significant benefits to renal and cardiovascular outcomes for CKD patients [64, 65]. However, the effect of conventional therapies with ACEIs, ARBs, or their combination (ACEIs/ARBs) is limited in patients with low eGFR or albuminuria, due to increased risks of hyperkalemia and AKI [65]. Thus, focusing on new therapeutic targets is warranted.

At present, Ang-2-associated inhibitors have been used for retinal disease in clinical practice. Anti-VEGF agents are the first-line treatment for retinal disease. However, the high treatment burden and low efficacy of anti-VEGF drugs have illuminated the need for new approaches. Many novel emerging drugs including ARP-1536, AKB-9778, AXT107, and the coformulation of nesvacumab and aflibercept for the retinal disease that target the Ang/Tie pathway are still in the early clinical trial phrase [66]. Nevertheless, faricimab, a dual Ang-2 and VEGF inhibitor, results in promising results in phase 2 clinical trials for treating diabetic macular edema (DME) [67] and neovascular age-related macular degeneration (nAMD) [68]. These findings showed that faricimab may improve visual acuity in patients with DME, and bring better treatment durability in both DME and nAMD patients when compared with standard care [69]. Following the positive phase 2 findings, the ongoing phase 3 trials aim to assess the faricimab's safety, efficacy, and durability in DME (YOSEMITE and RHINE) and nAMD (TENAYA and LUCERNE) patients. The potential mechanisms of action of faricimab in DME and AMD might be Tie-2 activation and vascular stabilization.

Tumor angiogenesis is a complex and highly coordinated process that requires the sequential activation of several factors, but the VEGF and Ang/Tie signaling pathways are generally viewed as key steps [70]. Many studies previously demonstrated that Ang-2 expression was upregulated in many types of cancer [71, 72]. Moreover, studies have shown that Ang2 promotes tumor angiogenesis and metastasis in conjunction with VEGFA [73–75]. Ang-2 inhibitors in cancer therapies are currently in various stages of clinical development. Nesvacumab (REGN 910) is a fully human anti-Ang-2 monoclonal antibody for treating advanced-stage solid tumors, completing phase 1 clinical study [76]. Trebananib (AMG 386) is a peptide antibody that inhibits Ang-1 and Ang-2 interaction with their receptor Tie-2, currently in phase 2 clinical study. Prior studies showed trebananib improved progression-free survival when combined with paclitaxel in recurrent ovarian cancer patients [77]. However, subsequent results of the phase 2 study showed that the addition of trebananib to chemotherapy often failed to improve patient outcomes [78]. Additionally, it was found that peripheral edema was linked to dual inhibition of Ang1 and Ang2 [79]. CVX 060 is a monoclonal antibody that can inhibit Ang2. In a preclinical study, CVX 060 alone or combined with VEGF inhibitors made certain progress in reducing tumor growth [80, 81]. Several agents targeting both Ang2 and VEGF, such as CVX-241, vanucizumab, BI836880, and double-antiangiogenic protein (DAAP), demonstrated antitumor activity in phase 1 and 2 clinical studies [78]. Moreover, faricimab has not been used for cancer treatment due to the uncertainty of its antitumor effect.

Although there are no effective inhibitors currently that specifically target Ang-2 in the setting of renal disease within clinical practice. However, the inspiring outcomes in retinal disease and cancer treatment may provide us insights into the potential pharmacological function of Ang-2 in kidney disease. Like DME and AMD, kidney disease is also a multifactorial disease involving dysfunction of microcirculation and ECs, as well as inflammation. Similar to tumor growth, the pathogenesis of kidney disease is also involved in abnormal angiogenesis. Many preclinical and clinical studies have shown that circulating

Ang-2 levels significantly increased in renal diseases, suggesting a key role of Ang-2 in the pathogenesis of kidney disease. Accordingly, inhibitors targeting Ang-2 may be a promising therapy to improve microvascular damage and endothelial dysfunction in renal diseases. Moreover, drugs targeting Ang-2 alone or targeting other factors together with Ang-2 may serve as a novel and desirable strategy to alleviate the occurrence and the development of AKI and CKD in the near future. Of note, potential interactions between Ang-2-Tie2 and VEGF-A pathways in the microenvironment have to be considered, to observe outcomes regarding efficacy and durability.

Conclusions

Recent preclinical and clinical studies provide us with a relatively comprehensive understanding of the role of Ang-2 in renal diseases. Overall, previous studies showed that Ang-2 levels in tissue or blood are greatly increased in certain kidney diseases, which might partly contribute to the dysfunction of endothelium, angiogenesis, and chronic inflammation involved in the pathogenesis of kidney diseases. Based on a line of independent studies, it can be claimed that increased Ang-2 levels qualify as a reliable diagnostic and prognostic marker in kidney diseases. Nevertheless, the pathogenic role of Ang-2 in kidney diseases is still incompletely understood. We are expecting that more largely high-level preclinical will be carried out in the future to explore the specific mechanisms of Ang-2 action in the pathogenesis of kidney diseases. At present, drugs targeting Ang-2 are under clinical trials, mainly as a combination in antiangiogenic therapy of cancer. Despite the extremely limited evidence regarding the use of Ang-2 inhibitors in nonneoplastic diseases in clinical practice [66], their putative therapeutic potential cannot be ignored. We postulate that Ang-2 alone or in combination with other biomarkers can be more widely applied in clinical practice in the near future.

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Conflict of Interest Statement

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References

1 Bondareva O, Sheikh BN. Vascular homeostasis and inflammation in health and disease-lessons from single cell technologies. Int J Mol Sci. 2020 Jun;21(13):4688.
2 Futrakul N, Butthep P, Futrakul P. Altered vascular homeostasis in chronic kidney disease. Clin Hemorheol Microcirc. 2008;38(3):201–7.
3 Maisonpierre PC, Suri C, Jones PF, Bartunkova S, Wiegand SJ, Radziejewski C, et al. Angiopoietin-2, a natural antagonist for Tie2 that disrupts in vivo angiogenesis. Science. 1997 Jul;277(5322):55–60.
4 Gale NW, Thurston G, Hackett SF, Renard R, Wang Q, McClain J, et al. Angiopoietin-2 is required for postnatal angiogenesis and lymphatic patterning, and only the latter role is rescued by Angiopoietin-1. Dev cell. 2002 Sep;3(3):411–23.
5 Fiedler U, Scharpfenecker M, Koidl S, Hegen A, Grunow V, Schmidt JM, et al. The Tie-2 ligand angiopoietin-2 is stored in and rapidly released upon stimulation from endothelial cell Weibel-Palade bodies. Blood. 2004 Jun;103(11):4150–6.
6 Krikun G, Schatz F, Finlay T, Kadner S, Mesia A, Gerrets R, et al. Expression of angiopoietin-2 by human endometrial endothelial cells: regulation by hypoxia and inflammation. Biochem Biophys Res Commun. 2000 Aug;275(1):159–63.
7 Hakanpaa L, Sipila T, Leppanen VM, Gautam P, Nurmi H, Jacquemet G, et al. Endothelial destabilization by angiopoietin-2 via integrin β1 activation. Nat Commun. 2015 Jan;6:5962.
8 Gnudi L. Angiopoietins and diabetic nephropathy. Diabetologia. 2016 Aug;59(8):1616–20.
9 Hazzard TM, Christenson LK, Stouffer RL. Changes in expression of vascular endothelial growth factor and angiopoietin-1 and -2 in the macaque corpus luteum during the menstrual cycle. Mol Hum Reprod. 2000 Nov;6(11):993–8.
10 Chang FC, Chiang WC, Tsai MH, Chou YH, Pan SY, Chang YT, et al. Angiopoietin-2-induced arterial stiffness in CKD. J Am Soc Nephrol. 2014 Jun;25(6):1198–209.
11 Zoccali C, Curatola G, Panuccio V, Tripepi R, Pizzini P, Versace M, et al. Paricalcitol and endothelial function in chronic kidney disease trial. Hypertension. 2014 Nov;64(5):1005–11.

12 Yilmaz MI, Stenvinkel P, Sonmez A, Saglam M, Yaman H, Kilic S, et al. Vascular health, systemic inflammation and progressive reduction in kidney function; clinical determinants and impact on cardiovascular outcomes. Nephrol Dial Transpl. 2011 Nov; 26(11):3537–43.
13 Hirata Y, Sugiyama S, Yamamoto E, Matsuzawa Y, Akiyama E, Kusaka H, et al. Endothelial function and cardiovascular events in chronic kidney disease. Int J Cardiol. 2014 May;173(3):481–6.
14 Annuk M, Soveri I, Zilmer M, Lind L, Hulthe J, Fellström B. Endothelial function, CRP and oxidative stress in chronic kidney disease. J Nephrol. 2005 Nov-Dec;18(6):721–6.
15 Kari JA, Donald AE, Vallance DT, Bruckdorfer KR, Leone A, Mullen MJ, et al. Physiology and biochemistry of endothelial function in children with chronic renal failure. Kidney Int. 1997 Aug;52(2):468–72.
16 Suri C, Jones PF, Patan S, Bartunkova S, Maisonpierre PC, Davis S, et al. Requisite role of angiopoietin-1, a ligand for the TIE2 receptor, during embryonic angiogenesis. Cell. 1996 Dec;87(7):1171–80.
17 Câmara NOS, Iseki K, Kramer H, Liu ZH, Sharma K. Kidney disease and obesity: epidemiology, mechanisms and treatment. Nat Rev Nephrol. 2017 Mar;13(3):181–90.
18 Vassalotti JA, Stevens LA, Levey AS. Testing for chronic kidney disease: a position statement from the National Kidney Foundation. Am J Kidney Dis. 2007 Aug;50(2):169–80.
19 Xue L, Shuyan T, Xiaoli L, Zilong L, Qiuling F, Lining W, et al. Glomerular proteomic profiles in the NZB/W F1 hybrid mouse model of lupus nephritis. Med Sci Monit. 2019 Mar;25:2122–31.
20 Lu YH, Deng AG, Li N, Song MN, Yang X, Liu JS. Changes in angiopoietin expression in glomeruli involved in glomerulosclerosis in rats with daunorubicin-induced nephrosis. Acta Pharmacol Sin. 2006 May;27(5):579–87.
21 Yuan HT, Tipping PG, Li XZ, Long DA, Woolf AS. Angiopoietin correlates with glomerular capillary loss in anti-glomerular basement membrane glomerulonephritis. Kidney Int. 2002 Jun;61(6):2078–89.
22 Davis B, Dei Cas A, Long DA, White KE, Hayward A, Ku CH, et al. Podocyte-specific expression of angiopoietin-2 causes proteinuria and apoptosis of glomerular endothelia. J Am Soc Nephrol. 2007 Aug;18(8): 2320–9.
23 Rizkalla B, Forbes JM, Cao Z, Boner G, Cooper ME. Temporal renal expression of angiogenic growth factors and their receptors in experimental diabetes: role of the renin-angiotensin system. J Hypertens. 2005 Jan; 23(1):153–64.
24 Ichinose K, Maeshima Y, Yamamoto Y, Kinomura M, Hirokoshi K, Kitayama H, et al. 2-(8-hydroxy-6-methoxy-1-oxo-1h-2-benzopyran-3-yl) propionic acid, an inhibitor of angiogenesis, ameliorates renal alterations in obese type 2 diabetic mice. Diabetes. 2006 May;55(5):1232–42.
25 Yamamoto Y, Maeshima Y, Kitayama H, Kitamura S, Takazawa Y, Sugiyama H, et al. Tumstatin peptide, an inhibitor of angiogenesis, prevents glomerular hypertrophy in the early stage of diabetic nephropathy. Diabetes. 2004 Jul;53(7):1831–40.
26 Ichinose K, Maeshima Y, Yamamoto Y, Kitayama H, Takazawa Y, Hirokoshi K, et al. Antiangiogenic endostatin peptide ameliorates renal alterations in the early stage of a type 1 diabetic nephropathy model. Diabetes. 2005 Oct;54(10):2891–903.
27 Luo C, Li T, Zhang C, Chen Q, Li Z, Liu J, et al. Therapeutic effect of alprostadil in diabetic nephropathy: possible roles of angiopoietin-2 and IL-18. Cell Physiol Biochem. 2014;34(3): 916–28.
28 Khairoun M, van der Pol P, de Vries DK, Lievers E, Schlagwein N, de Boer HC, et al. Renal ischemia-reperfusion induces a dysbalance of angiopoietins, accompanied by proliferation of pericytes and fibrosis. Am J Physiol Ren Physiol. 2013 Sep;305(6): F901–10.
29 de Oliveira Neves FM, Araújo CB, de Freitas DF, Arruda BFT, de Macedo Filho LJM, Salles VB, et al. Fibroblast growth factor 23, endothelium biomarkers and acute kidney injury in critically-ill patients. J Transl Med. 2019 Apr;17(1):121.
30 Robinson-Cohen C, Katz R, Price BL, Harju-Baker S, Mikacenic C, Himmelfarb J, et al. Association of markers of endothelial dysregulation Ang1 and Ang2 with acute kidney injury in critically ill patients. Crit Care. 2016 Jul;20(1):207.
31 Araújo CB, de Oliveira Neves FM, de Freitas DF, Arruda BFT, de Macedo Filho LJM, Salles VB, et al. Angiopoietin-2 as a predictor of acute kidney injury in critically ill patients and association with ARDS. Respirology. 2019 Apr;24(4):345–51.
32 Henry BM, de Oliveira MHS, Cheruiyot I, Benoit JL, Cooper DS, Lippi G, et al. Circulating level of Angiopoietin-2 is associated with acute kidney injury in coronavirus disease 2019 (COVID-19). Angiogenesis. 2021 Aug;24(3):403–6.
33 Jongman RM, van Klarenbosch J, Molema G, Zijlstra JG, de Vries AJ, van Meurs M. Angiopoietin/Tie2 dysbalance is associated with acute kidney injury after cardiac surgery assisted by cardiopulmonary bypass. PLoS One. 2015;10(8):e0136205.
34 Allegretti AS, Vela Parada X, Ortiz GA, Long J, Krinsky S, Zhao S, et al. Serum angiopoietin-2 predicts mortality and kidney outcomes in decompensated cirrhosis. Hepatology. 2019 Feb;69(2):729–41.
35 Kümpers P, Hafer C, David S, Hecker H, Lukasz A, Fliser D, et al. Angiopoietin-2 in patients requiring renal replacement therapy in the ICU: relation to acute kidney injury, multiple organ dysfunction syndrome and outcome. Intensive Care Med. 2010 Mar; 36(3):462–70.
36 Mansour SG, Bhatraju PK, Coca SG, Obeid W, Wilson FP, Stanaway IB, et al. Angiopoietins as prognostic markers for future kidney disease and heart failure events after acute kidney injury. J Am Soc Nephrol. 2022 Mar;33(3):613–27.
37 Sokolovska J, Stefanovics J, Gersone G, Pahirko L, Valeinis J, Kalva-Vaivode S, et al. Angiopoietin 2 and neuropeptide Y are associated with diabetic kidney disease in type 1 diabetes mellitus. Exp Clin Endocrinol Diabetes. 2020 Oct;128(10):654–62.
38 El-Asrar MA, Elbarbary NS, Ismail EAR, Bakr AA. Circulating angiopoietin-2 levels in children and adolescents with type 1 diabetes mellitus: relation to carotid and aortic intima-media thickness. Angiogenesis. 2016 Jul; 19(3):421–31.
39 Scurt FG, Menne J, Brandt S, Bernhardt A, Mertens PR, Haller H, et al. Systemic inflammation precedes microalbuminuria in diabetes. Kidney Int Rep. 2019 Jun;4(10): 1373–86.
40 Salem M, Sallam AAM, Abdel-Aleem E, El-Mesallamy HO. Effect of lisinopril and verapamil on angiopoietin 2 and endostatin in hypertensive diabetic patients with nephropathy: a randomized trial. Horm Metab Res. 2021 Jul;53(7):470–7.
41 Aly MH, Arafat MA, Hussein OA, Elsaid HH, Abdel-Hammed AR. Study of Angiopoietin-2 and vascular endothelial growth factor as markers of diabetic nephropathy onset in Egyptians diabetic patients with non-albuminuric state. Diabetes Metab Syndr. 2019 Mar- Apr;13(2):1623–7.
42 Tsai YC, Lee CS, Chiu YW, Lee JJ, Lee SC, Hsu YL, et al. Angiopoietin-2, renal deterioration, major adverse cardiovascular events and all-cause mortality in patients with diabetic nephropathy. Kidney Blood Press Res. 2018;43(2):545–54.
43 Shvetsov MY, Zheng A, Kozlovskaya LV, Serova AG, Travkina EV, Mukhin NA. Urinary excretion of angiogenesis regulatory factors and renal injury markers in chronic glomerulonephritis: significance in the assessment of progression. Ter Arkh. 2015; 87(6):75–82.
44 Chang FC, Lai TS, Chiang CK, Chen YM, Wu MS, Chu TS, et al. Angiopoietin-2 is associated with albuminuria and microinflammation in chronic kidney disease. PLoS One. 2013;8(3): e54668.
45 Tsai YC, Chiu YW, Tsai JC, Kuo HT, Lee SC, Hung CC, et al. Association of angiopoietin-2 with renal outcome in chronic kidney disease. PLoS One. 2014 Oct;9(10):e108862.

46 David S, Kümpers P, Lukasz A, Fliser D, Martens-Lobenhoffer J, Bode-Boger SM, et al. Circulating angiopoietin-2 levels increase with progress of chronic kidney disease. Nephrol Dial Transpl. 2010 Aug;25(8):2571–6.
47 Rasul S, Reiter MH, Ilhan A, Lampichler K, Wagner L, Kautzky-Willer A. Circulating angiopoietin-2 and soluble Tie-2 in type 2 diabetes mellitus: a cross-sectional study. Cardiovasc Diabetol. 2011 Jun;10:55.
48 Bijkerk R, Kallenberg MH, Zijlstra LE, van den Berg BM, de Bresser J, Hammer S, et al. Circulating angiopoietin-2 and angiogenic microRNAs associate with cerebral small vessel disease and cognitive decline in older patients reaching end-stage renal disease. Nephrol Dial Transpl. 2022 Feb;37(3):498–506.
49 Futrakul N, Butthep P, Futrakul P. Altered vascular homeostasis in type 2 diabetic nephropathy. Ren Fail. 2009;31(3):207–10.
50 David S, John SG, Jefferies HJ, Sigrist MK, Kumpers P, Kielstein JT, et al. Angiopoietin-2 levels predict mortality in CKD patients. Nephrol Dial Transpl. 2012 May;27(5):1867–72.
51 Chu C, Chen X, Hasan AA, Szakallova A, Kramer BK, Tepel M, et al. Angiopoietin-2 predicts all-cause mortality in male but not female end-stage kidney disease patients on hemodialysis. Nephrol Dial Transpl. 2021 Jun;37(7):1348–56.
52 Tsai YC, Lee CS, Chiu YW, Kuo HT, Lee SC, Hwang SJ, et al. Angiopoietin-2 as a prognostic biomarker of major adverse cardiovascular events and all-cause mortality in chronic kidney disease. PLoS One. 2015 Aug;10(8):e0135181.
53 Dubin RF, Whooley M, Pico A, Ganz P, Schiller NB, Meyer C. Proteomic analysis of heart failure hospitalization among patients with chronic kidney disease: the Heart and Soul Study. PLoS One. 2018 Dec;13(12):e0208042.
54 Bontekoe J, Lee J, Bansal V, Syed M, Hoppensteadt D, Maia P, et al. Biomarker profiling in stage 5 chronic kidney disease identifies the relationship between angiopoietin-2 and atrial fibrillation. Clin Appl Thromb Hemost. 2018 Dec;24(9_suppl):269s–276s.
55 Glorieux G, Vanholder R, Van Biesen W, Pletinck A, Schepers E, Neirynck N, et al. Free p-cresyl sulfate shows the highest association with cardiovascular outcome in chronic kidney disease. Nephrol Dial Transpl. 2021 May;36(6):998–1005.
56 Tsai YC, Lee CS, Chiu YW, Kuo HT, Lee SC, Hwang SJ, et al. Angiopoietin-2, angiopoietin-1 and subclinical cardiovascular disease in chronic kidney disease. Sci Rep. 2016 Dec;6:39400.
57 Shroff RC, Price KL, Kolatsi-Joannou M, Todd AF, Wells D, Deanfield J, et al. Circulating angiopoietin-2 is a marker for early cardiovascular disease in children on chronic dialysis. PLoS One. 2013;8(2):e56273.
58 David S, Kümpers P, Hellpap J, Horn R, Leitolf H, Haller H, et al. Angiopoietin 2 and cardiovascular disease in dialysis and kidney transplantation. Am J Kidney Dis. 2009 May;53(5):770–8.
59 Yang XX, Zhang H, Shi Y, Yu Z, Yan H, Ni Z, et al. Association of serum angiopoietin-2 with malnutrition, inflammation, atherosclerosis and valvular calcification syndrome and outcome in peritoneal dialysis patients: a prospective cohort study. J Transl Med. 2018 Nov;16(1):312.
60 Edelbauer M, Kshirsagar S, Riedl M, Billing H, Tonshoff B, Haffner D, et al. Soluble VEGF receptor 1 promotes endothelial injury in children and adolescents with lupus nephritis. Pediatr Nephrol. 2012 May;27(5):793–800.
61 Raptis V, Bakogiannis C, Loutradis C, Boutou AK, Lampropoulou I, Intzevidou E, et al. Levels of endocan, angiopoietin-2, and hypoxia-inducible factor-1a in patients with autosomal dominant polycystic kidney disease and different levels of renal function. Am J Nephrol. 2018;47(4):231–8.
62 Coban M, Inci A. The association of serum angiogenic growth factors with renal structure and function in patients with adult autosomal dominant polycystic kidney disease. Int Urol Nephrol. 2018 Jul;50(7):1293–300.
63 Zhang Y, He D, Zhang W, Xing Y, Guo Y, Wang F, et al. ACE inhibitor benefit to kidney and cardiovascular outcomes for patients with non-dialysis chronic kidney disease stages 3-5: a network meta-analysis of randomised clinical trials. Drugs. 2020 Jun;80(8):797–811.
64 Xie X, Liu Y, Perkovic V, Li X, Ninomiya T, Hou W, et al. Renin-angiotensin system inhibitors and kidney and cardiovascular outcomes in patients with CKD: a bayesian network meta-analysis of randomized clinical trials. Am J Kidney Dis. 2016 May;67(5):728–41.
65 Qiao Y, Shin JI, Sang Y, Inker LA, Secora A, Luo S, et al. Discontinuation of angiotensin converting enzyme inhibitors and angiotensin receptor blockers in chronic kidney disease. Mayo Clin Proc. 2019 Nov;94(11):2220–9.
66 Hussain RM, Neiweem AE, Kansara V, Harris A, Ciulla TA. Tie-2/Angiopoietin pathway modulation as a therapeutic strategy for retinal disease. Expert Opin Investig Drugs. 2019, Oct;28(10):861–9.
67 Sahni J, Patel SS, Dugel PU, Khanani AM, Jhaveri CD, Wykoff CC, et al. Simultaneous inhibition of angiopoietin-2 and vascular endothelial growth factor-A with faricimab in diabetic macular edema: BOULEVARD phase 2 randomized trial. Ophthalmology. 2019 Aug;126(8):1155–70.
68 Khanani AM, Patel SS, Ferrone PJ, Osborne A, Sahni J, Grzeschik S, et al. Efficacy of every four monthly and quarterly dosing of faricimab vs ranibizumab in neovascular age-related macular degeneration: the STAIRWAY phase 2 randomized clinical trial. JAMA Ophthalmol. 2020 Sep;138(9):964–72.
69 Khan M, Aziz AA, Shafi NA, Abbas T, Khanani AM. Targeting angiopoietin in retinal vascular diseases: a literature review and summary of clinical trials involving faricimab. Cells. 2020 Aug;9(8):1869.
70 Acker T, Plate KH. Role of hypoxia in tumor angiogenesis-molecular and cellular angiogenic crosstalk. Cell Tissue Res. 2003 Oct;314(1):145–55.
71 Bergers G, Benjamin LE. Tumorigenesis and the angiogenic switch. Nat Rev Cancer. 2003 Jun;3(6):401–10.
72 Kaelin WG, Jr.. The von Hippel-Lindau tumor suppressor gene and kidney cancer. Clin Cancer Res. 2004 Sep;10(18 Pt 2):6290s–6295s.
73 Shim WSN, Ho IAW, Wong PEH. Angiopoietin: a TIE(d) balance in tumor angiogenesis. Mol Cancer Res. 2007 Jul;5(7):655–65.
74 Augustin HG, Koh GY, Thurston G, Alitalo K. Control of vascular morphogenesis and homeostasis through the angiopoietin–Tie system. Nat Rev Mol Cell Biol. 2009 Mar;10(3):165–77.
75 Huang J, Bae JO, Tsai JP, Kadenhe-Chiweshe A, Papa J, Lee A, et al. Angiopoietin-1/Tie-2 activation contributes to vascular survival and tumor growth during VEGF blockade. Int J Oncol. 2009 Jan;34(1):79–87.
76 Papadopoulos KP, Kelley RK, Tolcher AW, Razak ARA, Van Loon K, Patnaik A, et al. A phase I first-in-human study of nesvacumab (REGN910), a fully human anti–angiopoietin-2 (Ang2) monoclonal antibody, in patients with advanced solid tumors. Clin Cancer Res. 2016 Mar;22(6):1348–55.
77 Monk BJ, Poveda A, Vergote I, Raspagliesi F, Fujiwara K, Bae DS, et al. Anti-angiopoietin therapy with trebananib for recurrent ovarian cancer (TRINOVA-1): a randomised, multicentre, double-blind, placebo-controlled phase 3 trial. Lancet Oncol. 2014 Jul;15(8):799–808.
78 Qi S, Deng S, Lian Z, Yu K. Novel drugs with high efficacy against tumor angiogenesis. Int J Mol Sci. 2022 Jun;23(13):6934.
79 Herbst RS, Hong D, Chap L, Kurzrock R, Jackson E, Silverman JM, et al. Safety, pharmacokinetics, and antitumor activity of AMG 386, a selective angiopoietin inhibitor, in adult patients with advanced solid tumors. J Clin Oncol. 2009 Jul;27(21):3557–65.
80 Parmar D, Apte M. Angiopoietin inhibitors: a review on targeting tumor angiogenesis. Eur J Pharmacol. 2021May;899:174021.
81 Wu FTH, Man S, Xu P, Chow A, Paez-Ribes M, Lee CR, et al. Efficacy of cotargeting angiopoietin-2 and the VEGF pathway in the adjuvant postsurgical setting for early breast, colorectal, and renal cancers. Cancer Res. 2016 Dec;76(23):6988–7000.