Diabetic nephropathy (DN) is the leading cause of ESRD in the United States, and its global occurrence is rising rapidly. As evidenced by the wide spectrum of debilitating macro- and microvascular complications that patients eventually experience, the vascular endothelium is a prominent target of longstanding diabetes. In the hunt for breakthrough discoveries that could someday transform the treatment of DN, signaling pathways that regulate the form and functions of blood vessels have recently garnered significant attention. In this issue of JASN, Dessapt-Baradez et al. have deployed a combination of conditionally transgenic mice and human DN biopsy studies to advance the proposition that therapies targeting the vasculature may ameliorate DN.1

In 2001, the National Institutes of Health initiated the Animal Models of Diabetic Complications Consortium to tackle the well known resistance of inbred mouse strains to DN and other diabetic manifestations, a limitation that had severely impeded investigation of the molecular pathogenesis underlying progressive diabetes. The field of vascular research in DN received a substantial boost in 2006 and 2007 when two groups reported in JASN that diabetic mice null for endothelial nitric oxide synthase (eNOS−/−) recapitulated major structural and functional features of advanced human DN.2,3 The enzyme eNOS is highly specifically expressed in the endothelium and is responsible for producing the vasodilator nitric oxide, which is thought to contribute to vascular homeostasis. Loss of eNOS-derived nitric oxide activity has been observed in diabetic patients well before the onset of severe end-organ complications (reviewed by De Vriese et al.4).

Although the regulation of eNOS is complex,5 two important growth factors upstream of eNOS have been implicated in DN—vascular endothelial growth factor-A (VEGF-A) and, more recently, angiopoietins-1 and -2 (Angpt-1 and -2). VEGF-A and Angpt-1 are constantly secreted by healthy podocytes and signal distinct receptors expressed on the surface of endothelial cells, VEGFR1/2 and Tie-2, respectively. As a potentially useful oversimplification, VEGF-A signals through VEGFR2 to induce angiogenesis and to attenuate barrier function, the latter activity accounting for VEGF’s original name of “vascular permeability factor.”6 Angpt-1 activates Tie-2 to stabilize newly sprouted vessels, and, importantly, it prevents vascular leakage and enhances basal microvascular barrier function.7–9 Systemic administration of the anti-VEGF antibody bevacizumab can produce glomerular endotheliosis, proteinuria, and thrombotic microangiopathy, indicating an essential role for VEGF in the maintenance of glomerular architecture and health.10 No comparable clinical data regarding Angpt-1 inhibition exist because drugs targeting this pathway are investigational. But unlike with VEGF-A, conditional knockout mice suggest that Angpt-1 is dispensable in the mature glomerulus.11

The available data for VEGF-A in DN appear to be conflicting. For example, VEGF-A has been reported to be upregulated12 or downregulated13 in human DN biopsies. Deletion of VEGF from podocytes has been shown to exacerbate DN in the streptozotocin (STZ) model of type 1 diabetes by Sivaskandarajah et al.,14 whereas Veron et al.15 reported that inducible overexpression of VEGF in the podocyte causes severe nodular glomerulosclerosis in the STZ model. Similar to the results of Veron et al., the Gnudi laboratory showed that podocyte overexpression of a naturally occurring VEGF inhibitor called sFlt-1 improves DN.16 An attempt to synthesize the VEGF literature in DN is beyond the current scope of this editorial, but in contrast, the current report from Dessapt-Baradez et al. adds to two prior independent experimental studies that collectively demonstrate a renoprotective role for Angpt-1 in DN.

In 2007, a group led by Park used a systemic viral gene therapy approach to achieve excess circulating Angpt-1 for 8 weeks starting during young adulthood in db/db type 2 diabetic mice.17 They found that renal levels of inflammatory adhesion proteins and profibrotic signaling molecules were reduced by Angpt-1 treatment. Urinary albumin excretion was reduced from approximately 150 μg/d to approximately 100 μg/d, and histopathologic changes, namely mesangial matrix expansion and glomerular basement membrane thickness, were similarly reduced by Angpt-1. The study was somewhat confounded because the Angpt-1 group also exhibited less severe elevation of fasting blood glucose levels and less visceral adiposity. In 2011, the Quaggin laboratory genetically deleted Angpt-1 at the end of in utero development and administered STZ 1–3 weeks after weaning.11 Unlike Lee et al. they observed no Angpt-1–dependent effect...
on glucose metabolism, as assessed by the percentage glycosylated hemoglobin. Twenty weeks after STZ administration, diabetic Angpt-1 knockouts had a urine albumin-to-creatinine ratio of 0.25 compared with 0.06 in wild-type diabetic controls. Although not scored, mesangial matrix expansion was much more prominent in the diabetic Angpt-1 knockouts than wild-type diabetic controls.

In the current report, the group led by Gnudi used a gain-of-function system (as did Park et al.), and they studied type 1 diabetes as modeled by STZ (as did Quaggin et al.). They overexpressed Angpt-1 in the podocyte beginning shortly after the induction of hyperglycemia, and they studied outcomes after 10 weeks of excess podocyte-expressed Angpt-1. They noted that excess Angpt-1 did not alter baseline renal physiology or structure. They showed that expression of the Angpt-1 receptor, Tie-2, falls in experimental diabetes, and that their “therapy” of locally expressed Angpt-1 enhances Tie-2 activation. Diabetic Angpt-1 transgenic mice had urinary albumin of 508 μg/d compared with 2101 μg/d in diabetic wild-type controls. Unlike in the prior reports, the authors did not observe any appreciable rescue of mesangial matrix expansion or of GBM thickening by Angpt-1, leaving the physical mechanisms by which diabetic proteinuria improved downstream of Angpt-1 unclear. Finally, using quantitative PCR to compare isolated glomeruli from 12 subjects with DN and 32 living donor controls, they found that DN was associated with induction of the endogenous antagonist of Tie-2, Angpt-2. 18

To summarize, experiments from three independent groups that added Angpt-1 to the diabetic milieu or genetically removed it suggest that Angpt-1 confers renoprotection in experimental diabetes. Because the basis of proteinuria in DN is incompletely understood, there is no consensus on how Angpt-1 is attenuating renal damage in diabetes. To wit, Dessapt-Baradez et al. examined glomerular cell proliferation, VEGF2 phosphorylation, eNOS activation, and nephrin expression in their model. Cross-talk between the VEGF and Angpt-1 signaling axes19,20 and, indeed, the larger network of pathways among endothelia and vascular smooth muscle cells may confound efforts to implicate a single linear cascade. It also remains unclear whether Angpt-1 actually “heals” DN because the interventional studies in mice have commenced at an early, preproteinuric stage of diabetes.

Because only a minority of patients with diabetes develop DN, it will be interesting to test whether polymorphisms in genes comprising vascular pathways or protein levels in the blood or urine can help identify at-risk patients. Conversely, long-term patients with diabetes who do not develop complications may possess a unique vascular-protective profile. If diabetes is fundamentally a metabolic disturbance, we should ask what triggers the dysregulation of vascular pathways21 and why there is a decades-long delay from hyperglycemia to overt complications. Finally, more studies are needed to explore how blood vessel destabilization may contribute to the final common pathway of fibrosis in diabetes as well as other forms of CKD. The study from Dessapt-Baradez et al. provides new evidence implicating Angpt-1 as a renoprotective factor in DN and, more broadly, reminds us that pathogenic molecular events with persistent functional consequences may be unfolding months to years before standard measures of chronic disease are manifest.

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DISCLOSURES

None.

REFERENCES


Antifibrotic Therapy: Is an Antioxidative Regimen the Answer?

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CKD is a major burden for the individual patient as well as for society in general. Whereas only 2% of all patients with CKD eventually reach the stage of dialysis dependency, cardiovascular risk rises steadily with the decline in kidney function, resulting in a robust increase in morbidity and mortality. Morphologically, chronic renal failure is most often characterized by glomerulosclerosis, tubular atrophy, and tubulointerstitial fibrosis, the latter often being the most prominent feature.1 Tubulointerstitial fibrosis consists of proliferation and activation of various cells to so-called myofibroblasts and synthesis of extracellular matrix by these cells. A long-standing controversy surrounds which cell type is the main precursor of these matrix-producing cells, including resident fibroblasts, fibrocytes, pericytes, endothelial cells, and epithelial cells. However, the individual contribution of these various cell types to myofibroblast formation may vary according to the method and model examined, pointing to a certain heterogeneity of these cells.2 However, once matrix-synthesizing myofibroblasts have been formed, renal interstitial fibrosis represents a final common pathway for a plethora of CKDs. Thus, renal interstitial fibrosis is a worthwhile target because antifibrotic therapy would make almost all patients with CKD suitable candidates for therapy. Moreover, antifibrotic therapy may also be of clinical importance in other organs, such as the lung and the liver.

Initial antifibrotic strategies focused mainly on the neutralization of profibrotic cytokines or the application of antifibrotic cytokines.3 However, in recent years the focus of antifibrotic therapy has shifted to antioxidant therapy. Although oxidation was thought to be of primary importance for inflammatory processes, recent evidence has shown that it is also critical for organ fibrosis in general and the kidney in particular.4 Many of these potential therapeutic agents have been tested experimentally and clinically in diabetic kidney disease.5 One of the more interesting substances in this regard is pirenidone, which has antifibrotic, anti-inflammatory, and antioxidant properties. The drug was used successfully to treat many fibrotic disorders, not only in the kidney but also in the lung and the liver. Furthermore, the drug has already been approved for use in patients with idiopathic pulmonary fibrosis; it is in clinical use in Europe but not the United States (the Food and Drug Administration has withdrawn approval in the United States). However, one clinical trial in patients with diabetic nephropathy gave only mixed results,6 whereas a second trial in patients with FSGS is still ongoing.7 Another interesting antioxidant and again antifibrotic agent is tranilast. This synthetic compound attenuates the induction of thioredoxin-interacting protein and oxidative stress. It is used as an antifibrotic agent in Southeast Asia for the treatment of keloid formation. In addition, the drug has been evaluated experimentally8 as well as clinically in diabetic nephropathy,9 although clinical use was confined to only a few patients. Pentoxifylline is a methylxanthine phosphodiesterase inhibitor that has been evaluated in more than 20 studies, mostly in patients with diabetic nephropathy. Unfortunately, most of these studies included only a handful of individuals and were of short duration.10 There was a tendency toward decreased serum creatinine values but no significant effects on proteinuria, and the current evidence does not support the use of the drug in patients with diabetic kidney disease.5

See related article, “Targeted Glomerular Angiopoietin-1 Therapy for Early Diabetic Kidney Disease,” on pages 33–42.