



Effects of candesartan on the proteinuria of chronic glomerulonephritis

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Angiotensin I-converting enzyme (ACE) inhibitors are commonly used for the treatment of hypertension, progressive chronic renal disease, diabetic nephropathy, and congestive heart failure. Because angiotensin II acts through membrane bound type 1 (AT₁) and type 2 (AT₂) receptors, ACE inhibitors and angiotensin II-receptor antagonists have distinct effects. ACE inhibitors inhibit production of angiotensin II thus suppressing the action of angiotensin II on both AT₁ and AT₂. In contrast, the effect of AT₁-receptor antagonists is to selectively block the activation of the AT₁ receptor. This AT₁-receptor blockade leaves the AT₂ receptors unopposed to

elevated levels of endogenous angiotensin II. Thus, there may be an advantage of AT₁-receptor blockade over ACE inhibition in the management of a variety of chronic vascular diseases, including chronic glomerulonephritis and other glomerular diseases. In a clinical trial candesartan, an AT₁-receptor antagonist, effectively lowered urinary protein excretion in patients with chronic glomerular nephritis. Evidence indicates that functionally active AT₁ receptors, as well as AT₂ receptors, are present in both afferent and efferent arteriole of the glomerulus, and that angiotensin II induces afferent and efferent arteriolar dilatation via AT₂ receptors.

Keywords: glomerulonephritis; angiotensin II; angiotensin II type 1 receptor; angiotensin II type 2 receptor; candesartan; ACE

Introduction

Angiotensin I-converting enzyme (ACE) inhibitors have been widely used in clinical practice for the treatment of disorders such as hypertension and congestive heart failure. In addition, these agents have been shown to be effective in preventing the progression of chronic renal disease, independent from their effect of lowering systemic blood pressure. ACE inhibitors reduce glomerular hydraulic pressure by blocking the preferential action of angiotensin II to constrict the glomerular efferent arterioles. Thus, in addition to their widely recognised effect as antihypertensive drugs, ACE inhibitors may also be regarded as a useful agent for renal disorders and thus could be called 'renal pills'. In fact, clinical studies have shown that they can be used to manage normotensive patients with chronic renal disease associated with primary glomerulonephritis and diabetic glomerulosclerosis.

The major action of ACE inhibitors seems to be inhibition of the production of angiotensin II rather than the degradation of bradykinin, although there is some controversy regarding the role of bradykinin in the overall effects of ACE inhibitors. Since it is known that angiotensin II acts through membrane-bound type 1 (AT₁) and type 2 (AT₂) receptors, ACE inhibitors and angiotensin II-receptor antagonists could have, in theory, distinct effects. There are a number of reasons for these differences. First, there are several pathways leading to the production of angiotensin II that are independent of ACE. Second, it has been well recognised that activation of AT₁ receptors by angiotensin II induces contraction and

proliferation of vascular smooth-muscle cells, stimulation of proximal tubular sodium reabsorption, and activation of tubuloglomerular feedback. These effects of angiotensin II mediate, in part, the hypertensive action of angiotensin II. Therefore, both ACE inhibitors and AT₁-receptor antagonists are able to lower systemic blood pressure. The third difference relates to the possibility of bradykinin action, which, although it may be small, is always present with ACE inhibition. By contrast, activation of the AT₂ receptor by angiotensin II has been suggested to have an apoptotic and antiproliferative action in vascular smooth-muscle cells and other cell types examined.

In contrast to the effect of ACE inhibitors, the effect of AT₁-receptor antagonists is to selectively block the activation of the AT₁ receptor. This blockade will elevate levels of endogenous angiotensin II, and lead to activation of the AT₂ receptors. This may theoretically be an additional advantage of AT₁-receptor blockade over ACE inhibition in the management of a variety of chronic vascular diseases, including chronic glomerulonephritis and other glomerular diseases, since it has been suggested that the AT₂ receptor activation may often offset the activation of AT₁ receptors in some cell types including vascular smooth-muscle cells. However results from chronic renal disease rat models have shown that both ACE inhibitors and AT₁-receptor antagonists are equally effective.

Candesartan in primary chronic glomerulonephritis

Early clinical trial of candesartan for chronic glomerulonephritis

We performed a study investigating the effect of candesartan in patients with primary chronic glomeru-

lonephritis with proteinuria. Patients enrolled in the study were aged between 20 to 65 years, and had proteinuria, with urine protein excretion greater than 0.5 g/day, and a serum creatinine less than 2.0 mg/dl. Candesartan cilexetil, an angiotensin II type 1 (AT₁) receptor antagonist, was orally given daily for 8–12 weeks. Changes in urine protein excretion, widely recognised as one of the best predictors of progression of chronic renal disease (including primary glomerulonephritis), were used as the study end-point. The doses of candesartan were 2, 4, and 8 mg/day. Thirty-three patients were included, 10 receiving 2 mg/day, 12 receiving 4 mg/day, and 11 receiving 8 mg/day. The male:female ratio was 4:6, 4:8, and 6:5 for the 2, 4, and 8 mg/day groups, respectively. The majority of patients had mesangial proliferative glomerulonephritis with IgA nephropathy diagnosed in three, seven, and five patients in the 2, 4, and 8 mg/day groups, respectively. A small number of patients had systemic hypertension (two patients in the 2 mg/day group, one in the 4 mg/day, and four in the 8 mg/day groups).

A greater than 50% reduction in urinary protein excretion at 8–12 weeks was seen in approximately 50% of patients with each dose of candesartan. Seventy to 80% of patients treated with candesartan demonstrated a greater than 25% reduction in urine protein by 8–12 weeks (Table 1). This beneficial effect on protein excretion was independent of the effect on systemic blood pressure since blood pressure was normal in the majority of patients studied. The serum creatinine levels did not change and there was a slight minimal rise in serum potassium, which stayed within the normal range.

Dose-finding study of candesartan

Based on these results, we then conducted a more extensive, double-blind, controlled dose-finding study to identify the optimal dose of candesartan in chronic glomerulonephritis. Inclusion criteria were the same as for the earlier trial, as were patient demographics. The doses of 2, 4, and 8 mg/day, were again given for 12 weeks. Eighty-three, 86, and 85 patients were entered in the 2, 4, and 8 mg/day groups, respectively. The mean age (years) was 44.7, 45.1, and 45 for the 2, 4, and 8 mg/day groups respectively; urinary protein excretion was 2.0, 1.6, and 1.9 g/day for the 2, 4, and 8 mg/day groups, respectively. Serum creatinine was 1.03, 1.07, and 1.03 mg/dl for the 2, 4, and 8 mg/day groups, respectively. The majority (60 to 70%) of patients

Table 1 Reduction in urine protein excretion

Candesartan cilexetil (mg/day)	UV protein decrease		
	>50%	25–50%	Combined
2	4/9 *(10)	3/9 *(10)	7/9 *(10)
4	7/11 *(12)	2/11 *(12)	9/11 *(12)
8	5/11	3/11	8/11

*Discrepancy with numbers in the text reflects the number of patients judged ineligible for overall evaluation.

had mesangial proliferative glomerulonephritis, mostly IgA nephropathy.

Changes in urinary protein excretion during treatment with candesartan are summarised in Figure 1. The urinary protein excretion decreased by more than 50% in 16% of patients receiving 2 mg/day. This effect was dose-dependent, with 26% of patients receiving 4 mg/day, and 35% of those receiving 8 mg/day, demonstrating a greater than 50% reduction in urinary protein excretion. Overall, 50 to 60% of patients achieved a reduction in urinary protein of at least 25% in response to candesartan. These results demonstrated that candesartan effectively lowered urinary protein excretion in patients with chronic glomerular nephritis.

Angiotensin II receptor actions in the kidney

To elucidate the potential effects of AT₂-receptor activation in the kidney, a greater understanding of the mechanisms involved in both AT₁- and AT₂-receptor action is needed. It is known that AT₁-receptor antagonists will leave AT₂ receptors unopposed to elevated levels of endogenous angiotensin II. Available data suggests that AT₂-receptor activation may be associated with lowering of systemic blood pressure. Furthermore, Yerkes *et al*¹ have reported that AT₂ receptor knockout mice show a variety of kidney and urinary tract anomalies, which may obstruct urine outflow, primarily due to defective apoptosis in mesenchymal cells surrounding the area of urinary tract formation in the foetus. This resembles the CAKUT syndrome (congenital anomalies of kidney and urinary tract), which is the most common cause of an abdominal mass in the newborn. Indeed, Yerkes *et al*¹ showed the presence of mutations in the AT₂-receptor gene in these patients. The CAKUT syndrome provides evidence indicating the importance of AT₂-receptor activation during foetal development.

To gain evidence for the presence of angiotensin II receptors in the adult kidney, we need to turn to studies done by both Brenner and associates,² and Noda *et al*.³ These investigators examined the glomerulosclerosis model that develops following 5/6 nephrectomy in rats. In this model, glomerulosclerosis was inhibited by the ACE inhibitor, enalapril,

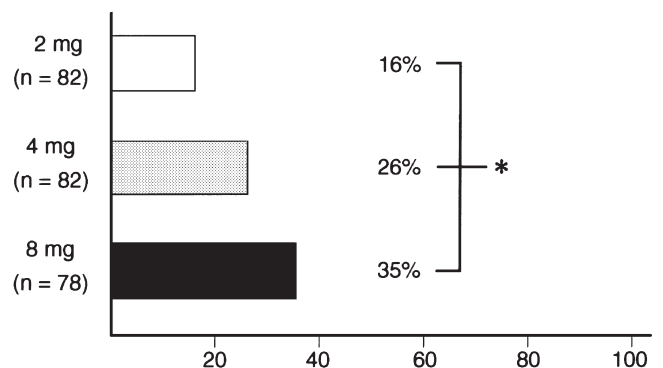


Figure 1 Efficacy (response rate): proportion of patients with reduction of urinary protein by 50% or more. Cochran–Armitage test; **P* = 0.007.

as well as by the AT₁-receptor antagonist, candesartan. While the precise molecular and cellular mechanisms underlying the development of glomerulosclerosis in the remnant kidney model are not well understood, a number of possibilities for the beneficial effect of these drugs have been proposed. These include correction of an increased glomerular hydraulic pressure, and hyperfiltration in the remnant kidney by blocking the action of angiotensin II on efferent arterioles.

Another marker found in glomerulosclerosis in the remnant kidney model is overexpression of transforming growth factor- β 1 (TGF- β 1), an agent known to trigger the sclerosing processes. Studies using the remnant kidney model have shown an increased staining for TGF- β 1 in the glomeruli.³ This elevated staining was prevented at 4 weeks by enalapril ($P < 0.01$), and by candesartan ($P < 0.05$). However, in a longer term follow-up at 16 weeks, while enalapril still inhibited the elevation of TGF- β 1 levels, this was less than that shown at 4 weeks, and candesartan now demonstrated greater activity (Figure 2). The reasons for this distinct effect of ACE inhibition and AT₁-receptor blockade are not yet clear.

Since glomerular hyperfiltration, and eventually, glomerulosclerosis, occur in the otherwise completely normal kidney that has simply had a mass reduction, glomerular haemodynamic change is likely to be critically involved as proposed by Brenner and associates. Recently, Ito and associates⁴ provided convincing evidence for the presence of functionally active AT₂ receptors regulating glomerular haemodynamics in the adult kidney. They performed studies using an isolated rabbit afferent arteriole system perfused *in vitro* and measured the change in diameter of the afferent arteriole. The effect of the AT₁-receptor antagonist, candesartan, which is the active metabolite of candesartan cilexetil, on angiotensin II action in afferent arterioles was measured in the presence or absence of the AT₂-receptor antagonist, PD123319. They found the vasoconstrictive effect of angiotensin II is blocked in a dose-dependent manner by increasing the AT₁ blockade. However, when PD123319 is added, selective blockade of the AT₂ receptor exaggerates the constriction of afferent arterioles in the presence of angiotensin II, suggesting that there is a functional AT₂ receptor present in afferent arterioles (Figure 3).

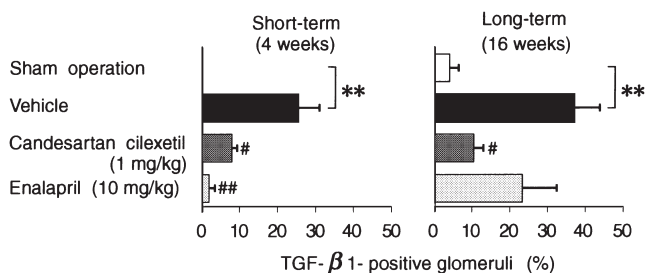


Figure 2 Effects of candesartan cilexetil and enalapril on TGF- β 1 expression in glomeruli in 5/6 nephrectomized rats. Mean \pm s.e.; ** $P < 0.01$ vs sham operation; # $P < 0.05$; ### $P < 0.01$ vs vehicle. (Reproduced from reference 3 with permission from the authors and the publisher).

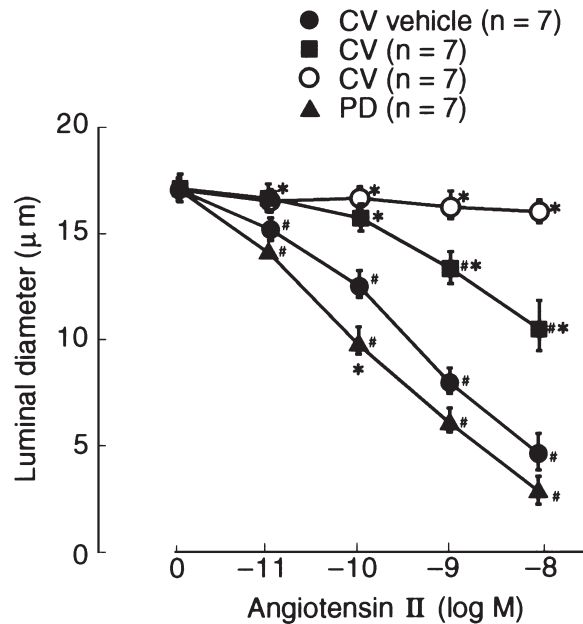


Figure 3 Effect of candesartan or PD123319 on angiotensin II-induced vasoconstriction in afferent arterioles. # $P < 0.01$ vs pre-angiotensin II; * $P < 0.01$ vs candesartan vehicle. (Reproduced from reference 4 with permission from the authors and the publisher).

A similar experiment was performed to confirm these results, in which the afferent arterioles were pre-constricted by noradrenaline. Increasing the dose of angiotensin II caused the pre-constricted afferent arterioles to further constrict. Addition of the AT₁-receptor antagonist, candesartan, led to vasodilatation that was completely blocked by candesartan plus PD123319. Therefore, it is clear that there is an AT₂ receptor for angiotensin II in afferent arterioles, and that stimulation of this receptor leads to vasodilatation. These effects are dependent on the presence of endothelium, since this effect was completely abolished by luminal perfusion of antibody against factor VIII-related antigen.

In addition, CGP42112, an AT₂-receptor agonist, induces afferent arteriole vasodilatation in the presence of noradrenaline-induced vasoconstriction. In addition to confirming that angiotensin II induces afferent arteriolar vasodilatation via AT₂ receptors, these results also suggest that the activation of AT₂ receptors leading to afferent arteriole vasodilatation is mediated by cytochrome P450-generated epoxygenase, as schematically shown (Figure 4).

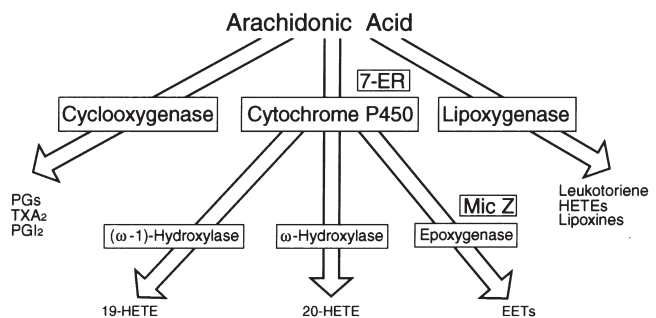


Figure 4 Cascade of events involved in the metabolism of arachidonic acid. (Reproduced from reference 4 with permission from the authors and the publisher).

Investigation into the effect on efferent arterioles has also been performed, showing that blockade of AT₂ receptors augments the angiotensin II-induced vasoconstriction *in vitro*. In efferent arterioles pre-constricted with noradrenaline, angiotensin II causes vasodilatation in the presence of the AT₁-receptor blockade candesartan, which again was completely eliminated by the addition of an AT₂-receptor antagonist. These data show that AT₂-receptor activation by angiotensin II causes efferent arteriolar dilatation. In pre-constricted efferent arterioles, the presence of noradrenaline, candesartan, and the addition of angiotensin II, leads to vasodilatation, as expected. However, this effect is completely obliterated in the presence of miconazole, which is known to inhibit the production of epoxyeicosatrienoic acid (EET).

Conclusion

It is clear that functionally active AT₁, as well as AT₂, receptors are present in both afferent and efferent arterioles, and that angiotensin II induces afferent and efferent arteriolar dilatation via AT₂ receptors. However, to elucidate the exact mechanisms, we need to know the regulation of tubuloglomerular feedback. For example, more detailed clarification of the effects of AT₂-receptor activation is needed. This is an important determinant of glomerular haemodynamics, of the tone of the afferent arterioles, and of renin secretion. These results may provide insight

into the mechanisms responsible for the differential effect of ACE inhibitors and AT₁-receptor blocking agents, such as candesartan, on glomerulosclerosis and glomerular TGF- β 1 expression in the remnant kidney model.

However, detailed analysis of the changing glomerular haemodynamics and tubuloglomerular feedback in the presence of AT₁-receptor blockade is absolutely necessary to advance our understanding of the biologic and clinical values of AT₁-receptor antagonists in the management of chronic renal disease. That is, the overall effects of angiotensin II via AT₂-receptor activation on glomerular haemodynamics and hydraulic pressure need to be determined to further predict the theoretical long-term effect of AT₁ receptor antagonism in chronic renal disease.

References

- 1 Yerkes E *et al.* Role of angiotensin in the congenital anomalies of the kidney and urinary tract in the mouse and the human. *Kidney Int* 1998; **54** (Suppl 67): S75–S77.
- 2 Mackenzie HS *et al.* Angiotensin receptor antagonists in experimental models of chronic renal failure. *Kidney Int* 1997; **52** (Suppl 63): S140–S143.
- 3 Noda M *et al.* Effects of candesartan cilexetil (TCV-116) and enalapril in 5/6 nephrectomized rats. *Kidney Int* 1997; **52** (Suppl 63): S136–S139.
- 4 Arima S *et al.* Possible role of P-450 metabolite of arachidonic acid in vasodilator mechanism of angiotensin II type 2 receptor in the isolated microperfused rabbit afferent arteriole. *J Clin Invest* 1997; **100**: 2816–2823.