

## EDITORIAL

### THE RENIN–ANGIOTENSIN SYSTEM AND PERITONEAL DIALYSIS

During continuous ambulatory peritoneal dialysis (CAPD), various morphological changes take place in the peritoneum, including mesothelial denudation, interstitial fibrosis, neovascularization, and such vascular alterations as the replication of basement membrane and fibrosis and hyalinization of the vascular wall (1–8). Among the suggested causes of these histological and functional alterations are recurrent peritonitis, the influence of plasticizers and advanced glycosylation end-products, and response to the nonphysiological nature of peritoneal dialysis (PD) solutions, in particular its high glucose content, hypertonicity, lactate, and low pH (8–10). Although the mechanisms underlying these alterations are not completely understood, it has been shown that growth factors and cytokines secreted by mesothelial cells (MC) and macrophages are associated with peritoneal fibrosis (11). Dobbie suggested that the initiating events that lead to peritoneal fibrosis and injury to MC are chronic irritation by dialysis solutions and severe or prolonged peritonitis (12). Damaged MC can cause peritoneal fibrosis by secreting extracellular matrix macromolecules (ECM) consisting of collagen, fibronectin, laminin, proteoglycans, and cytokines, including transforming growth factor beta 1 ( $TGF\beta_1$ ) and interleukin-1.

A key mediator of ECM accumulation in fibrotic diseases,  $TGF\beta_1$  has several properties that cause fibrosis. These include (1) increased synthesis of matrix proteins such as collagens, fibronectin, and proteoglycans; (2) decreased degradation of matrix proteins by suppression of matrix metalloproteinase (MMP) expression and increased production of protease inhibitors, such as tissue inhibitor of matrix metalloproteinase (TIMP) and plasminogen activator inhibitor type 1; and (3) increased synthesis of integrins (13). *In vitro*,  $TGF\beta_1$  orchestrates sequential cellular events that include loss of epithelial adhesion, F-actin reorganization, disruption of basement membrane integrity, and enhanced cell migration and

invasion (14). Reduction of  $TGF\beta_1$  overexpression decreases pathological matrix accumulation.

The  $TGF\beta$  superfamily regulates cell proliferation, differentiation, adhesion, and apoptosis, and thus controls embryonic development, tissue recycling, and wound repair (15).  $TGF\beta$  binds directly to the  $TGF\beta$  receptors II (a constitutively active transmembrane serine/threonine kinase that recruits  $TGF\beta$  receptors) and I, and phosphorylates one or more substrates to initiate a signal cascade such as that of Smad proteins and integrin-linked kinase (14).

Angiotensin II (ANG II), the main peptide of the renin–angiotensin system (RAS), is an oligopeptide of eight amino acids, formed from its precursor, angiotensinogen, by a series of two enzymatic cleavages. The effects of ANG II are mediated by binding to specific ANG II receptors: AT1 and AT2. Vascular and renal tubular actions are mediated primarily by AT1 receptors. The effects of AT2 receptors are less well understood; they may play a role of regulation of cell proliferation in the arterial wall. It is now clear, however, that there are extra-renal RAS and that ANG II can be synthesized at a variety of sites, including the kidney, vascular endothelium, adrenal gland, and brain. It is presumed that local ANG II production is important for the regulation of local processes that are activated by local factors such as prostaglandin, nitric oxide, and endothelin. Angiotensin II is considered a growth factor that regulates cell proliferation, apoptosis, and fibrosis. In models of renal injury, blockage of ANG II action via angiotensin-converting-enzyme inhibitors (ACE-I) or angiotensin-receptor blockers (ARBs) decreases proteinuria, inflammatory-cell infiltration, fibrosis, and gene expression of matrix proteins and growth factors (13,16,17). RAS blockers are commonly used in the treatment of hypertension and proteinuria in humans (18).

Angiotensin II stimulates macrophages and fibroblast-like cells to secrete  $TG\beta_1$ . A perivascular/

interstitial fibrosis, for instance, accompanies chronic elevation of either circulating ANG II or aldosterone (19) and, in the case of ANG II, occurs in response to abnormal vascular permeability and escape of macromolecules (20). ANG II also regulates the synthesis of proinflammatory cytokines (tumor necrosis factor  $\alpha$ , interleukin-6) and chemokines (monocyte chemoattractant protein-1) in the kidney (21). Importantly, ANG II may have a direct effect on downstream modulators of collagen metabolism, that is, MMPs, TIMP (22), and collagen tissue growth factor (23). The MMPs seem to play an increasingly prominent role as inflammatory mediators in the regulation of ECM proteins and cell proliferation via the TGF $\beta_1$  pathway (20). There is also some evidence of TGF $\beta_1$ -independent pathways of ANG II-induced fibrosis (24).

It has been shown that activated macrophages isolated from peritoneal exudates (formed after instillation of mineral oil into the peritoneal space) express ANG II receptors (25).

Peritoneal matrix accumulation is characteristic of peritoneal fibrosis. Continuous ambulatory PD patients who had persistent TGF $\beta_1$  in their effluent had an increased risk of peritoneal fibrosis. We know that TGF $\beta_1$  regulates cell proliferation, differentiation, adhesion, and apoptosis, and thus controls tissue recycling and wound repair. TGF $\beta_1$  binds directly to TGF $\beta$  receptors and phosphorylates one or more substrates to initiate a signal cascade, such as that of Smad proteins. Seven human Smad proteins have been identified. Smad1 is thought to be a mediator of bone morphogenic protein signaling, whereas Smad2 and Smad3 are responsible for TGF $\beta$  and activin signaling (26).

Recent studies have demonstrated increased evidence of ANG II involvement in the regulation of angiogenic cytokines, such as vascular endothelial growth factor (VEGF) and angiopoietin, and their receptors (27). ANG II plays a role in promoting angiogenesis. Rizkalla *et al.* clearly demonstrated that ANG II is a major stimulus of angiogenic cytokines and their receptors. Specifically, both AT1- and AT2-receptor-blockades antagonize the actions of ANG II on the expression of VEGF and angiopoietin (28).

Some *in vitro* studies have investigated the effects of RAS on peritoneum. Yao *et al.*, who investigated the role of the TGF $\beta_1$ -Smad pathway in preventing peritoneal fibrosis in human peritoneal mesothelial cells (HPMC), showed that dialysis with high glucose upregulated the expression of Smad2 and Smad4, whereas conditions of high osmolarity (as with mannitol) did not affect the TGF $\beta_1$ -Smad signaling pathway. They also showed that ARB (losartan) inhibited the concentration of TGF $\beta_1$  and the expression of Smad2. They concluded that ARBs might represent a

possible way to prevent and treat fibrosis of the peritoneum in long-term PD patients (29).

Noh *et al.* investigated the possibility that an independent RAS was operating in HPMC and examined the effects of glucose on it. They demonstrated that HPMC have an independent RAS and that high glucose concentrations upregulate it. ANG II mediates high-glucose-induced fibronectin secretion by HPMC, and blockade of RAS by the ACE-I imidapril may have a therapeutic benefit in long-term PD (30).

Using an encapsulating peritoneal sclerosis model in mice, Sawada *et al.* also showed that the ACE-I quinapril ameliorates peritoneal fibrosis (31).

Abe *et al.* investigated the role of bone-marrow-derived cells in the formation of peritoneal fibrotic tissue and the importance of ANG II type 1 receptor in bone-marrow-derived cells in the progression of peritoneal fibrosis in a chlorhexidine-induced fibrosis model in mice. They showed that ANG II acting via AT1 receptor in bone-marrow-derived cells might play a pivotal role in the progression of peritoneal fibrosis by elevating the level of growth factors and chemokines (32).

Imai *et al.* investigated changes in the expression of aquaporins (AQPs) in the peritoneum in rats and examined the effects of the RAS on the expression of AQPs with and without an ACE-I (benazepril) or an ARB (valsartan). Treatment with benazepril or valsartan significantly suppressed the expression of AQP-1 and AQP-4. Those authors suggest that the RAS plays an important role in the regulation of water transport in the peritoneum (33).

Using animal models, we investigated the effect of ACE-I or ARBs, or a combination of both, on peritoneal alterations induced by hypertonic PD solutions. Thus, in a rat model of once-daily injection of 3.86% glucose PD fluid for 4 weeks, we found that (1) oral administration of enalapril ameliorates changes in peritoneal function and morphology (34), (2) oral administration of lisinopril (an ACE-I) and valsartan (an ARB) have similar beneficial effects on peritoneal function and morphology (35), and (3) oral administration of quinapril plus valsartan has no synergistic effects on peritoneal alterations induced by 3.86% glucose PD solutions (36). On histological examination, we found less cell infiltration and less vascularization in all treated groups. According to results of Rizkalla *et al.* (28), inhibition of angiogenesis by ANG II blockage might partially explain the functional benefits observed in our experiments (34). Based on these findings, we concluded that RAS blockade ameliorates hypertonic PD solution-induced peritoneal injury by inhibiting the overexpression of cytokines (*i.e.*, TGF $\beta_1$  and VEGF), and that ACE-I and/or ARBs might preserve the viability of peritoneum in CAPD patients over the long term.

In the clinical setting, in a pilot study, Agraharkar *et al.* investigated the effects of RAS blockage on peritoneal protein loss in PD patients. They concluded that RAS blockage did not reduce protein loss into the peritoneal fluid during dialysis (37).

ACE-I and ARBs may have an important if indirect role in PD by affecting the rate of decline of residual renal function in PD patients, especially in those with diabetic nephropathy (38,39) and chronic proteinuric nephropathy (40,41). In a randomized study of 60 patients on PD, Li *et al.* investigated the effect of ACE-I on residual renal function. Over a 12-month period, the average residual glomerular filtration rate (GFR) declined by 2.07 mL/minute/1.73 m<sup>2</sup> in the ramipril (5 mg/day) group versus 3.00 mL/minute/1.73 m<sup>2</sup> in the control group ( $p = 0.03$ ). At 12 months, 14 patients in the ramipril group ( $n = 30$ ) and 22 in the control group ( $n = 30$ ) became anuric. Those authors concluded that the ACE-I ramipril might reduce the rate of decline of residual renal function in PD patients (42). Similar studies in patients with diabetic nephropathy and other chronic proteinuric nephropathies found that the benefits of ramipril seem to be independent of the level of systemic blood pressure.

In another study, Kikuta *et al.* examined the effects of valsartan on residual renal function in CAPD patients for 3 months to 2 years after the start of dialysis. Valsartan significantly retarded the progressive decline in residual renal function (residual GFR 4.1 to 3.2 vs 4.1 to 2.2 mL/min/1.73 m<sup>2</sup>) (43).

Ample evidence from *in vitro* experiments shows that the beneficial effect of ACE-I therapy is related to the paracrine effect of ANG II (accumulation of extracellular matrix) rather than its hemodynamic effect (44).

The plasma levels of ACE are modulated by ACE gene polymorphism. The polymorphism was classified into three genotypes: II, ID, and DD. An association between progressive renal disease and the DD genotype was found in a prospective follow-up study of 56 patients with type II diabetes (45) and in patients with type I diabetes (46). There are a few studies in the literature that clarify the role of genes related to ACE in PD patients. In one of them, Nishina, from Japan, investigated polymorphism of the ACE gene in 60 PD patients and 50 patients undergoing hemodialysis (47). The author found a significant difference in allele frequency between normal subjects ( $n = 100$ ) (I = 0.63, D = 0.37) and dialysis patients ( $n = 110$ ) (I = 0.46, D = 0.54,  $p < 0.001$ ). The mean plasma ACE activity and the mean rate of decrease in residual urinary volume were higher with the DD genotype ( $p < 0.05$ ). The mean rate of decrease in residual urinary volume was positively correlated with plasma ACE activity ( $r =$

0.133 89,  $p < 0.02$ ). The mean cardiothoracic ratio was higher in PD patients with the DD genotype. The author concluded that PD patients with the DD genotype lost residual renal function more rapidly and had a larger heart than patients with the other genotypes (47).

In another study, Wang *et al.* investigated cardiac hypertrophy and remodeling in relation to ACE and angiotensinogen genotypes in Chinese PD patients. They found that polymorphism of the angiotensinogen M235T gene, but not the ACE I/D gene, is associated with greater left ventricular mass and relative wall thickness, indicating more concentric left ventricular hypertrophy, in Chinese PD patients (48).

Finally, Varagunam *et al.* found a significant difference in erythropoietin requirement in the II/ID group compared to the DD group. They concluded that ACE genotype has predictive value when determining erythropoietin dosage, as the II/ID genotype may be associated with a suboptimal response (49).

What we learned from these three studies is that gene polymorphism is also important for evaluating PD patients, and we need to clarify the relation between gene polymorphism and transport status and developing peritoneal fibrosis.

## CONCLUSION

The evidence to date suggests that the renin-angiotensin system plays a role in high-glucose-induced peritoneal injury by regulating proinflammatory cytokines, increasing aquaporin expression, and increasing progression of fibrosis. Evidence is emerging that ACE-I and ARBs may have a beneficial effect in preventing the long-term alterations of peritoneal membrane induced by high glucose solutions, and possibly decreasing the rate of decline of residual renal function in these patients. Well-designed, randomized controlled trials are required to determine whether the renin-angiotensin system and its blockade will play an important role in the long-term outcome of PD patients.

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## REFERENCES

1. Gokal R. Taking peritoneal dialysis beyond the year 2000. *Perit Dial Int* 1999; 19(Suppl 3):S35–42.
2. Davies SJ, Bryan J, Phillips L, Russell GI. Longitudinal changes in peritoneal kinetics: the effects of peritoneal dialysis and peritonitis. *Nephrol Dial Transplant* 1996; 11:498–506.
3. Selgas R, Fernandez-Reyes M-J, Bosque E, Bajo M-A, Borrego F, Jimenez C, et al. Functional longevity of the human peritoneum: how long is continuous peritoneal dialysis possible? Results of a prospective median long-term study. *Am J Kidney Dis* 1994; 23:64–73.
4. Heimbürger O, Waniewski J, Werynski A, Tranæus A, Lindholm B. Peritoneal transport in CAPD patients with permanent loss of ultrafiltration capacity. *Kidney Int* 1990; 38:495–506.
5. Di Paolo N, Sacchi G. Atlas of peritoneal histology. *Perit Dial Int* 2000; 20(Suppl 3):S5–96.
6. Williams JD, Craig KJ, Topley N, Von Ruhland C, Fallon M, Newman GR, et al. Peritoneal Biopsy Study Group. Morphologic changes in the peritoneal membrane of patients with renal disease. *J Am Soc Nephrol* 2002; 13:470–9.
7. Honda K, Nitta K, Horita S, Yumura W, Nihei H. Morphological changes in the peritoneal vasculature of patients on CAPD with ultrafiltration failure. *Nephron* 1996; 72:171–6.
8. Rubin J, Herrera GA, Collins D. An autopsy study of the peritoneal cavity from patients on continuous ambulatory peritoneal dialysis. *Am J Kidney Dis* 1991; 18:97–102.
9. Fracasso A, Baggio B, Ossi E, Del Prete D, Bonfante L, Bazzato G, et al. Glycosaminoglycans prevent the functional and morphological peritoneal derangement in an experimental model of peritoneal fibrosis. *Am J Kidney Dis* 1999; 33:105–10.
10. Honda K, Nitta K, Horita S, Yumura W, Nihei H, Nagai R, et al. Accumulation of advanced glycation end products in the peritoneal vasculature of continuous ambulatory peritoneal dialysis patients with low ultrafiltration. *Nephrol Dial Transplant* 1999; 14:1541–9.
11. Chaimovitz C. Peritoneal dialysis. *Kidney Int* 1994; 45:1226–40.
12. Dobbie JW. Pathogenesis of peritoneal fibrosing syndromes (sclerosing peritonitis) in peritoneal dialysis. *Perit Dial Int* 1992; 12:14–27.
13. Ruiz-Ortega M, Lorenzo O, Suzuki Y, Ruperez M, Egido J. Proinflammatory actions of angiotensins. *Curr Opin Nephrol Hypertens* 2001; 10:321–9.
14. Yang J, Liu Y. Dissection of key events in tubular epithelial to myofibroblast transition and its implications in renal interstitial fibrosis. *Am J Pathol* 2001; 159:1465–75.
15. Strutz F, Muller GA. Transdifferentiation comes of age. *Nephrol Dial Transplant* 2000; 15:1729–31.
16. Wolf G, Neilson EG. Angiotensin II as a renal growth factor. *J Am Soc Nephrol* 1993; 3:1531–40.
17. Mezzano SA, Ruiz-Ortega M, Egido J. Angiotensin II and renal fibrosis. *Hypertension* 2001; 38:635–8.
18. Andersen S, Tarnow L, Rossing P, Hansen BV, Parving HH. Renoprotective effects of angiotensin II receptor blockade in type 1 diabetic patients with diabetic nephropathy. *Kidney Int* 2000; 57:601–6.
19. Border WA, Noble NA. TGF  $\beta$  in kidney fibrosis: a target for gene therapy. *Kidney Int* 1997; 51:1388–96.
20. Peters H, Border WA, Noble NA. Targeting TGF  $\beta$  overexpression in renal disease: maximizing the antifibrotic action of angiotensin II blockade. *Kidney Int* 1998; 54:1570–80.
21. Ruiz-Ortega M, Ruperez M, Lorenzo O, Esteban V, Blonco J, Mezzano S, et al. ANG II also regulates the synthesis of proinflammatory cytokines and chemokines in the kidney. *Kidney Int Suppl* 2002; 82:S12–22.
22. Lods N, Ferrari P, Frey FJ, Kappeler A, Berthier C, Vogt B, et al. Angiotensin-converting enzyme inhibition but not angiotensin II receptor blockade regulates matrix metalloproteinase activity in patients with glomerulonephritis. *J Am Soc Nephrol* 2003; 14:2861–72.
23. Liu BC, Sun J, Chen Q, Ma KL, Ruan XZ, Phillips AO. Role of connective tissue growth factor in mediating hypertrophy of human proximal tubular cells induced by angiotensin II. *Am J Nephrol* 2003; 23:429–37.
24. Ma LJ, Yang H, Gaspert A, Carlesso G, Barty MM, Davidson JM, et al. Transforming growth factor-beta-dependent and -independent pathways of induction of tubulointerstitial fibrosis in beta6(-/-) mice. *Am J Pathol* 2003; 163:1261–73.
25. Thomas DW, Hoffman MD. Identification of macrophage receptor for angiotensin: a potential role in antigen uptake for T lymphocyte responses? *J Immunol* 1984; 132:2807–12.
26. Lagna G, Hata A, Massagué J. Partnership between DPC4 and SMAD protein in TGF-beta signalling pathways. *Nature* 1996; 383:832–6.
27. Fujiyama S, Matsubara H, Nozawa Y, Maruyama K, Mori Y, Tsutsumi Y, et al. Angiotensin AT(1) and AT(2) receptors differentially regulate angiotensin-2 and vascular endothelial growth factor expression and angiogenesis by modulating heparin binding-epidermal growth factor (EGF)-mediated EGF receptor transactivation. *Circ Res* 2001; 88:22–9.
28. Rizkalla B, Forbes JM, Cooper ME, Cao Z. Increased renal vascular endothelial growth factor and angiotensin II infusion is mediated by both AT1 and AT2 receptors. *J Am Soc Nephrol* 2003; 14:3061–71.
29. Yao Q, Lindholm B, Qian J. Inhibition of the effect of high glucose (HG) on the expression of Smad in human peritoneal mesothelial cells [Abstract]. *J Am Soc Nephrol* 2003; 14:215A.
30. Noh H, Kim HJ, Yu MR, Ha H, Lee HB. Renin angiotensin system in human peritoneal mesothelial cells [Abstract]. *J Am Soc Nephrol* 2003; 14:218A.
31. Sawada T, Ishii Y, Tojimbara T, Nakajima I, Fuchinoue S, Teraoka S. The ACE inhibitor, quinapril, ameliorates peritoneal fibrosis in an encapsulating peritoneal sclerosis model in mice. *Pharmacol Res* 2002; 46:505–10.
32. Abe K, Miyazaki M, Yoshio Y, Furusu A, Harada T, Sugaya T, et al. Importance of angiotensin II type 1

- receptor in bone marrow derived cells in the progression of peritoneal fibrosis [Abstract]. *J Am Soc Nephrol* 2003; 14:36A.
33. Imai H, Nakamoto H, Ishida Y, Yamanouchi Y, Inoue T, Okada H, *et al.* Renin-angiotensin system plays an important role in the regulation of water transport in the peritoneum. *Adv Perit Dial* 2001; 17:20-4.
  34. Duman S, Gunal AI, Sen S, Asci G, Ozkahya M, Terzioglu E, *et al.* Does enalapril prevent peritoneal fibrosis induced by hypertonic (3.86%) peritoneal dialysis solution? *Perit Dial Int* 2001; 21:219-24.
  35. Duman S, Şen S, Aşçi G, Başçi A, Akçiçek F. Effect of valsartan versus lisinopril on peritoneal alterations in rats [Abstract]. *Perit Dial Int* 2002; 22:112.
  36. Duman S, Şen S, Duman C, Aşçi G, Basci A, Akcicek F. Improvement of peritoneal alterations induced by hypertonic PD solutions by ACE inhibitors and by AR blockers [Abstract]. *Nephrol Dial Transplant* 2003; 18(Suppl 4):s474.
  37. Agraharkar M, Du Y, Man-Wan C, Henry S, Kuo YF, Ahuja T. Angiotensin II receptor blockade (ARB) and peritoneal protein loss in peritoneal dialysis patients [Abstract]. *J Am Soc Nephrol* 2003; 14:858A.
  38. Andersen S, Tarnow L, Rossing P, Hansen BV, Parving HH. Renoprotective effects of angiotensin II receptor blockade in type 1 diabetic patient with diabetic nephropathy. *Kidney Int* 2000; 57:601-6.
  39. Bauer JH, Reams GP, Hewett J, Klachko D, Lau A, Messina C, *et al.* A randomized, double blind, placebo-controlled trial to evaluate the effect of enalapril in patients with clinical diabetic nephropathy. *Am J Kidney Dis* 1992; 20:443-57.
  40. Ruggenti P, Perna A, Gherardi G, Gaspari F, Benini R, Remuzzi G. Renal function and requirement for dialysis in chronic nephropathy patients on long-term ramipril: REIN follow-up trial. Gruppo Italiano di Studi Epidemiologici in Nefrologia (GISEN). Ramipril Efficacy in Nephropathy. *Lancet* 1998; 352:1252-6.
  41. Ruggenti P, Perna A, Gherardi G, Garini G, Zoccali C, Salvadori M, *et al.* Renoprotective properties of ACE-inhibition in non-diabetic nephropathies with non-nephrotic proteinuria. *Lancet* 1999; 354:359-64.
  42. Li PK, Chow KM, Wong TY, Leung CB, Szeto CC. Effects of an angiotensin-converting enzyme inhibitor on residual renal function in patients receiving peritoneal dialysis. A randomized, controlled study. *Ann Intern Med* 2003; 139:105-12.
  43. Kikuta T, Watanabe Y, Kanno Y, Tomori K, Nakamoto H, Okada H, *et al.* Effects of angiotensin II-receptor blocker, valsartan, on residual renal function in patients on CAPD [Abstract]. *J Am Soc Nephrol* 2003; 14:482A.
  44. Kagami S, Border WA, Miller DE, Noble NA. Angiotensin II stimulates extracellular matrix protein synthesis through induction of transforming growth factor-beta expression in rat glomerular mesangial cells. *J Clin Invest* 1994; 93:2431-7.
  45. Fava S, Azzopardi J, Ellard S, Hattersley AT. ACE gene polymorphism as a prognostic indicator in patients with type 2 diabetes and established renal disease. *Diabetes Care* 2001; 24:2115-20.
  46. Azar ST, Zalloua PA, Medlej R, Halabi G. The DD genotype of the ACE gene polymorphism is associated with diabetic nephropathy in the type-1 diabetics. *Endocr Res* 2001; 27:99-108.
  47. Nishina M. A study on angiotensin-I converting enzyme polymorphism in CAPD patients. *Nippon Jinzo Gakkai Shi* 1996; 38:595-602.
  48. Wang AY, Chan JC, Wang M, Poon E, Lui SF, Li PK, *et al.* Cardiac hypertrophy and remodeling in relation to ACE and angiotensinogen genes genotypes in Chinese dialysis patients. *Kidney Int* 2003; 63:1899-907.
  49. Varaganam M, McCloskey DJ, Sinnott PJ, Raftery MJ, Yaqoob MM. Angiotensin-converting enzyme gene polymorphism and erythropoietin requirement. *Perit Dial Int* 2003; 23:111-15.

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