Pathophysiology of Diabetic Nephropathy

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Diabetic nephropathy (DN) is now the commonest cause of end-stage renal failure in the Western world. Recent studies examining the pathogenesis of diabetic complications have focused on the complex interaction between genetic and hemodynamic mechanisms in addition to metabolic factors such as advanced glycation, protein kinase C (PKC) activation, and polyol production. The importance of the various components, particularly with regard to the progression of DN, is currently being explored with the assistance of targeted drug intervention studies.

DIABETES IS NOW the leading cause of end-stage renal failure in the Western world.[1] Diabetic nephropathy (DN) is characterized by the development of overt proteinuria, increasing systemic blood pressure, and declining renal function.[2] However, before the onset of overt proteinuria, there are various functional changes including renal hyperfiltration, hyperperfusion, and increasing permeability to macromolecules such as albumin. These are accompanied by ultrastructural changes including glomerular basement membrane thickening, glomerular hypertrophy, and mesangial expansion with the later development of glomerulosclerosis and tubulointerstitial fibrosis.[3] DN occurs in up to 40% of type I diabetic patients and in a smaller proportion of type II diabetic patients. However, since the prevalence of type II diabetes is much higher, this form of diabetes now contributes at least 50% of diabetic cases in end-stage renal failure programs.[1]

It has been postulated that DN occurs as a result of the interplay of metabolic and hemodynamic factors in the renal microcirculation (Fig 1). Recent studies have focused on the role of cytokine production in the diabetic kidney and noted that both metabolic and hemodynamic factors influence their expression.[4]

GLYCATION

Since diabetes is a state of chronic hyperglycemia, it is likely that glucose-dependent processes participate in the genesis of diabetic complications. In the process of advanced glycation, there is a spontaneous nonenzymatic reaction between glucose and lipids and proteins that leads to the formation of advanced glycosylation end products (AGEs).[5] This family of products, not yet fully characterized, has been shown to accumulate in the diabetic kidney.[6] Aminoguanidine, an inhibitor of AGE formation, has been reported not only to reduce accumulation of renal AGEs but also to attenuate various markers of diabetic renal injury including albuminuria and mesangial expansion.[7] Recently, more powerful inhibitors of advanced glycation have been developed,[8] and their role in the prevention and treatment of diabetic complications is keenly awaited.

An alternative approach to inhibit the effects of advanced glycation may involve the use of the thiazolium compound phenacylthiazolium bromide (PTB), an agent that cleaves AGE-derived protein cross-links.[9] Administration of PTB has been shown to be associated with reduced AGE cross-links in rat tail collagen from diabetic animals. It has been suggested that since this agent can cleave preformed AGE cross-links, it may have a role in reversing AGE-mediated tissue damage and be of particular relevance in a diabetic subject with established renal disease.

It is believed that AGEs interact with specific receptors to cause injury. Several of these receptors have been cloned. One of these, known as the receptor for advanced glycation (RAGE), has been shown by several groups to be present in tissues that undergo vascular injury.[10] RAGE has been detected in cultured mesangial cells and more recently in vivo, where immunohistochemistry has demonstrated localization to the glomerulus.[11] Several of the other putative receptors for AGEs have also been reported to be present in cultured mesangial cells.[12]

POLYOL PATHWAY

Another metabolic route that has been investigated in detail in the genesis of diabetic complications is the polyol pathway.[13] In this pathway, glucose is reduced to sorbitol by the enzyme aldose reductase. Sorbitol accumulation is associated with depletion of myoinositol and changes in the cellular redox potential.[14] The advent of inhibitors of aldose reductase has allowed investigators to explore in vivo the role of the polyol pathway. Results in experimental DN are conflicting, but include both short- and long-term effects on the glomerular filtration rate (GFR), albuminuria, and renal structure.[14-17] The galactose-fed rat has been used to explore the polyol pathway and has been shown to have some renal hemodynamic abnormalities similar to those observed in the streptozotocin-induced diabetic rat.[18] These include a loss of the ability of the afferent arteriole in these rats to constrict in response to an increase in renal perfusion pressure.[18] This phenomenon provides further evidence for interactions between metabolic and hemodynamic pathways. The status of aldose reductase inhibitors in clinical DN remains to be fully ascertained, with several studies reporting beneficial effects on glomerular hyperfiltration and albuminuria.[19,20] Although advanced glycation and polyol metabolism are considered separate glucose-dependent pathways, in the polyol pathway, sorbitol is converted to fructose, a reactive sugar that can lead to production of AGEs. A recent study has shown that aldose reductase inhibition with epalrestat not only inhibited sorbitol formation but was associated with reduced AGE formation.[21]

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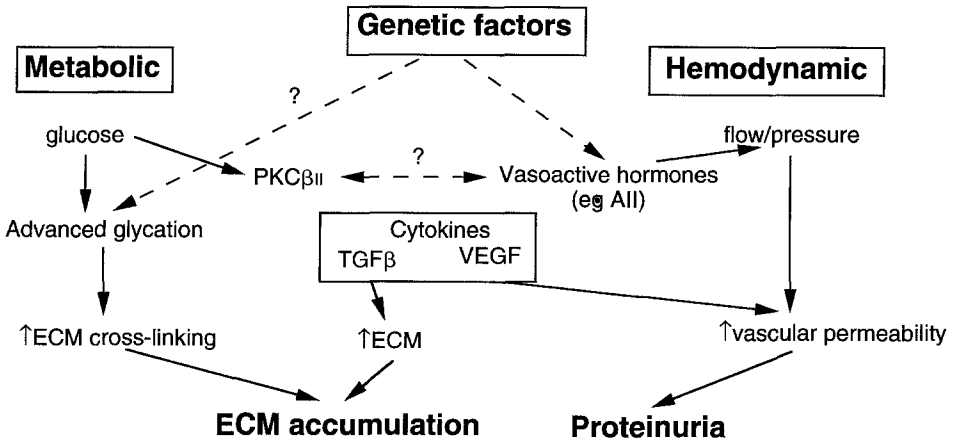

Fig 1. Schema outlining potential interactions among metabolic, hemodynamic, and genetic factors in the pathogenesis of DN. VEGF, vascular endothelial growth factor.

PROTEIN KINASE C

Protein Kinase C (PKC) is a family of serine-threonine kinases that influence a range of functions including cellular proliferation, blood flow, and vascular permeability. PKC activity has been reported to be increased in diabetic glomeruli.[22] Recently, an orally effective inhibitor of the β_{II} isoform of PKC has been developed, LY333531.[23] This compound has been reported to prevent the development of various renal functional changes in diabetic rats, including hyperfiltration and albuminuria.[23]

HEMODYNAMIC FACTORS

DN is commonly associated with systemic hypertension, which would presumably be transmitted to the diabetic glomerulus. Indeed, micropuncture studies have shown that in experimental diabetes, there is an elevation of intraglomerular pressure even in the absence of systemic hypertension.[24] This relates to relative afferent versus efferent arteriolar vasodilation.

A range of vasoactive substances have been postulated to be intimately involved in the genesis of these renal hemodynamic abnormalities. Angiotensin II (AII) has a range of actions including effects on afferent and efferent arteriolar tone, mesangial contractility, and proximal tubular solute transport. Anderson et al[25] have reported an increased expression of various components of the renin-angiotensin system (RAS) in the diabetic kidney. Furthermore, the diabetic kidney has an increased sensitivity to AII.[26] These findings provide a rationale for the use of angiotensin-converting enzyme (ACE) inhibitors in DN. This class of agents that act to inhibit the formation of AII is now widely used in DN. However, these drugs not only act to inhibit AII formation but also to prevent degradation of the vasodilator bradykinin. These non–AII-dependent effects of ACE inhibitors have been investigated in a range of experimental and clinical contexts including DN. Our group has compared ACE inhibition with AII receptor antagonism, an approach that leads to inhibition of the effects of AII without directly acting on kinin pathways, in experimental diabetes. In short-term studies, infusion of the active metabolite of ramipril, ramiprilat, led to a reduction in the GFR in rats after 3 weeks of diabetes.[27] However, this effect was not reproduced by an AII receptor antagonist or by concomitant administration of the bradykinin receptor antagonist icatibant. These findings are consistent with some of the acute effects of ACE inhibition involving bradykinin-dependent pathways. In contrast, in recently performed long-term studies, the beneficial effects of ACE inhibition on albuminuria, glomerular basement membrane thickness, and glomerular hypertrophy were reproduced by the AII receptor antagonist valsartan and could not be blocked by icatibant.[28] This suggests that the chronic effects of ACE inhibitors in the diabetic kidney occur primarily via AII-dependent pathways.

DN involves not only a range of renal functional abnormalities but also pathological changes, the hallmark being extracellular matrix (ECM) accumulation.[3] In the kidney, this ECM consists of structural proteins such as type IV collagen, as well as a range of cytokines including prosclerotic growth factors such as transforming growth factor beta (TGFβ).[29] This growth factor has been shown to play a pivotal role in ECM accumulation in the diabetic kidney. Administration of antibodies to TGFβ prevent diabetes-associated renal hypertrophy.[30] The role of this growth factor is further suggested by both in vitro and in vivo findings indicating that putative mediators of DN such as AII and AGEs promote expression of this cytokine. Therefore, TGFβ may play an important role in the interaction between metabolic and hemodynamic factors in mediating ECM accumulation in the diabetic kidney.

AII has been shown in vitro to promote collagen IV production via TGFβ in mesangial cells.[31] In the model of subtotal nephrectomy, an animal model of progressive renal injury with many hemodynamic and structural similarities to diabetes, our group has shown that in vivo inhibition of AII by ACE inhibition or AII receptor antagonism is associated with reduced gene expression of TGFβ1.[32] These treatments led not only to reduced ECM accumulation but also to attenuation of glomerular and tubulointerstitial injury and preservation of renal function.[32]

Exogenous administration of AGEs upregulates a range of cytokines including TGFβ in the kidney.[33] Recent studies by our group have explored the relationship between TGFβ1, collagen, and AGEs in diabetic vessels, and have shown that the increase in gene expression of TGFβ1 in diabetic vessels can be prevented by administration of the inhibitor of advanced glycation, aminoguanidine.[34] Similar effects in preventing overexpression of TGFβ1 and the matrix protein, type IV collagen, within diabetic blood vessels have also been observed using the

ACE inhibitor perindopril. Whether these effects of inhibitors of glycation and the RAS on growth factor and matrix protein expression are also observed in the kidney also requires evaluation.

GENETIC FACTORS

It is likely that genetic factors play a role in the susceptibility to DN. Indeed, diabetic siblings of probands with DN have a higher incidence of renal disease.[35] A family history of hypertension has been associated with an increased risk of DN. Some investigators have noted a relationship with red blood cell sodium lithium countertransport activity,[36] which is viewed as a risk marker for essential hypertension. As outlined previously, the RAS has been postulated to play a pathogenic role in the development of DN. Therefore, polymorphisms of genes relevant to the RAS such as the angiotensinogen, ACE, and angiotensin type I receptor genes have been evaluated.[37] Polymorphisms of the ACE gene have been suggested to be linked to DN, particularly in type I diabetes, by some investigators.[38] Of particular interest is the finding that ACE gene polymorphism may represent a genetic determinant of the renal response of an individual to ACE inhibition.[39]

Longitudinal studies by our group in a cohort of type I diabetic patients have demonstrated that before the onset of microalbuminuria, most patients already have an increase in serum prorenin, the precursor of the active enzyme renin.[40] To further explore the role of renin in the evolution of DN, diabetes was induced in transgenic Ren 2 rats, a rat strain generated by insertion of the mouse renin Ren 2 gene into the genome.[41] This hypertensive strain has elevated prorenin levels, and induction of diabetes leads to the rapid development of glomerulosclerosis, tubulointerstitial injury, and renal impairment.[42] These changes occur despite blood pressure levels similar to those in the diabetic spontaneously hypertensive rat, a model that does not develop such severe injury.[43] Furthermore, recent studies indicate that the ACE inhibitor perindopril can attenuate renal injury in these diabetic Ren 2 rats.[44] These findings are consistent with the underlying hypothesis that DN is a result of the interaction between hemodynamic and metabolic perturbations that occur in the diabetic kidney. Despite these promising findings that suggest a role for various genetic factors in the pathogenesis of DN, genetic screening cannot yet be considered appropriate as part of routine clinical practice in diabetic patients.

RELEVANCE OF PATHOGENETIC MECHANISMS TO THE TREATMENT APPROACHES

As outlined previously, a number of pathogenetic mechanisms have been clearly shown to be relevant to the development of DN. Glucose-dependent pathways are clearly relevant, and therefore any therapeutic approach that involves intensification of glycemic control should be considered as part of the treatment regimen for a diabetic patient. Indeed, in both type I and type II diabetic subjects, intensified insulin therapy has been shown to retard the development of diabetic renal disease.[45,46] In addition, trials are in progress to directly inhibit some of the pathways that are activated by chronic hyperglycemia. For example, aminoguanidine is presently being investigated as an inhibitor of advanced glycation in type I and type II diabetic subjects with varying degrees of DN.[47]

The importance of hemodynamic factors, including systemic and intraglomerular hypertension, in the pathogenesis of DN has stimulated a large number of clinical studies using a range of antihypertensive agents, including ACE inhibitors.[48] These agents have been shown to retard the development of overt nephropathy in microalbuminuric diabetic patients[49,50] and in proteinuric subjects to reduce the incidence of end-stage renal failure.[51] Therefore, the mainstay of treatment in the diabetic patient at risk of or with established nephropathy remains optimization of glycemic control and reduction in blood pressure.

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