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Review

Differential role of mesangial cells and podocytes in TGF-ß-induced mesangial matrix synthesis in chronic glomerular disease

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Summary. Glomerulosclerosis is characterized by mesangial matrix accumulation that is mediated primarily by activation of transforming growth factor-ß (TGF-ß). Unlike podocytes, mesangial cells secrete TGF-ß in response to common in vitro fibrogenic stimuli. However, mesangial immunostaining for active TGF-ß1 in chronic glomerular disease is almost negligible, despite increased mesangial TGF-ß1 mRNA expression, while podocytes covering the sclerotic glomerular segments exhibit increased TGF-ß1 protein expression. The mechanisms whereby TGF-ß is activated in the diseased glomeruli and how the activated TGF-ß leads to mesangial matrix overproduction are not clear. We provide evidence that TGF-ß secreted as latent complexes by mesangial cells is stored in the mesangial matrix, from which soluble forms of latent TGF-ß are released and localized to the podocyte surface in chronic glomerular disease. Podocyte-derived reactive oxygen species, plasmin and thrombospondin-1, particularly renin-angiotensin-aldosterone system-induced oxidative stress, seem to be involved in TGF-ß activation in podocytes. We also provide evidence that the TGF-ß-induced secretion of connective tissue growth factor and vascular endothelial growth factor by podocytes acts as a paracrine regulatory mechanism on mesangial cells, which may cause mesangial matrix accumulation culminating in the development of glomerulosclerosis. Collectively, these data bring new insights into our understanding of the roles of the mesangial cells and podocytes in the TGF-ß-induced mesangial matrix synthesis in chronic glomerular disease.

Key words: Chronic glomerular disease, Glomerulosclerosis, Mesangial matrix, Podocytes, TGF-ß activation

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Introduction

Glomerulosclerosis frequently complicates most renal diseases, and is characterized by the collapse of the glomerular tuft and accumulation of extracellular matrix (ECM) in the mesangial area. Transforming growth factor-ß (TGF-ß) is a key regulator of ECM. TGF-ß is secreted as latent complexes, which are stored in the ECM to provide stability to the active molecule and a readily activable source of it (Lawrence, 2001). Overexpression of active TGF-ß1 in transgenic mice caused mesangial expansion, interstitial fibrosis and renal insufficiency (Kopp et al., 1996), while monoclonal antibody to TGF-ß reduces the glomerulosclerosis in experimental proliferative glomerulonephritis (GN) (Yu et al., 2004) and diabetic nephropathy (Ziyadeh et al., 2000; Benigni et al., 2003).

Mesangial cells secrete TGF-ß in response to common in vitro fibrogenic stimuli (Lee and Song, 2009), and mainly produce matrix protein in chronic glomerular disease. As yet, mesangial immunostaining for active TGF-ß1 is frequently negative in chronic glomerular disease in humans (Stein-Oakley et al., 1997; Kim et al., 2002, 2003; Wahab et al., 2005), while podocytes covering the sclerotic segments exhibit increased expression of TGF-ß1 protein (Kim et al., 2002, 2003; Wahab et al., 2005). Recent studies suggest that podocytes may play a significant role in the formation of glomerulosclerosis (Wolf et al., 2005; Matsusaka et al., 2005; Ziyadeh and Wolf, 2008). Nonetheless, common in vitro metabolic stimulators, such as high glucose (Iglesias-de la Cruz et al., 2002), angiotensin II (Ang II) (Chen et al., 2005), and oxidized low-density lipoprotein (H.S.L., unpublished data), and serotonin (Xu et al., 2007) fail to stimulate the expression of TGF-ß1 in cultured immortalized mouse podocytes (Mundel et al., 1997). In this regard, this article will discuss the mechanisms whereby latent TGF-ß secreted by mesangial cells would be activated in the

diseased glomeruli, and how activated TGF-ß could lead to enhanced mesangial matrix synthesis culminating in the development of glomerulosclerosis.

Activation of latent TGF-ß

The activity of all TGF-ß isoforms is under strict control during developmental and pathological processes. TGF-ß is secreted as latent complexes in which a 25 kD homodimer growth factor is noncovalently associated with a latency-associated peptide (LAP, 75 KD). Some cells secrete TGF-ß in the form of a TGF-ß/LAP complex, referred to as the small latent complex. However, most cells secrete TGF-ß as part of a large latent complex, in which latent TGF-ß binding protein (LTBP, 125-160 kD) is linked to the small latent complex. LTBP has a role in targeting the transport of latent TGF-ß complex into the ECM (Hyytiäinen et al., 2004; Koli et al., 2008). Intense mesangial immunostaining for LTBP-1 is observed in anti-Thy1.1 nephritis associated with severe but transient mesangial matrix accumulation (Porst et al., 2006).

The large latent complex is susceptible to proteolysis, within which LTBP is first cleaved at the protease sensitive hinge region. The soluble large latent complex is then released from the ECM and is activated by another proteolytic event that releases TGF-ß from LAP (Koli et al., 2001). The TGF-ß released then binds to its receptor and exerts its cellular functions.

Under in vitro conditions, latent TGF-ß is activated by heating, acid or alkaline treatment, irradiation, reactive oxygen species (ROS), proteases including plasmin, cathepsin, calpain, matrix metalloproteinase (MMP)-2 and MMP-9, some integrins, or thrombospondin-1 (TSP-1) (reviewed in Koli et al., 2001).

Several activation mechanisms may exist in vivo as reviewed below (Table 1).

TGF-ß activation by proteolysis

Plasmin could function as an in vivo activator of TGF-ß (Lyons et al., 1990; Edgtton et al., 2004). Plasmin can release the large latent TGF-ß complex from the ECM by cleaving LTBP at the amino terminal hinge region (Taipale et al., 1992). Furthermore, it can cleave LAP that releases active TGF-ß dimer from the large latent TGF-ß1 complex (Lyons et al., 1990; Annes et al., 2003; George et al., 2005) (Table 1).

MMP-2 and MMP–9 have also been implicated in the cleavage of LAP and the release of mature TGF-ß at the cell surface (Yu et al., 2000).

TSP-1-mediated activation of TGF-ß

TSP-1 is known to be a major physiologic activator of latent TGF-ß (Crawford et al., 1998). A specific peptide sequence within TSP-1, KRFK, binds to the LSKL sequence in LAP, and releases mature TGF-ß by inducing conformational changes in the protein (Crawford et al., 1998; Lawrence, 2001; Koli et al., 2008) (Table 1). However, it is not clear whether TSP-1 alone could directly activate latent TGF-ß (Abdelouahed et al., 2000; Grainger and Frow, 2000; Otsuka et al., 2007).

Overexpression of both TSP-1 and active TGF-ß occurs in podocytes in patients with focal segmental glomerulosclerosis (FSGS) (Kim et al., 2003) and diabetic nephropathy (Wahab et al., 2005). In various experimental renal disease models, TSP-1 is co-localized with TGF-ß and predicts the development of tissue fibrosis (Hugo et al., 1998).

In rats with anti-Thy1.1 nephritis, active TGF-ß1-positive area/glomerulus was 18%, while treatment with antisense TSP-1 oligodeoxynucleotides reduced it to 9% (Daniel et al., 2003). In wild-type and TSP-1 deficient diabetic mice, the TGF-ß1-positive area in the glomerulus was 5% and 3%, respectively (Daniel et al., 2007). Thus, TSP-1 seems to activate mesangial TGF-ß1 more actively in acute mesangial proliferate GN than in chronic glomerular disease.

ROS-mediated activation of TGF-ß

ROS produced by ionizing radiation was found to induce rapid TGF-ß activation in vivo (Barcellos-Hoff and Dix, 1996). ROS can activate TGF-ß directly through oxidation-induced conformational change in LAP, in which the unique methionine residue in the TGF-ß1/LAP functions as a redox switch center (Jobling et al., 2006), or indirectly through the activation of proteolytic enzymes (Koli et al., 2008) (Table 1).

Integrins in TGF-ß binding and activation

Integrins can bind to the RGD recognition domain in the LAP of TGF-ß1 and TGF-ß3. Particularly, αvß6 and αvß8 are known to activate the TGF-ß complex (Munger et al., 1999; Mu et al., 2002), leading to release of TGF-ß either by tractional force (αvß6) or by MT1-MMP-dependent proteolytic activity (αvß8) (Koli et al., 2008). By cell movement, an αvß6–integrin-expressing cell causes sufficient traction for the release of mature TGF-

Table 1. Possible mechanisms of latent TGF-ß activation in chronic glomerular disease.

Activators	Mechanisms of action
Plasmin	Proteolytic nicking of LTBP releasing large latent TGF-ß complex from the ECM Proteolytic nicking of the LAP
TSP-1	Induction of conformational changes in LAP
ROS	Induction of conformational changes in LAP Inducing activation of proteolytic enzymes
Integrin, αvß6	Release of TGF-ß from LAP by tractional force

TGF-ß, transforming growth factor-ß; LTBP, latent TGF-ß binding protein; ECM, extracellular matrix; TSP-1, thrombospondin-1; LAP, latency-associated peptide; ROS, reactive oxygen species.

ß from the ECM-anchored large latent complex (Annes et al., 2004) (Table 1). αvß6 integrin is expressed in the diseased kidneys confined to the distal tubules and collecting ducts (Trevillian et al., 2004).

Expression of TGF-ß protein in chronic glomerular disease

Mesangial expression of TGF-ß

In the early stage of human diabetic nephropathy (Wahab et al., 2005) and IgA nephropathy (IgAN) with mesangial cell proliferation (Stein-Oakley et al., 1997), some mesangial cells show immunoreactivity for TGF-ß1. Nonetheless, mesangial immunostaining for active TGF-ß1 is very weak or almost negligible in most human IgAN (Stein-Oakley et al., 1997; Kim et al., 2002), FSGS (Kim et al., 2003) and diabetic nodular glomerulosclerosis (Wahab et al., 2005), despite increased mesangial TGF-ß1 mRNA levels (Kim et al., 2002, 2003; Wahab et al., 2005).

Mesangial expression of TGF-ß isoforms is transiently upregulated in acute anti-Thy1.1 nephritis (Ito et al., 2001; Hartner et al., 2003), in which TGF-ß1 expression is only segmentally and weakly distributed (Ito et al., 2001). In the glomeruli of experimental diabetic nephropathy, only a few cells show positive immunostaining for TGF-ß1 (Hill et al., 2000). Mutations of the Wilms' tumour suppressor gene, WT1, induce Denys-Drash syndrome (DDS) characterized by diffuse mesangial sclerosis. In DDS mice, no TGF-ß1 expression occurred in the mesangium (Patek et al., 2003).

Altogether, immunohistochemical antigen detection for mesangial TGF-ß1 might be difficult in progressive glomerular disease, suggesting that latent TGF-ß complexes secreted by mesangial cells might be stored in the mesangial matrix to provide stability to the active molecule.

Expression of TGF-ß in podocytes

In contrast to mesangial cells, podocytes covering the sclerotic segments exhibit increased expression of TGF-ß1 mRNA and protein in human IgAN (Kim et al., 2002), FSGS (Kim et al., 2003) and end-stage diabetic nephropathy (Wahab et al., 2005), and in DDS mice (Patek et al., 2003).

Profibrotic growth factors mediating the downstream fibrogenic activity of TGF-ß

Connective tissue growth factor (CTGF)

CTGF is a major autocrine growth factor induced by TGF-ß. TGF-ß1 induces CTGF mRNA and protein expression in mesangial cells (Riser et al., 2000; Blom et al., 2001; Ito et al., 2001; Chen et al., 2002; Cooker et al., 2007), podocytes (Ito et al., 2001), and parietal epithelial cells (Kanemoto et al., 2003). However, CTGF administration stimulates CTGF, but not TGF-ß, expression in mesangial cells (Riser et al., 2000). Both TGF-ß and CTGF independently induced collagen or fibronectin expression in mesangial cells (Riser et al., 2000; Blom et al., 2001; Gore-Hyer et al., 2002; Weston et al., 2003). CTGF can interact with, and influence the signaling of, TGF-ß and bone morphogenic proteins (BMPs) (Abreu et al., 2002; Nguyen et al., 2008), and vascular endothelial growth factor (VEGF) (Hashimoto et al., 2002). Maximal TGF-ß1 induction of CTGF requires synergy between Smad and Ras/extracellular signal-regulated kinase signaling (Chen et al., 2002).

Expression of CTGF mRNA and/or protein in the mesangium and podocytes is upregulated in human chronic glomerular disease (Ito et al., 1998; Wahab et al., 2005), but not in acute postinfectious GN (Ito et al., 1998). CTGF is also overexpressed in the glomeruli of experimental diabetic glomerulosclerosis (Riser et al., 2000) and mesangial proliferative GN (Ito et al., 2001).

VEGF

Besides its angiogenic actions, VEGF may play an important role in TGF-ß1-induced glomerular fibrosis (Wang et al., 2004; Chen et al., 2005). TGF-ß1 stimulates VEGF expression in podocytes (Iglesias-de la Cruz et al., 2002) and mesangial cells (Wang et al., 2004), although treatment with anti-TGF-ß slightly prevented the enhanced VEGF mRNA expression in the db/db kidneys (Ziyadeh et al., 2000). VEGF increases type I collagen and fibronectin synthesis in mesangial cells via a TGF-ß1-induced mitogen-activated protein kinase activity (Wang et al., 2004). VEGF and/or VEGF receptor expression is increased in the glomeruli of diabetic animals, particularly in the podocytes (Wendt et al., 2003; Sung et al., 2006).

Platelet-derived growth factor (PDGF)

PDGF-B and PDGF–D chains mediate mesangial cell proliferation and matrix accumulation in anti-Thy1.1 model (Ostendorf et al., 2006) and in diabetic nephropathy (Nakagawa et al., 2000). PDGF-B induces TGF-ß in mesangial cells (Yamabe et al., 2000) or vice versa (Chow et al., 2005). PDGF-B antagonism had no effect on the glomerular expression of TGF-ß system components in rat anti-Thy1.1 model despite reduced ECM protein accumulation (Ostendorf et al., 2002). Thus, PDGF-B could act downstream of TGF-ß, or PDGF-B and TGF-ß might act independently of each other in vivo.

Activation of latent TGF-ß in the diseased glomeruli

Activation of TGF-ß by mesangial cells

The latent TGF-ß complexes stored in the mesangial matrix may be catabolized by plasmin. The plasmin-mediated TGF-ß activation, however, may be neutralized via feedback inhibition, since TGF-ß-induced

plasminogen activator (PA) inhibitor-1 production decreases the formation of active plasmin in mesangial cells (Baricos et al., 2003). Furthermore, accumulation of mesangial matrix progressed in association with enhanced mesangial fibrin deposition in rats with anti-Thy 1.1 nephritis (Liu et al., 2004). Thus, the mesangial cell surface surrounded by an enlarged matrix may not express active plasmin to liberate mature TGF-ß.

Unlike mesangial cells, podocytes do not overexpress TGF-ß1 in response to common in vitro metabolic stimuli (Iglesias-de la Cruz et al., 2002; Chen et al., 2005), although albumin load or mechanical strain upregulates TGF-ß1 mRNA expression in them (Abbate et al., 2002; Durvasula et al., 2004). In this regard, Wolf et al. (2005) suggested that podocytes respond to paracrine TGF-ß coming from the mesangial cell. Even though free active TGF-ß is liberated from the mesangial cells, it has a very short half-life in plasma (2-3 min) (Coffey et al., 1987), in contrast to the latent TGF-ß complex with a significantly longer half-life (>100 min) (Wakefield et al., 1990). Thus, latent TGF-ß, rather than active TGF-ß, may be localized to the podocyte surface after its release from the mesangial matrix.

Activation of latent TGF-ß by podocytes

The renin-angiotensin-aldosterone system (RAAS) seems to be involved in podocyte injury through the induction of oxidative stress in experimental renal disease (Chiba et al., 2002; Nagase et al., 2006; Whaley-Connell et al., 2006, 2008; Shibata et al., 2007). Diabetic podocytopathy is also mediated by Ang II (Ziyadeh and Wolf, 2008). Furthermore, podocyte-specific overexpression of the antioxidant metallothionein reduces the glomerular injury in diabetic mice, suggesting that oxidative damage to the podocyte plays a pathogenetic role in diabetic nephropathy (Zheng et al., 2008). Salt loading in spontaneously hypertensive stroke-prone rats induces renal inflammation/fibrosis, podocyte injury, and an increase in plasmin and MMP-2 activity (Gianella et al., 2007). Collectively, injured podocytes mediated by RAAS seem to overproduce ROS, which can activate the latent TGF-ß localized to their surface, either directly or indirectly, through the activation of proteolytic enzymes in chronic renal disease.

Control renal biopsies exhibit expression of urokinase-type PA (uPA) mRNA confined to the podocytes. Strong expression of uPA protein and mRNA is sometimes observed in podocytes within crescents, suggesting that damaged podocytes can release plasmin (Lee et al., 2001). In human FSGS lesions, the expression levels of TGF-ß1, TSP-1 and TGF-ß type II receptor mRNAs and proteins and phosphorylated Smad2/Smad3 are increased in podocytes (Kim et al., 2003). These observations suggest that damage to podocytes may stimulate the TGF-ß/Smad signaling pathway, and that TSP-1 may activate TGF-ß1 in podocytes in chronic glomerular disease.

Altogether, podocyte-derived ROS, plasmin and TSP-1 may release mature TGF-ß from LAP localized to the podocytes in the diseased glomeruli. The activated TGF-ß may bind to its receptor on podocytes, activating the TGF-ß/Smad signaling pathway to induce the overexpression of its target genes, such as CTGF and VEGF.

Paracrine interaction of growth factors between podocytes and mesangial cells in the diseased glomeruli

Podocyte-specific injury in adult mice leads to mesangial expansion and glomerulosclerosis (Matsusaka et al., 2005), suggesting that some humoral factors produced by podocytes seem to have fibrogenic effects on mesangial cells (Table 2). In support of this hypothesis, CTGF mRNA expression occurred only in the podocytes and glomerular parietal epithelial cells during the early stage of the anti-Thy-1.1 nephritis, a model of mesangiopathy (Ito et al., 2001). Furthermore, induction of diabetes in podocyte-specific CTGF-transgenic mice results in an increased mesangial CTGF expression with more severe mesangial expansion than diabetic wild-type mice (Yokoi et al., 2008) (Table 2). Although the glomerular expression of VEGF seems to be confined to the podocytes (Wendt et al., 2003), anti-

Table 2. Animal experiments to support the hypothesis that podocyte-derived growth factors induce mesangial matrix accumulation.

Experiments	Results	1st author
Podocyte-specific injury in mice	Mesangial expansion	Matsusaka (2005)
Anti-Thy-1.1 nephritis	CTGF mRNA expression confined to podocytes	Ito (2001)
Diabetes in podocyte-specific CTGF-transgenic mice	Increased mesangial CTGF expression and more severe mesangial expansion than wild-type mice	Yokoi (2008)
Anti-VEGF administration into diabetic mice	Decreased mesangial matrix expansion	Flyvbjerg (2002)
Diabetes in podocyte-specific BMP-7-transgenic mice	Prevents glomerular fibrosis without change in TGF-ß levels	Wang (2006)

TGF-ß, transforming growth factor-ß; CTGF, connective tissue growth factor; VEGF, vascular endothelial growth factor; BMP-7, bone morphogenic protein-7.

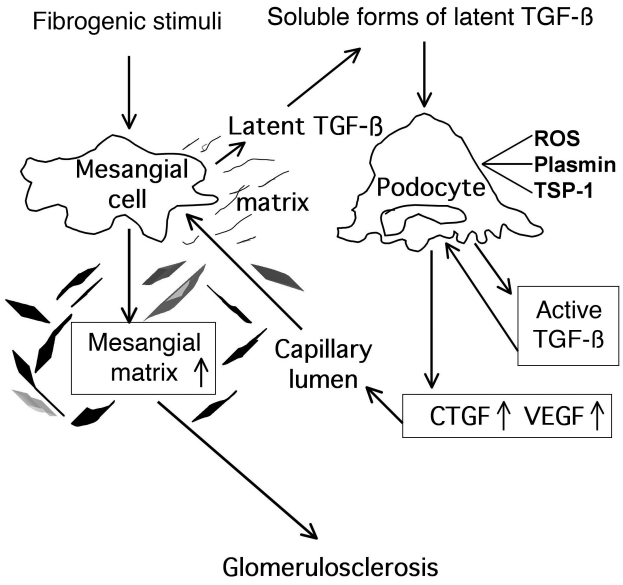

Fig. 1. Hypothetical pathway for mesangial matrix accumulation via activation of latent TGF-ß by podocytes in chronic glomerular disease. CTGF, conntective tissue growth factor; VEGF, vascular endothelial growth factor; TGF-ß, transforming growth factor-ß; TSP-1, thrombospondin-1; ROS, reactive oxygen species.

VEGF attenuates the mesangial matrix expansion in diabetic mice (Flyvbjerg et al., 2002). BMP-7 counteracts the fibrogenic action of TGF-ß, possibly by inhibiting the fibrogenic Smad signaling. Transgenic expression of BMP-7 in podocytes of diabetic mice prevents glomerular fibrosis without changing renal TGF-ß levels (Wang et al., 2006), suggesting that the activated TGF-ß/Smad signaling in podocytes may lead to mesangial matrix accumulation.

Contrary to the general perception that solutes cannot move against the flow of glomerular filtration, about one third of VEGF secreted from podocytes would reach the capillary lumen and accumulate there (Katavetin and Katavetin, 2008). Indeed, diffusion dominates the transport of most solute in the glomerular basement membrane (Deen et al., 2001). Although it is not clear whether this is also the case for CTGF, the experiments performed by Yokoi et al. (2008) support that possibility.

To sum up, latent TGF-ß1 complexes secreted by mesangial cells seem to be stored in the mesangial matrix in chronic glomerular disease. Soluble forms of latent TGF-ß released from the mesangial matrix may be localized to the podocyte surface, where ROS, plasmin and TSP-1 seem to activate the latent TGF-ß in podocytes. TGF-ß-induced CTGF and VEGF secretion by podocytes may act as a paracrine regulatory mechanism, necessary for the mesangial matrix accumulation (Fig. 1).

Conclusions

Experimental and clinical data suggest that latent TGF-ß complexes secreted by mesangial cells are stored in the mesangial matrix, from which soluble forms of latent TGF-ß may be released and localized to the podocyte surface in chronic glomerular disease. Podocyte-derived ROS, plasmin and TSP-1, particularly RAAS-induced oxidative stress, seem to be involved in TGF-ß activation. Active TGF-ß may induce CTGF and VEGF overexpression in podocytes, which may act as a paracrine regulatory mechanism on mesangial cells to stimulate mesangial matrix synthesis. Research on the glomerular production and activation of TGF-ß and its downstream mediators, CTGF and VEGF, will further our comprehension of the mesangial matrix accumulation and provide new therapeutic strategies to prevent the development of glomerulosclerosis in patients with chronic kidney disease.

References

Abbate M., Zoja C., Morigi M., Rottoli D., Angioletti S., Tomasoni S., Zanchi C., Longaretti L., Donadelli R. and Remuzzi G. (2002). Transforming growth factor-ß1 is up-regulated by podocytes in response to excess intraglomerular passage of proteins: a central pathway in progressive glomerulosclerosis. Am. J. Pathol. 161, 2179-2193.

Abdelouahed M., Ludlow A., Brunner G. and Lawler J. (2000). Activation of platelet transforming growth factor ß-1 in the absence of thrombospondin-1. J. Biol. Chem. 275, 17933-17936.

Abreu J.G., Ketpura N.I., Reversade B. and Robertis E.M. (2002). Connective-tissue growth factor (CTGF) modulates cell signalling by BMP and TGF-ß. Nature Cell Biol. 4, 599-604.

Annes J.P., Chen Y., Munger J.S. and Rifkin D.B. (2004). Integrin αvß6-mediated activation of latent TGF-ß requires the latent TGF-ß-binding protein-1. J. Cell Biol. 165, 723-734.

Annes J.P., Munger J.S. and Rifkin D.B. (2003). Making sense of latent TGFß activation. J. Cell Sci. 116, 217-224.

Barcellos-Hoff M.H. and Dix T.A. (1996). Redox-mediated activation of latent transforming growth factor-ß1. Mol. Endocrinol. 10, 1077-1083.

Baricos W.H., Reed J.C. and Cortez S.L. (2003). Extracellular matrix degradation by cultured mesangial cells: mediators and modulators. Exp. Biol. Med. 228, 1018-1022.

Benigni A., Zoja C., Corna D., Zatelli C., Conti S., Campana M., Gagliardini E., Rottoli D., Zanchi C., Abbate M., Ledbetter S. and Remuzzi G. (2003). Add-on anti-TGF-ß antibody to ACE inhibitor arrests progressive diabetic nephropathy in the rat. J. Am. Soc. Nephrol. 14, 1816-1824.

Blom I.E., van Dijk A.J., Wieten L., Duran K., Ito Y., Kleij L., deNichilo M., Rabelink T.J., Weening J.J., Aten J. and Goldschmeding R. (2001). In vitro evidence for differential involvement of CTGF, TGFß, and PDGF-BB in mesangial response to injury. Nephrol. Dial. Transplant. 16, 1139-1148.

Chen S., Lee J.S., Iglesias-de la Cruz M.C., Wang A., Izquierdo-Lahuerta A., Gandhi N.K., Danesh F.R., Wolf G. and Ziyadeh F.N. (2005). Angiotensin II stimulates α3(IV) collagen production in mouse podocytes via TGF-ß and VEGF signaling: implications for

diabetic glomerulopathy. Nephrol. Dial. Transplant. 20, 1320-1328.

Chen Y., Blom I.E., Sa S., Goldschmeding R., Abraham D.J. and Leask A. (2002). CTGF expression in mesangial cells: Involvement of SMADs, MAP kinase, and PKC. Kidney Int. 62, 1149-1159.

Chiba Y., Ando K. and Fujita T. (2002). The protective effects of taurine against renal damage by salt loading in Dahl salt-sensitive rats. J. Hypertens. 20, 2269-2274.

Chow E.K., O'Connell R.M., Schilling S., Wang X-F., Fu X-Y. and Cheng G. (2005). TLR agonists regulate PDGF-B production and cell proliferation through TGF-ß/type I IFN crosstalk. EMBO J. 24, 4071-4081.

Coffey R.J. Jr, Kost L.J., Lyons R.M., Moses H.L. and LaRusso N.E. (1987). Hepatic processing of transforming growth factor ß in the rat. Uptake, metabolism, and biliary excretion. J. Clin. Invest. 80, 750-757.

Cooker L.A., Peterson D., Rambow J., Riser M.L., Riser R.E., Najmabadi F., Brigstock D. and Riser B.L. (2007). TNF-α, but not IFN-γ, regulates CCN2 (CTGF), collagen type I, and proliferation in mesangial cells: possible roles in the progression of renal fibrosis. Am. J. Physiol. Renal Physiol. 293, F157-F165.

Crawford S.E., Stellmach V., Murphy-Ullrich J.E., Ribeiro S.M., Lawler J., Hynes R.O., Boivin G.P. and Bouck N. (1998). Thrombospondin-1 is a major activator of TGF-ß1 in vivo. Cell 93, 1159-1170.

Daniel C., Schaub K., Amann K., Lawler J. and Hugo C. (2007). Thrombospondin-1 is an endogenous activator of TGF-ß in experimental diabetic nephropathy in vivo. Diabetes 56, 2982-2989.

Daniel C., Takabatake Y., Mizui M., Isaka Y., Kawashi H., Rupprecht H., Imai E. and Hugo C. (2003). Antisense oligonucleotides against thrombospondin-1 inhibit activation of TGF-ß in fibrotic renal disease in the rat in vivo. Am. J. Pathol. 163, 1185-1192.

Deen W.M., Lazzara M.J. and Myers B.D. (2001). Structural determinants of glomerular permeability. Am. J. Physiol. Renal Physiol. 281, F579-F596.

Durvasula R.V., Petermann A.T., Hiromura K., Blonski M., Pippin J., Mundel P., Pichler R., Griffin S., Couser W.G. and Shankland S.J. (2004). Activation of a local tissue angiotensin system in podocytes by mechanical strain. Kidney Int. 65, 30-39.

Edgtton K.L., Gow R.M., Kelly D.J., Carmeliet P. and Kitching A.R. (2004). Plasmin is not protective in experimental renal interstitial fibrosis. Kidney Int. 66, 68-76.

Flyvbjerg A., Dagnaes-Hansen F., De Vriese A.S., Schrijvers B.F., Tilton R.G. and Rasch R. (2002). Amelioration of long-term renal changes in obese type 2 diabetic mice by a neutralizing vascular endothelial growth factor antibody. Diabetes 51, 3090-3094.

George S.J., Johnson J.L., Smith M.A., Angelini G.D. and Jackson C.L. (2005). Transforming growth factor-ß is activated by plasmin and inhibits smooth muscle cell death in human saphenous vein. J. Vasc. Res. 42, 247-254.

Gianella A., Nobili E., Abbate M., Zoja C., Gelosa P., Mussoni L., Bellosta S., Canavesi M., Rottoli D., Guerrini U., Brioschi M., Banfi C., Tremoli E., Remuzzi G. and Sironi L. (2007). Rosuvastatin treatment prevents progressive kidney inflammation and fibrosis in stroke-prone rats. Am. J. Pathol. 170, 1165-1177.

Gore-Hyer E., Shegogue D., Markiewicz M., Lo S., Hazen-Martin D., Greene E.L., Grotendorst G. and Trojanowska M. (2002). TGF-ß and CTGF have overlapping and distinct fibrogenic effects on human renal cells. Am. J. Physiol. Renal Physiol. 283, F707-F716.

Grainger D.J. and Frow E.K. (2000). Thrombospondin 1 does not activate transforming growth factor-ß 1 in a chemically defined system or in smooth muscle-cell cultures. Biochem. J. 350, 291-298.

Hartner A., Hilgers K.F., Bitzer M., Veelken R. and Schöcklmann H.O. (2003). Dynamic expression patterns of transforming growth factor-ß(2) and transforming growth factor-ß receptors in experimental glomerulonephritis. J. Mol. Med. 81, 32-42.

Hashimoto G., Inoki I., Fujii Y., Aoki T., Ikeda E. and Okada Y. (2002). Matrix metalloproteinases cleave connective tissue growth factor and reactivate angiogenic activity of vascular endothelial growth factor 165. J. Biol. Chem. 277, 36288-36295.

Hill C., Flyvbjerg A., Gronbaek H., Petrik H., Hill D.J., Thomas C.R., Sheppard M.C. and Logan A. (2000). The renal expression of transforming growth factor-ß isoforms and their receptors in acute and chronic experimental diabetes in rats. Endocrinology 141, 1196-1208.

Hugo C., Shankland S.J., Pichler R.H., Couser W.G. and Johnson R.J. (1998). Thrombospondin-1 precedes and predicts the development of tubulointerstitial fibrosis in glomerular disease in the rat. Kidney Int. 53, 302-311.

Hyytiäinen M., Penttinen C. and Keski-Oja J. (2004). Latent TGF-ß binding proteins: extracellular matrix association and roles in TGF-ß activation. Crit. Rev. Clin. Lab. Sci. 41, 233-264.

Iglesias-de la Cruz M.C., Ziyadeh F.N., Isono M., Kouahou M., Han D.C., Kalluri R., Mundel P. and Chen S. (2002). Effects of high glucose and TGF-ß1 on the expression of collagen IV and vascular endothelial growth factor in mouse podocytes. Kidney Int. 62, 901-913.

Ito Y., Aten J., Bende R.J., Oemar B.S., Rabelink T.J., Weening J.J. and Goldschmeding R. (1998). Expression of connective tissue growth factor in human renal fibrosis. Kidney Int. 53, 853-861.

Ito Y., Goldschmeding R., Bende R.J., Claessen N., Chand M.A., Kleij L., Rabelink T.J., Weening J.J. and Aten J. (2001). Kinectics of connective tissue growth factor expression during experimental proliferative glomerulonephritis. J. Am. Soc. Nephrol. 12, 472-484.

Jobling M.F., Mott J.D., Finnegan M.T., Jurukovski V., Erickson A.C., Walian P.J., Taylor S.E., Ledbetter S., Lawrence C.M., Rifkin D.B. and Barcellos-Hoff M.H. (2006). Isoform-specific activation of latent transforming growth factor-ß (LTGF-ß) by reactive oxygen species. Radiat. Res. 166, 839-848.

Kanemoto K., Usui J., Tomari S., Yokoi H., Mukoyama M., Aten J., Weening J.J. and Nagata M. (2003). Connective tissue growth factor participates in scar formation of crescentic glomerulonephritis. Lab. Invest. 83, 1615-1625.

Katavetin P. and Katavetin P. (2008). VEGF inhibition and renal thrombotic microangiopathy. N. Engl. J. Med. 359, 205-206.

Kim H.W., Moon K.C., Park S.Y., Hong H.K. and Lee H.S. (2002). Differential expression of platelet-derived growth factor and transforming growth factor ß in relation to progression of IgA nephropathy. Nephrology 7, S131-S139.

Kim J.H., Kim B.K., Moon K.C., Hong H.K. and Lee H.S. (2003). Activation of the TGF-ß/Smad signaling pathway in focal segmental glomerulosclerosis. Kidney Int. 64, 1715-1721.

Koli K., Myllärniemi M., Keski-Oja J. and Kinnula V.L. (2008). Transforming growth factor-ß activation in the lung: focus on fibrosis and reactive oxygen species. Antioxid. Redox. Signal 10, 333-342

Koli K., Saharinen J., Hyytiäinen M., Penttinen C. and Keski-Oja J. (2001). Latency, activation, and binding proteins of TGF-ß. Microsc. Res. Tech. 52, 354-362.

Kopp J.B., Factor V.M., Mozes M., Nagy P., Sanderson N., Boettinger E.P., Klotman P.E. and Thorgeirsson S.S. (1996). Transgenic mice

with increased plasma levels of TGF-ß1 develop progressive renal disease. Lab. Invest. 74, 991-1003.

Lawrence D.A. (2001). Latent TGF-ß: An overview. Mol. Cell Biochem. 219,163-170.

Lee H.S., Park S.Y., Moon K.C., Hong H.K., Song C.Y. and Hong S.Y. (2001). mRNA expression of urokinase and plasminogen activator inhibitor-1 in human crescentic glomerulonephritis. Histopathology 39, 203-209.

Lee H.S. and Song C.Y. (2009). Oxidized low-density lipoprotein and oxidative stress in the development of glomerulosclerosis. Am. J. Nephrol. 29, 62-70.

Liu N., Makino T., Nogaki F., Kusano H., Suyama K., Muso E., Honda G., Kita T. and Ono T. (2004). Coagulation in the mesangial area promotes ECM accumulation through factor V expression in MsPGN in rats. Am. J. Physiol. Renal Physiol. 287, F612-F620.

Lyons R., Gentry L.E., Purchio A.F. and Moses H.L. (1990). Mechanism of activation of latent recombinant transforming growth factor-ß1 by plasmin. J. Cell Biol. 110, 1361-1367.

Matsusaka T., Xin J., Niwa S., Kobayashi K., Akatsuka A., Hashizume H., Wang Q-c., Pastan I., Fogo A.B. and Ichikawa I. (2005). Genetic engineering of glomerular sclerosis in the mouse via control of onset and severity of podocyte-specific injury. J. Am. Soc. Nephrol. 16, 1013-1023.

Mu D., Cambier S., Fjellbirkeland L., Baron J.L., Munger J.S., Kawakatsu H., Sheppard D., Broaddus V.C. and Nishimura S.L. (2002). The integrin αvß8 mediates epithelial homeostasis through MT1-MMP-dependent activation of TGF-ß1. J. Cell Biol. 157, 493-507.

Mundel P., Reiser J., Zuniga-Mejia-Borja A., Pavenstädt H., Davidson G., Kriz W. and Zeller R. (1997). Rearrangements of the cytoskeleton and cell contacts induce process formation during differentiation of conditionally immortalized mouse podocyte cell lines. Exp. Cell Res. 236, 248-258.

Munger J.S., Huang X., Kawakatsu H., Griffiths M.J., Dalton S.L., Wu J., Pittet J.F., Kaminski N., Garat C., Matthay M.A., Rifkin D.B. and Sheppard D. (1999). The integrin αvß6 binds and activates latent TGFß 1: a mechanism for regulating pulmonary inflammation and fibrosis. Cell 96, 319-328.

Nagase M., Shibata S., Yoshida S., Nagase T., Gotoda T. and Fujita T. (2006). Podocyte injury underlies the glomerulopathy of Dahl salt-hypertensive rats and is reversed by aldosterone blocker. Hypertension 47, 1084-1093.

Nakagawa H., Sasahara M., Haneda M., Koya D., Hazama F. and Kikkawa R. (2000). Immunohistochemical characterization of glomerular PDGF B-chain and PDGF ß-receptor expression in diabetic rats. Diabetes Res. Clin. Pract. 48, 87-98.

Nguyen T.Q., Roestenberg P., van Nieuwenhoven F.A., Bovenschen N., Li Z., Xu L., Oliver N., Aten J., Joles J.A., Vial C., Brandan E., Lyons K.M. and Goldschmeding R. (2008). CTGF inhibits BMP-7 signaling in diabetic nephropathy. J. Am. Soc. Nephrol. 19, 2098-2107.

Ostendorf T., Kunter U., van Roeyen C., Dooley S., Janjic N., Ruckman J., Eitner F. and Floege J. (2002). The effects of platelet-derived growth factor antagonism in experimental glomerulonephritis are independent of the transforming growth factor-ß system. J. Am. Soc. Nephrol. 13, 658-657.

Ostendorf T., Rong S., Boor P., Wiedemann S., Kunter U., Haubold U., van Roeven C.R., Eitner F., Kawachi H., Starling G., Alvarez E., Smithson G. and Floege J. (2006). Antagonism of PDGF-D by human antibody CR002 prevents renal scarring in experimental glomerulonephritis. J. Am. Soc. Nephrol. 17, 1054-1062.

Otsuka G., Stempien-Otero A., Frutkin A.D. and Dichek D.A. (2007). Mechanisms of TGF-ß1-induced intimal growth. Plasminogen-independent activities of plasminogen activator inhibitor-1 and heterogeneous origin of intimal cells. Circ. Res. 100, 1300-1307.

Patek C.E., Fleming S., Miles C.G., Bellamy C.O., Ladomery M., Spraggon L., Mullins J., Hastie N.D. and Hooper M.L. (2003). Murine Denys-Drash syndrome: evidence of podocyte de-differentiation and systemic mediation of glomerulosclerosis. Hum. Mol. Genet. 12, 2379-2394.

Porst M., Daniel C., Plank C., Schocklmann H.O., Reinhardt D.P. and Hartner A. (2006). Induction and coexpression of latent transforming growth factor ß-binding protein-1 and fibrillin-1 in experimental glomerulonephritis. Nephron Exp. Nephrol. 102, e99-e104.

Riser B.L., Denichilo M., Cortes P., Baker C., Grondin J.M., Yee J. and Narins R.G. (2000). Regulation of connective tissue growth factor activity in cultured rat mesangial cells and its expression in experimental diabetic glomerulosclerosis. J. Am. Soc. Nephrol. 11, 25-38.

Shibata S., Nagase M., Yoshida S., Kawachi H. and Fujita T. (2007). Podocyte as the target for aldosteone. Roles of oxidative stress and Sgk1. Hypertension 49, 355-364.

Stein-Oakley A.N., Maguire J.A., Dowling J., Perry G. and Thomson N.M. (1997). Altered expression of fibrogenic growth factors in IgA nephropathy and focal and segmental glomerulosclerosis. Kidney Int. 51, 195-204.

Sung S.H., Ziyadeh F.N., Wang A., Pyagay P.E., Kanwar Y.S. and Chen S. (2006). Blockade of vascular endothelial growth factor signaling ameliorates diabetic albuminuria in mice. J. Am. Soc. Nephrol. 17, 3093-3104.

Taipale J., Koli K. and Keski-Oja J. (1992). Release of transforming growth factor-ß1 from the pericellular matrix of cultured fibroblasts and fibrosarcoma cells by plasmin and thrombin. J. Biol. Chem. 267, 25378-25384.

Trevillian P., Paul H., Millar E., Hibberd A. and Agrez M.V. (2004). αvß6 integrin expression in diseased and transplanted kidneys. Kidney Int. 66, 1423-1433.

Wahab N.A., Schaefer L., Weston B.S., Yiannikouris O., Wright A., Babelova A., Schaefer R. and Mason R.M. (2005). Glomerular expression of thrombospondin-1, transforming growth factor beta and connective tissue growth factor at different stages of diabetic nephropathy and their interdependent roles in mesangial response to diabetic stimuli. Diabetologia 48, 2650-2660.

Wakefield L.M., Winokur T.S., Hollands R.S., Christopherson K., Levinson A.D. and Sporn M.B. (1990). Recombinant latent transforming growth factor ß1 has a longer plasma half-life in rats than active transforming growth factor ß1, and a different tissue distribution. J. Clin. Invest. 86, 1976-1984.

Wang L., Kwak J.H., Kim S.I., He Y. and Choi M.E. (2004). Transforming growth factor-ß1 stimulates vascular endothelial growth factor 164 via mitogen-activated protein kinase kinase 3-p38α and p38δ mitogen-activated protein kinase-dependent pathway in murine mesangial cells. J. Biol. Chem. 279, 33213-33219.

Wang S., de Caestecker M., Kopp J., Mitu G., LaPage J. and Hirschberg R. (2006). Renal bone morphogenetic protein-7 protects against diabetic nephropathy. J. Am. Soc. Nephrol. 17, 2504-2512.

Wendt T.M., Tanji N., Guo J., Kislinger T.R., Qu W., Lu Y., Bucciarelli L.G., Rong L.L., Moser B., Markowitz G.S., Stein G., Bierhaus A., Liliensiek B., Arnold B., Nawroth P.P., Stern D.M., D'Agati V.D. and

Schmidt A.M. (2003). RAGE drives the develolpment of glomerulosclerosis and implicates podocyte activation in the pathogenesis of diabetic nephropathy. Am. J. Pathol. 162, 1123-1137.

Weston B.S., Wahab N.A. and Mason R.M. (2003). CTGF mediates TGF-ß-induced fibronectin matrix deposition by upregulating active α5ß1 integrin in human mesangial cells. J. Am. Soc. Nephrol. 14, 601-610.

Whaley-Connell A., Habibi J., Nistala R., Cooper S.A., Karuparthi P.R., Hayden M.R., Rehmer N., DeMarco V.G., Andresen B.T., Wei Y., Ferrario C. and Sowers J.R. (2008). Attenuation of NADPH oxidase activation and glomerular filtration barrier remodeling with statin treatment. Hypertension 51, 474-480.

Whaley-Connell A.T., Chowdhury N.A., Hayden M.R., Stump C.S., Habibi J., Wiedmeyer C.E., Gallagher P.E., Tallant E.A., Cooper S.A., Link C.D., Ferrario C. and Sowers J.R. (2006). Oxidative stress and glomerular filtration barrier injury: role of the renin-angiotensin system in the Ren2 transgenic rat. Am. J. Physiol. Renal Physiol. 291, F1308-F1314.

Wolf G., Chen S. and Ziyadeh F.N. (2005). From the periphery of the glomerular capillary wall toward the center of disease: podocyte injury comes of age in diabetic nephropathy. Diabetes 54, 1626-1634.

Xu J., Yao B., Fan X., Langworthy M.M., Zhang M-Z. and Harris R.C. (2007). Characterization of a putative intrarenal serotonergic system. Am. J. Physiol. Renal Physiol. 293, F1468-F1475.

Yamabe H., Osawa H., Kaizuka M., Tsunoda S., Shirato K-i., Tateyama F. and Okumura K. (2000). Platelet-derived growth factor, basic fibroblast growth factor, and interferon γ increase type IV collagen production in human fetal mesangial cells via a transforming growth factor-ß-dependent mechanism. Nephrol. Dial. Transplant. 15, 872-876.

Yokoi H., Mukoyama M., Mori K., Kasahara M., Suganami T., Sawai K., Yoshioka T., Saito Y., Ogawa Y., Kuwabara T., Sugawara A. and Nakao K. (2008). Overexpression of connective tissue growth factor in podocytes worsens diabetic nephropathy in mice. Kidney Int. 73, 446-455.

Yu L., Border W.A., Anderson I., McCourt M., Huang Y. and Noble N.A. (2004). Combining TGF-ß inhibition and angiotensin II blockade results in enhanced antifibrotic effect. Kidney Int. 66,1774-1784.

Yu Q. and Stamenkovic I. (2000). Cell surface-localized matrix metalloproteinase-9 proteolytically activates TGF-ß and promotes tumor invasion and angiogenesis. Genes Dev. 14, 163-176.

Zheng S., Carlson E.C., Yang L., Kralik P.M., Huang Y. and Epstein P.N. (2008). Podocyte-specific overexpression of the antioxidant metallothionein reduces diabetic nephropathy. J. Am. Soc. Nephrol. 19, 2077-2085.

Ziyadeh F.N. and Wolf G. (2008). Pathogenesis of the podocytopathy and proteinuria in diabetic glomerulopathy. Curr. Diabetes Rev. 4, 39-45.

Ziyadeh F.N., Hoffman B.B., Han D.C., Iglesias-de la Cruz M.C., Hong S.W., Isono M., Chen S., McGowan T.A. and Sharma K. (2000). Long-term prevention of renal insufficiency, excess matrix gene expression, and glomerular mesangial matrix expansion by treatment with monoclonal antitransforming growth factor-ß antibody in db/db diabetic mice. Proc. Natl. Acad. Sci. USA 97, 8015-8020.

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