The Renin System: Is Direct Renin Inhibition Different From Blockade at the AT_1 Receptor or the ACE Step?

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A substantial level of evidence supports the use of renin system blockade for many patients with hypertension. Two lines of evidence, based on very high-dose angiotensin blocker treatment or combination therapy with angiotensin-converting enzyme inhibitor and angiotensin receptor blocker, suggest that more complete blockade leads to improved clinical outcomes. The recent development of a powerful renin inhibitor that acts at the initial, rate-limiting step in the renin cascade would also favor more complete blockade of the system. For many patients, this is likely to lead to improved treatment.

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Although description of the renin system is truly venerable, the first publication appearing in 1897, progress in understanding the system was slow until the 1950s, when a creative explosion occurred. During that decade, the structures of angiotensin I (Ang I) and angiotensin II (Ang II) were identified,[1] and the angiotensin-converting enzyme (ACE) was described.[2] In this first report on ACE, Skeggs and his associates made a compelling argument that "since renin is the initial and rate-limiting substance in the renin–angiotensin system (RAS), it

would seem that renin inhibition . . . would be the most likely to succeed."[2] In fact, the development of renin inhibitors has lagged well behind blockade at the level of the ACE step or at the AT_1 receptor. The recent spate of reviews on renin inhibition is indicative of the recent development of renin inhibitors that are likely to have an important clinical impact.[3-9]

No one would minimize the contribution of ACE inhibition to the management of cardiovascular, metabolic, and renal disease. Indeed, most would trumpet this advance as one of the triumphs of modern therapeutics. Nevertheless, no pharmacologist would have chosen the ACE step as part of a planned approach to interrupting the renin system.[4] The development of ACE inhibitors was an unexpected byproduct of snake venom toxicology. The development of AT_1 receptor blockers was more planned, but its inception was accidental—a product of high-throughput screening that identified AT_1 receptor blockade by an unlikely molecule, an imidazole derivative.

In the hierarchy of evidence that supports the interesting notion of "evidence-based medicine," the large randomized, controlled clinical trial is at the top. These types of trials provide the most compelling evidence for similarity or difference between drug classes. There are very few such studies for ACE inhibitors and AT_1 receptor blockers, and none involving renin inhibition. The only comparative studies on renin inhibition involve blood pressure response, comparing a renin inhibitor with an ACE inhibitor or angiotensin receptor blocker (ARB) as reviewed elsewhere in this supplement. In the absence of such information, the evidence turns to information from studies of mechanism, drug pharmacology, pathophysiology of the disease, epidemiology and genetics, and tolerability of the agents available. These themes are developed in this article.

Particularly interesting to Skeggs and his coworkers[2] was the finding that the interaction of renin with its substrate is rate limiting. But what does the term "rate limiting" actually mean?

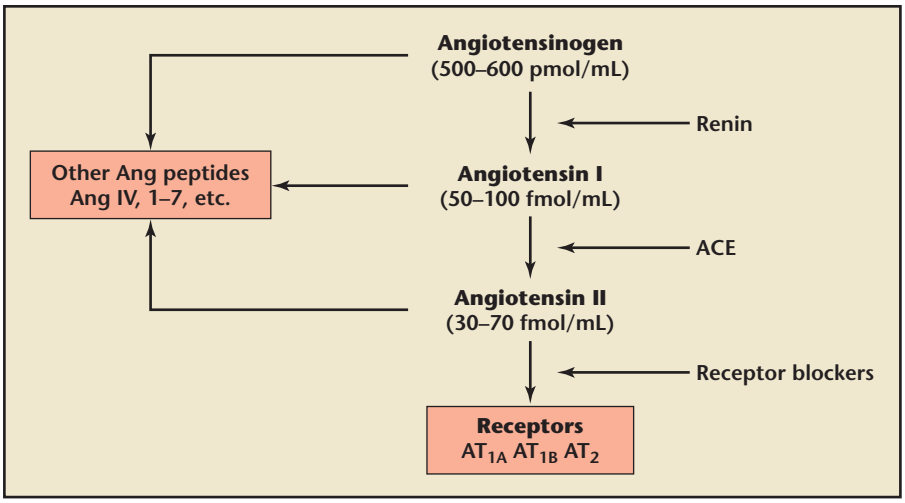
The Rate-Limiting Step

Three locations in the renin-angiotensin cascade are available for blockade: first, the interaction of renin with its substrate, angiotensinogen (Figure 1); next, the ACE step, the conversion of Ang I to Ang II; and finally, the AT_1 receptor. Some would add to this list the reduction in renin release induced by β-adrenergic blocking agents.

An enzyme cascade consists of several component steps, each of which may limit the rate of the overall reaction to a different extent. Each step is characterized by a rate constant, and the lowest rate constant is rate limiting, the determinant of the rate of overall reaction—in most but not all circumstances.[4] For example, a change in conditions may differently affect the specific activity of different enzymatic reactions. It has been argued as an alternative, however, that the rate-limiting step is the most sensitive step, the step that, if perturbed, can cause the largest change in overall velocity.

Unfortunately, most of us cannot translate a rate constant, much less a ratio of rate constants, into a physically meaningful model. As an alternative approach, Navar and colleagues[10] included in their description of the renin cascade not only the steps but also the concentrations of the various substrates and products along the way (Figure 1). Although the data in the figure represent findings in the rat, the findings in humans are very similar. The concentration of Ang II, the powerful vasoconstrictor and stimulus to aldosterone release, was in the range of 30 to 70 fmol/mL. The concentration of Ang I, the substrate for ACE and the precursor to Ang II, was about double the concentration of

Figure 1. *The renin cascade and the rate-limiting step. The concentrations of angiotensinogen, angiotensin I, and angiotensin II are shown in parentheses. Note that the concentration of angiotensin I is about twice that of angiotensin II, and the concentration of angiotensinogen, the renin substrate, is 5000-fold that of angiotensin I. The renin-catalyzed reaction is the rate-limiting step. Ang, angiotensin; ACE, angiotensin-converting enzyme. Adapted with permission from Fisher and Hollenberg.[3]*



Ang II, at 50 to 100 fmol/mL. Thus, the concentration gradient favored conversion catalyzed by ACE.

The concentration of the substrate angiotensinogen is in the neighborhood of 500 to 600 pmol/mL. Thus, the renin-catalyzed step from angiotensinogen to Ang I is favored by a 5000-fold concentration gradient. Clearly, if you want to block the system, the renin step should be the prime target. Blockade at this level can be more complete, for several reasons.

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Expectations: Models, Pathways, and Products

It is reasonable to ask, what are the expected responses to blocking each of the steps in the cascade? The expectations are determined mostly by the model one employs for understanding the system and the pathways and metabolic products represented in that model.

For example, if one accepts the classical view of the RAS as a system defined by concentrations of the relevant mediators in the circulation, as is still believed by some, then blockade at the renin step, the ACE step, and the AT_1 receptor should induce an equivalent response. Indeed, that is the finding when arterial blood pressure is used as the marker. It would be reasonable to argue that perhaps this classical view provides an adequate description of the relation between blood pressure and the renin system. Although there is substantial debate about whether different classes of antihypertensives influence natural history differently, no one seems to debate the efficacy of these various classes on blood pressure: they are very similar.[11] Thus, if there is to be a difference, it is going to be at the tissue level, where more effective blockade can affect the natural history of disease that goes beyond blood pressure.

There are a number of models other than the classical. As one example, some arguments have been made for significant non-ACE pathways in the generation of Ang II.[12] Should non-ACE pathways be important, then blockade induced by an AT_1 receptor blocker or renin inhibitor might produce a response substantially larger than that induced by an ACE inhibitor. This possibility is reviewed below for the kidney. Other researchers have argued for a wide range of products of Ang I metabolism, each with a distinctive pharmacology.[13] This fascinating area remains controversial, with little evidence as yet for an important role in humans. Another potentially important model involves prorenin. Indeed, as the evidence for prorenin's important role in pathology accumulates, and because of its implications for renin blockade, the prorenin story merits special review.

Prorenin

By the mid-1990s it had become clear that prorenin was powerfully associated with the genesis of microvascular disease—nephropathy and retinopathy—in patients with diabetes mellitus.[14] Prorenin is present in the human circulation in very high concentration, about 10 times greater than the concentration of renin. As prorenin seemed to have no action on blood pressure, blood vessels, or aldosterone release, it was thought to be metabolic waste, and so was considered to be a "marker" of disease rather than being involved mechanistically. On the other hand, the concordance between plasma prorenin concentration and microvascular disease was so powerful that it was difficult to avoid thinking about a mechanistic relationship. But the responsible pathway had not yet been identified.

That situation changed dramatically in 2002 with the identification of a receptor, isolated from cultured human mesangial cells, that avidly binds renin.[15] This receptor not only binds renin, but binds prorenin equally well. Surprisingly, binding to the receptor increases the catalytic activity of renin 5-fold and provides prorenin with complete catalytic activity. Perhaps even more surprising is the accumulating evidence that activation of this receptor, now called the prorenin receptor, is capable not only of generating Ang I and Ang II, but also of activating potentially important intracellular pathways without an intervening involvement of Ang I or Ang II.[15-17] Given that these pathways lead to the release of agents important in the development of tissue fibrosis, an obvious link to disease pathogenesis now exists.[16,17]

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Recently, Huang and colleagues[17] confirmed and extended this line of investigation by testing the hypothesis

that renin, independent of its enzymatic action to enhance angiotensin synthesis, would lead to the release of important mediators of fibrosis. In vitro, renin in relatively low concentrations induced unambiguous increases in transforming growth factor (TGF)-β_1 that were both dose and time dependent. The responses were not altered by adding the Ang II receptor antagonist losartan or the ACE inhibitor enalapril in high concentration, nor were the responses influenced by a direct renin inhibitor. Renin also led to an increase in plasminogen activator inhibitor 1 and collagen-1 messenger ribonucleic acid (RNA) in vitro, a response partially blocked by neutralizing antibodies to TGF-β. Tissue Ang I and Ang II levels were extremely low. Perhaps most important, these researchers employed RNA interference to decrease expression of the renin receptor and demonstrated blockade of the induction of TGF-β in vitro. Although studies performed in vitro are a long way from the situation of patients in the clinic, it is difficult to ignore this emerging story. It is also difficult to ignore Huang and colleagues' overall conclusion: "Thus, renin may contribute to renal fibrotic disease, particularly when therapeutic Ang II blockade elevates plasma renin."

Treatment of patients with ACE inhibitors and ARBs leads to a sharp increase in renin release and plasma renin concentration, probably via the short feedback loop. The only blocker of the system that can render it quiescent is renin inhibition. Whether this will also render quiescent any actions via the prorenin receptor is the important next question.

Responses at the Tissue Level

Multiple observations in a range of tissues and models have provided compelling evidence that when agents that block the RAS are effective in changing the natural history of disease, their action occurs primarily at the tissue level.[1] With the identification of important species differences in the pathways for Ang II generation,[18] human studies became crucial. The logic employed for studying the human kidney was straightforward. If all of the Ang II acting on the renal circulation was formed through the classical pathway—with Ang I conversion to Ang II occurring only in the transit of blood through the lung—one would expect ACE inhibition, renin inhibition, and Ang II antagonists to induce an identical renal response, measured as an increase in renal plasma flow.[18]

Our research group chose renin inhibition as the initial pathway for exploring the control mechanism, for several reasons. Given the remarkable substrate specificity of the renin reaction, mechanistic specificity of the renin inhibitor was very likely. Moreover, as both ACE and renin inhibition produce a fall in plasma Ang II concentration, this would facilitate comparison of the degree of blockade achieved. Our anticipated result was that the renal hemodynamic response to ACE inhibition in healthy volunteers on a low-salt diet would reflect not only a fall in local Ang II formation but also a reduced kinin degradation. The result would be accumulation of vasodilator products, including bradykinin, kinin-dependent prostaglandin formation, and activation of endothelial nitric oxide release. To our surprise, the renal vasodilator response to the renin inhibitor available at that time, enalkiren, exceeded the response to the ACE inhibitor captopril.[18]

To follow up on this unexpected finding, we performed a more elaborate, double-blind study in which volunteers were studied 3 times: on one day they received placebo, on another day enalkiren, and another day captopril—in random order. There was no effect of placebo in this study. Captopril increased renal plasma flow by 90 to 100 mL/min/1.73 m^2, essentially identical to the earlier study (Figure 2). The renin inhibitor produced a 50% larger

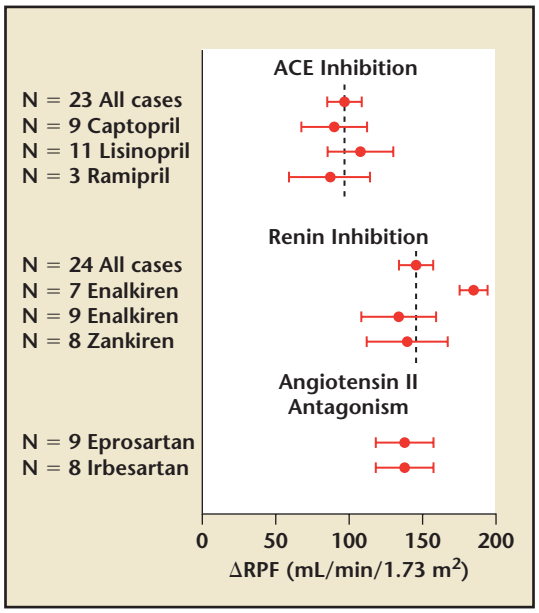

Figure 2. *Meta-analysis of renovascular response to pharmacological interruption of the renin system in healthy young men who were in balance on a 10 mEq sodium intake. Each agent was studied at the top of its dose–renal vascular relationship. From the ratio of flow increase induced by angiotensin converting enzyme (ACE) inhibition, approximately two-thirds of angiotensin II formation under these conditions is ACE dependent, and one-third is generated by alternative, non-ACE pathways. There is no evidence for non–renin-dependent generation of angiotensin. RPF, renal plasma flow. Reprinted with permission from Fisher and Hollenberg.*[3]

response, around 140 to 150 mL/min/1.73 m^2. Although renin is a fastidious enzyme with great substrate specificity, a possible interpretation of these findings was that renin inhibitors act via a mechanism unrelated to renin. Against this possibility is our finding that a high-salt diet blunted the renal response to renin inhibition. In that context, development of the Ang II antagonist class created the possibility of a "tie breaker." If the renin inhibitor acted via an alternative, non–angiotensin-dependent mechanism, one would expect Ang II antagonists to provide a lesser renovascular response under the conditions of our study. Conversely, if the renin inhibitor acted only through blockade of renin-dependent Ang II formation, one would expect an identical response.

We went on to study angiotensin II antagonists. At the top of the dose–response relationship, the Ang II antagonist induced a response similar to or slightly less than the response to the renin inhibitor. In this study, we probably underestimated the response to renin inhibition, as we reported findings only during the first several hours of administration.[18] In a follow-up study, we learned that a response to the renin inhibitor showed a continued increase over several hours, and thus we probably underestimated the peak.[19]

Combination Therapy

Following the introduction of ARBs, there has been an outpouring of reports on the influence on the kidney of ACE-ARB combinations. A recent review described 10 studies on patients with diabetic nephropathy and an additional 8 studies on patients with nephropathy unrelated to diabetes.[20] This output of papers reflected a number of factors. First, although ACE inhibitors are unambiguously effective, in individual patients their effect was often less than satisfactory. Second, there is a major ethical problem in dealing with 2 closely related drug classes. In view of the unambiguous efficacy of ACE inhibition, how could we justify withholding ACE inhibitors as part of a therapeutic trial?

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All of the studies reported were deeply flawed, because insufficient attention was given to the issue of drug dose. In a proper study, at least 1 of the 2 agents used in combination should be administered in a dose documented to achieve a maximal response. Otherwise the studies are not interpretable.

Is there a rationale for combination treatment if both drugs involve the same final pathway—blockade of angiotensin II production or its action? The answer is a somewhat tentative yes. The sigmoid shape of dose–response relationships is the integral of a normal distribution.[21] One feature of that distribution is a predictable relation between dose and magnitude of response: the response will increase linearly over the range 16% of maximum to 84% of maximum; the inflection point in the dose–response relationship occurs at these 2 points. To move from 84% to near 100% of maximum response requires a very large increase in dose. Thus, one could argue, parsimony dictates that the 2 agents be administered at the dose required to achieve a response at 84% of maximum. This will reduce the drug load and presumably the cost. Unfortunately, for none of the drugs involved do we have any idea of the maximum response and the dose to reach 84% of maximum.

In this area, the combination of a renin inhibitor with either an ACE or an ARB is more attractive than ACE-ARB combinations because of the short feedback loop and the reactive renin response to ACE inhibition and ARBs cited earlier. There is a reasonable chance that the renin response to blockade contributes to disease pathogenesis, especially in the case of the kidney.[17] Only renin inhibition renders the system quiescent.

Tolerability

Another important issue involves tolerability. Indeed, it has been an important theme in the evolution of renin system blockade. Within a few years, studies on quality-of-life measures made it clear that the ACE inhibitors were much better tolerated than the agents available up to that time: β-adrenergic blockers, methyldopa, and diuretics. The earlier agents were responsible for a great deal of fatigue, substantial depression, and a striking frequency of sexual difficulties. Not that the ACE inhibitors were free of adverse effects. Cough was frequent and very annoying. A rash was almost as frequent. Perhaps most important, the sporadic appearance of angioneurotic edema was potentially devastating.

When the ARBs became available, it was their tolerability that made them such an attractive advance. Their use in patients was free of cough, free of rash, and free of angioneurotic edema. Given that these agents are imidazole derivatives, and given the rather dramatic pharmacology of this chemical class, one would have expected a substantial frequency of adverse effects. In fact,

there have been none. This factor alone may well account for the remarkable growth in the use of ARBs, which antedated substantially the appearance of data on natural history.

Given the remarkable specificity of renin, which has a single substrate and a single product, one would expect the adverse effects associated with renin inhibition to be minimal. Lack of adverse events with renin inhibition depends not only on specificity of renin for its substrate angiotensinogen but also on the specificity of the renin inhibitor for renin. Aliskiren's excellent tolerability profile is related to its specificity for renin. This may not be true for other (future) renin inhibitors.

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Unanswered Questions

We already know, from a series of clinical trials reviewed elsewhere in this supplement, that aliskiren, the first orally effective renin inhibitor, is well tolerated. We also know from studies in patients with uncomplicated mild-to-moderate essential hypertension that the blood pressure response to aliskiren does not differ from the response induced by ARBs and ACE inhibitors. We do not yet know the response to this agent in patients who are likely to be especially responsive, including subclasses of patients with hypertension that are difficult to treat or are associated with diabetes mellitus, obesity, or advanced atherosclerosis.

The early evolution of the role of captopril provides an excellent example. The first human exposure to captopril occurred in late 1996. Full-scale clinical investigation began in 1977, and the drug was approved for the management of difficult hypertension in 1981, much the shortest time to approval for any antihypertensive agent. Why? Captopril was found to be remarkably effective for patients with difficult hypertension who had responded poorly to "standard triple therapy"—the combination of a diuretic, a β-blocker, and a vasodilator. With clear evidence that captopril met an unmet clinical need, it was easy for the U.S. Food and Drug Administration to provide an accelerated review and early acceptance.

Tissue protection is crucial. It is easy to forget that we treat high blood pressure not to reduce the blood pressure number but to change the natural history of disease. There are compelling reasons, reviewed above, to hope that renin inhibition—providing a degree of blockade of the system not easily achieved with alternative agents—will result in improved tissue protection. The obvious candidate is renal injury, but ultimately the logic extends to vascular injury, myocardial infarction, heart failure, and stroke.

A role for interference with the renin receptor, and in that way the contribution of prorenin, represents an equally attractive target.

The studies required to address these unanswered questions are already under way. What should we do in the interim, until the relevant data become available? Most of us are likely to do what we have done in the past. There are patients for whom the construct developed in this article is especially attractive, particularly if they are doing poorly on current therapy. We do not have the option of telling patients in need to come back in 3 years when we have the data required for a definitive decision. In our clinical practice we have to act now to deal with problems now, and very often have to make do with the best available information. The introduction of renin inhibition is no exception. For example, for the patient with type 1 diabetes mellitus and proteinuria who has an inadequate response to an ACE inhibitor, it is entirely reasonable to try renin inhibition—monitoring the influence on proteinuria. It is probably equally appropriate to add a renin inhibitor to an ACE inhibitor. On the basis of identical logic, captopril was widely used for patients with proteinuria well before appearance of the compelling evidence required for regulatory approval.[22]

Fortunately, on the basis of the experience of success with ACE inhibition and AT_1 receptor blockade, we will have the information required on the influence of the renin inhibitor much earlier in its development. ■

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Main Points

- Based on concentrations of substrates and products in the renin–angiotensin system (RAS) the renin-catalyzed step is the prime target for blockade of the system.
- A potentially important model of RAS blockade involves prorenin, which is powerfully associated with the genesis of microvascular disease in patients with diabetes mellitus.
- Studies of renin inhibition in the human kidney found the renal vasodilator response to renin inhibitor (enalkiren) exceeded the response to angiotensin-converting enzyme (ACE) inhibitor (captopril).
- A more elaborate, double-blind study showed that renin inhibitor produced a 50% larger increase in renal plasma flow than captopril.
- The combination of a renin inhibitor with either an ACE or an angiotensin receptor blocker (ARB) is more attractive than ACE-ARB combinations because of the short feedback loop and the reactive renin response to ACE inhibition and ARBs.
- When ARBs became available, it was their increased tolerability over ACE inhibitors that made them such an attractive advance; renin inhibitors seem to be similar to ARBs in this regard.