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ASK1: a new therapeutic target for kidney disease

Greg H. Tesch,1,2 Frank Y. Ma,1,2 and David J. Nikolic-Paterson1,2

1Department of Nephrology, Monash Medical Centre, Clayton, Victoria, Australia; and 2Monash University Department of Medicine, Monash Medical Centre, Clayton, Victoria, Australia

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THE PROGRESSION OF KIDNEY INJURY to end-stage renal disease is a major health problem. Current therapies aimed at blood pressure control or immunosuppression can slow but not halt the progression of most forms of kidney disease. Furthermore, immunosuppressive drugs can have toxic side effects. Thus, there is an urgent need to develop new therapies that target the underlying mechanisms of renal injury.

Inflammation and fibrosis are major mechanisms of renal injury that are not targeted by current therapies, and this may explain the inability of current therapies to halt progressive disease reaching end-stage renal failure. The development of these pathological mechanisms is partly dependent on stress-induced activation of protein kinase signaling pathways (p38 MAPK and JNK) in kidney cells (39). Previous studies have shown that therapeutic blockade of p38 MAPK or JNK can inhibit the development of inflammation and fibrosis in animal models of kidney disease (39). However, current strategies that inhibit these kinases can have deleterious side effects [p38 MAPK: liver toxicity, skin rash, and dizziness (21); JNK: exacerbation of albuminuria (26, 33)] alongside their therapeutic benefits due to the roles of p38 MAPK and JNK in homeostatic functions.

Apoptosis signal-regulating kinase 1 (ASK1)/MAP3K5 has been identified as an upstream signaling kinase of p38 MAPK and JNK (25). Importantly, ASK1 is activated only in pathological states, thereby providing a novel target that may interrupt pathological but not homeostatic functions of the downstream p38 MAPK and JNK signaling pathways. Studies in animal models of kidney disease have shown that genetic deficiency or selective inhibition of ASK1 reduces activation of p38 MAPK and JNK (41, 61). Additional in vitro studies on Ask1−/− kidney cells suggest that ASK1 promotes p38 MAPK activation induced by oxidative stress but not innate immunity (41). Therefore, ASK1 inhibitors are currently seen as a potential strategy for targeting the pathological effects of p38 MAPK and JNK activation in kidney disease, which are associated with oxidative stress.

Role of p38 MAPK in Kidney Disease

Three isoforms of p38 MAPK (α, β, and δ) are found in the kidney and are activated by phosphorylation of the activation loop. The structure and function of these p38 MAPK isoforms has been described previously (9). Immunostaining and Western blotting of phosphorylated p38 MAPK has identified activation of p38 MAPK in normal human and rodent kidneys, which is restricted to a small proportion of epithelial cells in glomeruli (podocytes, parietal epithelial cells), tubules, and the macula densa (53, 56). Studies using kinase inhibitors have demonstrated a critical role for p38 MAPK in protecting...
kidney cells from hyperosmotic stress, suggesting that p38 MAPK activation in normal kidney is probably due to fluctuations in osmotic stress that occur in normal kidney function (12, 51).

Human renal biopsy analysis has revealed increased activation of p38 MAPK in a wide range of glomerular diseases and diabetic nephropathy, but not thin membrane disease (1, 56). Kidney activation of p38 MAPK is also substantially higher in proliferative forms of glomerulonephritis (involving IgA nephropathy, postinfectious glomerulonephritis, vasculitis, and systemic lupus erythematosus) than nonproliferative forms of glomerulonephritis (minimal change nephrotic syndrome, membranous glomerulonephritis, and primary focal and segmental glomerulosclerosis) (56). In human nephropathies and rodent models of diabetic nephropathy and antiglomerular basement membrane (GBM) glomerulonephritis, increased activation of p38 MAPK is evident in both intrinsic renal cells (podocytes, tubular cells, mesangial cells, and endothelial cells) and in infiltrating cells (macrophages, neutrophils, and myofibroblasts) and correlates with disease progression (1, 53, 56). These studies suggest a role for p38 MAPK signaling in the development of acute and chronic renal inflammation and renal fibrosis.

Selective inhibitors of p38 MAPK (NPC31145, NPC31169, FR167653, and SB203580) that predominantly target the p38α isoform have been shown to reduce podocyte injury, inflammation, and fibrosis in rodent models of anti-GBM glomerulonephritis (54, 64), ureretic obstruction (55), puromycin aminonucleoside and Adriamycin nephropathies (32), diabetic nephropathy (29), and lupus nephritis (28). In some of these models, protection against kidney damage was associated with reductions in proteinuria and preservation of renal function (28, 32, 54, 64). These studies provided a strong rationale for exploring the therapeutic potential of p38 MAPK inhibition in human kidney diseases.

Role of JNK in Kidney Disease

Two isoforms of JNK (JNK1 and JNK2) are expressed in the kidney. The structures of these JNK isoforms have been described in detail elsewhere (30). JNK signaling in the kidney can be identified by phosphorylation of the JNK activation loop or by phosphorylation of its downstream target c-Jun at Ser63. Immunostaining and Western blotting studies have demonstrated JNK activation in normal human and rodent kidneys, which is localized to some cortical tubules, podocytes, and glomerular parietal epithelial cells (10, 53). A study in cultured epithelial cells has found that JNK is activated by hypertonic stimuli (59), suggesting that JNK activation may be a normal kidney response to fluctuations in osmotic stress. Analysis of patients and rodents with kidney disease has shown that JNK activation is increased in glomeruli and tubules in most types of renal injury and can also be found in resident endothelial cells and infiltrating macrophages, lymphocytes, and myofibroblasts during the progression of kidney injury (10, 11, 33, 38, 53).

Selective JNK inhibitors (SP600125, CC401, and CC930) have been shown to reduce inflammation in rodent models of anti-GBM glomerulonephritis, ischemia-reperfusion injury and diabetic nephropathy (11, 14, 33, 37), and fibrosis in models of ureteric obstruction, anti-GBM glomerulonephritis, and ischemia-reperfusion injury (11, 14, 37, 38). In addition, JNK inhibition improves renal function in models of anti-GBM glomerulonephritis and ischemia reperfusion injury (11, 14, 37). Interestingly, in the ureteric obstruction model, mice genetically deficient in either Jnk1 or Jnk2 are not protected from renal fibrosis, and only Jnk1−/− mice exhibit protection from tubular apoptosis (38). Therefore, it appears that blockade of both JNK1 and JNK2 isoforms is required to provide full protection from the pathological effects of JNK signaling in the kidney.

Therapeutic Limitations of p38 MAPK and JNK Inhibitors

Activation of p38 MAPK and JNK is seen in a wide range of tissues in normal health and to a much greater degree in diseased states. Therefore, it is likely that activation of these pathways has some role in normal physiology in addition to their pathological roles. Current inhibitors of p38 MAPK and JNK can be highly effective at suppressing the activity of their target; however, some of these inhibitors have been shown to have both beneficial and harmful effects (e.g., hepatotoxicity) in disease settings (39). Therefore, there may limitations to blocking all p38 MAPK or JNK signaling as a treatment for disease.

Activation of the p38α isoform induces production of a range of proinflammatory and profibrotic cytokines [e.g., TNFα, monocyte chemoattractant protein-1 (MCP-1) and transforming growth factor-β1 (TGF-β1)] and is a major target of current p38 MAPK inhibitors. Genetic deletion of the ubiquitous p38α isoform results in embryonic death in mice at ~12 days due to effects on erythropoiesis (diminished erythropoietin production) and placental angiogenesis (lack of vascularization of the labyrinth layer) (44, 58). Analysis of p38α−/− mice has found renal structural abnormalities that are apparent at 8 wk of age and progress by 21 wk of age (42). These abnormalities include enlargement of Bowman’s capsule, dilation of proximal tubules, vacuolar degeneration, inflammation, and focal interstitial fibrosis. Consequently, p38α appears to play an important role in the structural and functional integrity of the kidney.

Selective inhibitors of p38 MAPK have been shown to be effective in rodent models of acute and chronic diseases (e.g., ureretic obstruction, anti-GBM glomerulonephritis, and diabetic nephropathy) (29, 54, 55, 64), but these successes have not translated into clinical practice due to lack of efficacy in patients with rheumatoid arthritis and concerns about hepatotoxicity and adverse effects on the central nervous system (21). Therefore, the clinical future of p38 MAPK inhibitors remains in doubt.

Current inhibitors of JNK target both the JNK1 and JNK2 isoforms, which are widely expressed. Genetic deletion of both of these isoforms in mice is fetal lethal on day 12 of embryo development due to neural tube defects (18); however, mice deficient in either isoform alone appear to develop normally with no effects on kidney structure or function (7, 18). Therefore, JNK1 and JNK2 exhibit considerable redundancy and could be individually targeted without the expectation of serious side effects. In contrast, the possibility of combined JNK1/2 blockade having some effects on normal health remains somewhat uncertain.
JNK inhibition during the induction (days 0–14) or progression (days 7–24) of anti-GBM glomerulonephritis in rodents provides significant protection against renal injury, which includes reductions in proteinuria, inflammation, and fibrosis and preservation of renal function (14, 37). In comparison, studies in rodent models of type 1 and type 2 diabetic nephropathy have found that JNK inhibition for 10 wk starting at the onset of albuminuria can exacerbate albuminuria while reducing renal inflammation (26, 33). Whether these differences are due to the duration of JNK blockade or reflect the contribution of JNK activation in different disease settings remains to be determined. Interestingly, a recent phase 2 clinical trial of a JNK inhibitor in idiopathic pulmonary fibrosis (NCT01203943) was terminated on the basis that the benefit/risk profile did not support continuation (study data not available). Therefore, the clinical scope for using JNK inhibitors in patients may be limited to acute injuries to avoid side effects, or it may be dependent on development of inhibitors with greater isoform specificity.

**Upstream Activators of p38 MAPK and JNK Signaling**

An alternative approach to directly targeting the p38 or JNK enzymes is to inhibit the upstream kinases in these signaling pathways, with the goal of suppressing the pathological and not homeostatic functions of these pathways. A summary of the kinases involved in the p38 MAPK and JNK signaling pathways is provided in Fig. 1.

The immediate upstream kinases of p38 MAPK signaling are MKK3 (MAP2K3) and MKK6 (MAP2K6). Mice genetically deficient in these kinases are viable and have normal health (18). Experimental studies have shown that Mkk3-deficient mice are protected against renal injury during ureteric obstruction and the development of type 2 diabetic nephropathy, which were associated with reduced kidney activation of p38 MAPK (34, 40). The impact of Mkk6 deficiency in kidney disease has not yet been explored; however, mice deficient in Mkk6 are protected against collagen-induced arthritis (22). By contrast, mice which are genetically deficient in the kinases immediately upstream of JNK, MKK4 (MAP2K4), or MKK7 (MAP2K7) are not viable due to their role in embryonic development (18). Mice deficient in Mkk4 die on day 12 of embryonic development due to anemia and liver defects, whereas deficiency of Mkk7 in mice causes embryonic death due to reasons undefined (18).

Further upstream, there are more than 20 different MAP3K enzymes that can facilitate activation of the MAP2Ks, which leads to p38 MAPK and/or JNK signaling. Examples of enzymes that activate both pathways are ASK1, mixed-lineage kinase 3 (MLK3), TGF-β-activated kinase 1 (TAK1), and MAPK/ERK kinase kinase 3 (MEKK3) (4). Targeted disruption of the genes encoding these kinases has identified that mice deficient in Ask1 or Mlk3 are viable and healthy, whereas deletion of Tak1 results in fetal lethality in mice on day 11 of development due to neural tube defects and deletion of Mekk3 prevents angiogenesis past day 10 of development, resulting in embryo death by day 12 (18). The normal phenotype of Mlk3−/− mice has been attributed to extensive functional redundancy between MLK3 and the other MLK family members, particularly MLK1/2 (6). In contrast, the normal phenotype of Ask1−/− mice is thought to be due to ASK1 being activated by pathological oxidative stress, which suggests that inhibition of ASK1 could be protective against tissue injury without interfering with normal physiology.

**Activation of ASK1 in the Kidney**

Under normal conditions, ASK1 is maintained in an inactive state through binding to the reduced form of thioredoxin-1 (in mitochondria ASK1 binds to Trx2). However, under pathological conditions, mitochondrial stress, endoplasmic reticulum (ER) stress, and receptor-mediated inflammatory signals can induce intracellular oxidative stress, which activates ASK1 by dissociation and oxidation of thioredoxin (TRX). Oxidation of TRX also causes dissociation of TRX and the thioredoxin-interacting protein (TXNIP), allowing TXNIP to bind Nod-like receptor 3 and activate the Nod-like receptor 3 inflammasome (68). Furthermore, increased levels of thioredoxin-interacting protein (TXNIP) can sequester thioredoxin in its re-
Reduced form, thereby prolonging the activation of ASK1 (52). These conditions allow active ASK1 to facilitate downstream p38 MAPK and JNK signaling, resulting in progression of injury (see Fig. 2).

ASK1 is expressed in most tissues, including kidney, heart, lung, liver, and brain (62). One study has reported phosphorylation of the ASK1 activation loop in glomeruli, peritubular capillaries, and tubular epithelial cells in renal allograft rejection, which was absent in normal human kidney (3). In addition, Western blotting has shown that kidney levels of phosphorylated (activated) (p)-ASK1 are increased in rodent models of kidney ischemia reperfusion injury (60, 65). Furthermore, these increases in p-ASK1 correlated with increased levels of p-MKK3, p-p38 MAPK, p-JNK, and tubular cell apoptosis, suggesting a role for ASK1 in tubular cell injury induced by ischemia. Similarly, increased activation of MKK3/6, p38 MAPK, and JNK in mouse kidneys following ureter obstruction is dependent on ASK1 and is associated with tubular cell apoptosis and interstitial inflammation and fibrosis (41). Therefore, in vivo evidence supports a role for ASK1 in tubular cell apoptosis during kidney injury.

In vitro studies have also suggested that ASK1 can promote kidney disease. High glucose induces oxidative stress and increased ASK1 activation in mouse mesangial cells, which correlates with increased levels of cleaved caspase 3 (52). Antioxidant treatment or knockdown of TXNIP in these mesangial cells reduced both ASK1 activation and apoptosis. Similarly, Adriamycin induces oxidative stress and increases p-ASK1 and p-p38 MAPK in mouse podocytes, which is associated with reduced cell viability (15). Treatment with antioxidants or AMPK suppresses ASK1-p38 MAPK signaling in these cells, providing protection against podocyte injury. These studies suggest that ASK1 may play a critical role in podocyte or mesangial cell apoptosis induced by oxidative stress during diabetes or acute cell injury.

**Role of ASK1 in Kidney Injury Following Ischemia or Ureteric Obstruction**

Studies performed in transgenic mice have provided valuable insights into the role of ASK1 in tubulo-interstitial injury in the kidney. Ask1−/− mice are protected from renal injury resulting from kidney ischemia–reperfusion (IR) or unilateral ureter obstruction (UUO) (41, 60). In the IR model, Ask1 deficiency reduces kidney activation of both p38 MAPK and JNK by ~50% and inhibits tubular cell apoptosis (80% reduction), infiltrating interstitial leukocytes (70% reduction) and loss of kidney function (50% reduction) (60). In the UUO model, Ask1 deficiency reduces the activation of p38 MAPK by 90% and JNK by 60% and suppresses tubular apoptosis (35% reduction), interstitial macrophage accumulation of Nod-like receptor 3, and interstitial fibrosis (50% reduction) (41). Therefore, in some types of kidney injury, ASK1 is an important mediator of tubular damage and possibly the tubular response to injury.

Cell culture studies have examined some of the underlying mechanisms for ASK1 causing tubular injury in the IR and UUO models. Tubular epithelial cells (TEC) from Ask1−/− mice show reduced caspase-3 activity and cell death under hypoxic conditions that promote endoplasmic reticulum stress (60). In contrast, when Ask1−/− TEC are transfected with an adenovirus encoding dominant active ASK1, caspase-3 activity and cell death are increased in a dose-dependent manner. Interestingly, TEC transfected with a dominant inactive ASK1 secrete less of the chemokine MCP-1/CCL2 under hypoxic conditions compared with transfection controls; however, it is unclear whether this is related to reduced cell death (60). Additional studies have shown that Ask1−/− TEC have reduced p38 MAPK activation and produce less TGF-β1, PDGFβ, and...
MCP-1 in response to stimulation with angiotensin II or hydrogen peroxide but not IL-1α or LPS (41). These findings suggest that endoplasmic reticulum stress, oxidative stress, and signaling through specific receptors are major inducers of ASK1 activity in tubular cells. The exact mechanisms by which ASK1 promotes interstitial inflammation and fibrosis following tubular injury are less clear. Reduced levels of the chemokine MCP-1 are found in the kidneys of Ask1−/− mice in both the IR and UUO models (41, 60), indicating a potential role for ASK1 in macrophage-mediated inflammation following tubular injury. However, tubular cells are the major source of MCP-1 in the kidney, and therefore, the lower levels of MCP-1 in these Ask1−/− kidneys may be an indirect result of reduced tubular cell injury in the absence of ASK1. In the UUO model, fibrosis occurs in the interstitium and is dependent on the accumulation and activity of interstitial fibroblasts. Interestingly, fibroblasts obtained from the UUO kidneys of wild-type and Ask1−/− mice showed similar proliferation and matrix production, suggesting that ASK1 acts indirectly on fibroblasts to promote interstitial fibrosis in this model (41). Therefore, further studies are needed in mice using conditional gene deletion in specific cell types (e.g., proximal tubular cells, macrophages, T cells, and fibroblasts) to determine how ASK1 regulates inflammation and fibrosis during the development of tubulointerstitial injury.

Potential Role of ASK1 in Chronic Kidney Diseases

Oxidative stress and activation of p38 MAPK and JNK signaling are features of chronic kidney diseases such as glomerulonephritis, diabetic nephropathy, and hypertensive nephropathy, suggesting a potential role for ASK1 in development of these diseases. Glomerulonephritis. Although intervention studies have established clear roles for p38 MAPK and JNK in several models of glomerulonephritis, little is known about the role of ASK1 in these glomerular diseases. One study has shown that ASK1 is active in glomeruli of rats with Heymann nephritis, in which complement induces subthalamic injury to glomerular epithelial cells (GEC) causing oxidative stress and proteinuria (49). Furthermore, in vitro studies demonstrated that complement-induced lysis was increased in GEC, which overexpressed ASK1 and was attenuated in GEC that overexpress a dominant negative ASK1 mutant (49). These findings suggest that ASK1 is a mediator of C5b-9-dependent cell injury. Studies in Ask1−/− mice have revealed that ASK1 promotes the expression of cytokines (MCP-1, IL-1β, and TNFα) required for the recruitment and activation of macrophages during skin and cardiac injury (46, 47). Therefore, it is possible that ASK1 may play a role in macrophage-mediated glomerular injury during the development of glomerulonephritis.

Diabetic nephropathy. In vivo and in vitro data indirectly supports a role for ASK1 in diabetic nephropathy. High glucose levels induce mitochondrial oxidative stress (45) and upregulate Txnip in cultured kidney tubular cells, mesangial cells, and podocytes (16, 48, 50). Furthermore, Txnip levels are increased in human and experimental diabetic nephropathy and correlate with kidney fibrosis (2). In addition, transgenic mice overexpressing Trx (presumably with reduced ASK1 activation) are protected from albuminuria and mesangial matrix expansion in streptozotocin-induced diabetic nephropathy (20).

In vitro studies have also shown that advanced glycation end products (AGEs) induce proinflammatory effects in kidney cells by activating p38 MAPK signaling (8). Therefore, ASK1 may be critical for mediating the effects of hyperglycemia and AGEs in diabetic kidneys that induce injury. A postulated role of ASK1 in diabetic nephropathy is shown in Fig. 2.

Hypertensive nephropathy. Indirect evidence also suggests that ASK1 may contribute to hypertensive nephropathy. Patients with hypertension display marked upregulation of ACE and downregulation of ACE2 in their kidneys, which is associated with increased levels of angiotensin II and kidney activation of p38 MAPK (31). Blockade of p38 MAPK in kidney tubular cells abolishes angiotensin II-induced upregulation of CTGF and collagen I and downregulation of ACE2, indicating that activation of p38 MAPK signaling can contribute to injury in hypertensive kidneys (31, 67). In addition, in vitro studies show that angiotensin II-induced activation of p38 MAPK and production of TGF-β1, PDGFβ, and MCP-1 by tubular epithelial cells is reduced in the absence of ASK1 (41). The effects of hypertension have not yet been explored in the kidneys of Ask1−/− mice. However, Ask1−/− mice are known to be protected from cardiac injury (hypertrophy, apoptosis, inflammation, and fibrosis) induced by angiotensin II infusion or aldosterone/salt (27, 46), and similar disease mechanisms are seen in hypertensive nephropathy.

Therapeutic Evaluation of ASK1 Inhibitors in Kidney Disease

Growing interest in ASK1 as a potential therapeutic target has led to the development of selective synthetic inhibitors of ASK1 (Table 1). These ASK1 inhibitors have been shown to have therapeutic benefits in various animal disease models, including multiple sclerosis (19), contact hypersensitivity (43), cardiac and renal ischemia-reperfusion injury (13, 17), tumor growth (24), acetaminophen hepatotoxicity (66), and diabetic nephropathy (61) (Table 1). Administration of the ASK1 inhibitor NQDI-1 to uninephrectomized rats 1 h prior to commencing 45 min of renal ischemia resulted in protection from kidney oxidative stress, tubular cell apoptosis, loss on renal function, and histological damage (13). This study demonstrated that prophylactic treatment with an ASK1 inhibitor could protect kidneys from ischemia-reperfusion injury and was consistent with previous findings in Ask1−/− mice.

A recent study has evaluated a potent and highly selective ASK1 inhibitor, GS-44217, as an early or late intervention treatment in a model of diabetic nephropathy in hypertensive NOS3−/− mice (Table 1) (61). In a panel of 451 kinases, GS-44217 has more than 50 times greater affinity for ASK1 than any other kinase (61), which is an unusually high degree of selectivity for a kinase inhibitor drug. Treatment with GS-44217 prevented p38 MAPK activation in diabetic kidneys but had no impact on hypertension. Early intervention with GS-44217 from weeks 2 to 8 of diabetes reduced the development of glomerulosclerosis and loss of renal function but had no effect on the development of albuminuria. Late intervention with GS-44217 from weeks 8 to 15 of diabetes halted the progression of glomerulosclerosis (Fig. 3), renal inflammation, and tubular injury and improved renal function despite having no effect on established albuminuria. This study iden-
tified ASK1 inhibition as a potential intervention treatment for diabetic nephropathy.

Currently, Gilead Sciences is performing a phase 2 placebo-controlled clinical trial (NCT02177786) with an ASK1 inhibitor (GS-4997) in 300 patients with type 2 diabetes and stage 3 or 4 diabetic kidney disease receiving standard of care therapy (35). The design of this trial involves examination of three doses of GS-4997, with the primary end point being estimated glomerular filtration rate at 48 wk (35). This trial was expected to be completed in August 2016, with final collection of data for primary outcomes occurring in July 2016. A total of 334 patients have been recruited for this study.

Potential Therapeutic Limitations of ASK1 Inhibitors in Disease

Because Ask1−/− mice develop normally, the expectation is that ASK1 inhibitors will not affect normal physiology, and therefore, they will be safe for therapeutic use in patients. However, given that ASK1 signaling activates p38 MAPK and JNK and that inhibitors of p38 MAPK and JNK have side effects, some caution is warranted. One study in a model of colitis has shown that bowel inflammation is increased in Ask1−/− mice due to suppression of the immune response to bacteria, which was attenuated with antibiotics (23). Furthermore, this study demonstrated that bone marrow macrophages from Ask1−/− mice have a reduced capacity to kill the bacteria causing colitis due to a lack of p38 MAPK activation. Therefore, ASK1 inhibition could potentially suppress innate immunity, which may be either beneficial or detrimental depending on the disease being targeted.

Further Issues To Be Addressed

Despite recent advances made possible with the availability of Ask1−/− mice and selective ASK1 inhibitors, there are still a number of important issues to be addressed to obtain a greater understanding of the role of ASK1 in kidney diseases and the therapeutic potential of its inhibition. For example, it will be important to know whether ASK1 contributes to the innate and adaptive immune responses seen in various forms of kidney disease caused by infection (e.g.,

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<th>ASK1 Inhibitor</th>
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<td>EAE model of multiple sclerosis (mice) (19)</td>
<td>Reduced demyelination of spinal cord and optic nerve</td>
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<td>Compound inhibited ATP binding to ASK1 catalytic site</td>
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<td>Diabetic nephropathy (mice) (61)</td>
<td>Glomerulosclerosis halted, renal function improved</td>
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<td>Diabetic nephropathy (human, phase 2 clinical trial, trial identifier NCT02177786) (35)</td>
<td>Trial in progress; expected completion August 2016</td>
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ASK1, apoptosis signal-regulating kinase 1; NA, not available.
pyelonephritis) or autoimmunity (e.g., lupus nephritis), which will require investigations in the relevant disease models. In addition, the association of single-nucleotide polymorphisms in the ASK1 gene with insulin sensitivity (5) argues that ASK1 variants should be investigated in patients with type 2 diabetic nephropathy. Other challenges will include defining the role of ASK1 in cancer and the endoplasmic reticulum unfolding protein response, which may place limitations on the therapeutic use of ASK1 inhibitors in chronic diseases.

Conclusions

Evidence from experimental animal models has demonstrated that inhibition of ASK1 is protective as a prophylactic treatment in kidney IR injury and as an intervention therapy in diabetic nephropathy. Given that downstream p38 MAPK/JNK signaling is known to cause injury in many types of kidney disease, there is significant scope for ASK1 inhibition to become a therapy in other progressive kidney diseases, including glomerulonephritis and hypertensive nephropathy. Furthermore, since ASK1 inhibition appears to have no effect on normal physiology or hypertension, it is likely to be well tolerated and potentially useful as an adjunct treatment with current antihypertensive therapy. The current clinical trial of an ASK1 inhibitor in patients with diabetic nephropathy is expected to provide valuable insight into the potential of ASK1 as a therapy for kidney injury, which may lead to further trials in nondiabetic forms of chronic kidney disease.

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DISCLOSURES

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AUTHOR CONTRIBUTIONS

G.T. drafted manuscript; G.T., F.Y.M., and D.J.N.-P. edited and revised manuscript; G.T. approved final version of manuscript.

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