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REVIEW ARTICLE

INHIBITORS OF RENIN AS POTENTIAL THERAPEUTIC AGENTS

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INTRODUCTION

The importance of the renin-angiotensin system (RAS) in blood pressure (BP) regulation and sodium homeostasis has been revealed by extensive experimental and clinical studies with inhibitors of converting-enzyme[1,2]. The successful therapeutic application of converting enzyme inhibitors in the treatment of essential hypertension and congestive heart failure[2–5] has stimulated interest in compounds that interfere with the RAS at other points in the formation or action of angiotensin II (ANG II) (Figure 1)[6–8]. Recently, interest has focused on inhibitors of renin (EC 3.4.99.19) because of the unique substrate specificity of this enzyme. Theoretically, inhibitors of renin should block the RAS only, in contrast to inhibitors of converting enzyme which have the potential to interfere with other peptidic pathways.

In this review the biochemistry of human renin is briefly summarised, the structures and the *in vitro* and *in vivo* activity of various renin inhibitors are described and the therapeutic potential of renin inhibitors is discussed.

THE RENIN-ANGIOTENSIN SYSTEM

Renin is synthesized and stored in the epithelioid cells of the juxtaglomerular apparatus in the kidney. It is released into the blood by various stimuli such as hypovolemia, hypotension and sodium depletion. Release is mediated via the sympathetic nervous system and intrarenal vascular and tubular mechanisms[9,10]. ANG II exerts a negative feed-back inhibition of renin release (Figure 1). Although the kidney is the major source of plasma renin, other tissues such as the brain, heart, adrenal medulla and blood vessels have been shown to contain renin or to be capable of renin synthesis[11–14]. However, the physiological role of tissue renin is not known. A major portion of renin in the plasma is in an inactive form that can be activated by various

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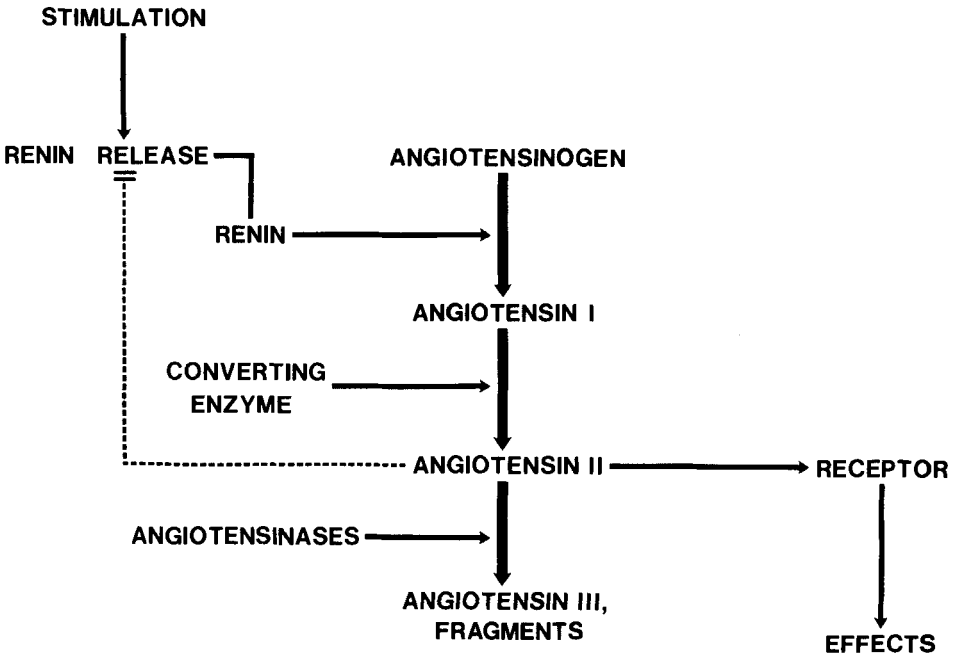

FIGURE 1 The reaction sequence of the renin-angiotensin system.

procedures *in vitro*[15,16]. Although inactive renin has been shown to be identical to prorenin[17,18], its physiological significance and the mechanism for its activation *in vivo* are not clear.

The reaction sequence of the RAS consists of several steps which are outlined in Figure 1. Renin is an endopeptidase that catalyses the first and rate-limiting step in the formation of ANG II. It specifically hydrolyses the amide bond between the amino acids in positions 10 and 11 of the N-terminal of angiotensinogen to produce the decapeptide angiotensin I (ANG I)[6,7,12]. Angiotensinogen is an alpha-2-globulin that is synthesized mainly in the liver and released into the circulation[19]. ANG I is transformed into the biologically active octapeptide ANG II by converting enzyme (Kininase II), a zinc-containing dicarboxypeptidase found in many tissues[1,2,6,7]. Unlike renin, converting enzyme has more than one substrate; it is involved in the degradation of various peptides. ANG II is degraded by exo- and endopeptidases into small inactive fragments. Only one of the degradation products, des-Asp-ANG II (ANG III), shows biological activity but its physiological role is a matter of debate[2,14].

ANG II may increase BP by its direct vasoconstrictor action, by a potentiation of the activity of the sympathetic nervous system and by its sodium conserving properties[1,2,12,20]. Sodium conservation may be a consequence of renal vasoconstriction, stimulation of aldosterone release or an effect of ANG II on tubular sodium reabsorption. These vascular and renal effects of ANG II account for the important role the RAS plays in the short- and long-term regulation of BP.

The systemic and renal vasoconstrictor effects of ANG II become apparent after stimulation of the RAS by sodium and volume loss or sodium restriction. Under these conditions, blockade of the formation of ANG II by converting-enzyme inhibitors results in a substantial reduction in blood pressure and an increase in renal blood

flow[1,2,5,21,22]. Moreover, a small but significant hypotensive effect has been shown in healthy volunteers on a normal salt diet[21,22]. This indicates that the RAS participates in the maintenance of blood pressure even in the salt-replete state.

The RAS plays a causal role in several forms of experimental and clinical renal hypertension (e.g. unilateral renal artery stenosis) and blood pressure can be normalized by blockade of the RAS with ANG II-antagonists or converting-enzyme inhibitors[2,4,5,21,22]. In addition, patients with essential hypertension who have a normal plasma renin activity also respond to treatment with converting-enzyme inhibitors. Thus, converting-enzyme inhibitors have proved to be effective antihypertensive agents in a high proportion of patients with hypertension. The effectiveness of converting-enzyme inhibitors in congestive heart failure appears to be due to a reduction in afterload via a decreased systemic vascular resistance and to an improvement in the renal capacity for sodium excretion[2,3,5].

BIOCHEMICAL PROPERTIES OF RENIN AND ANGIOTENSINOGEN

Renin belongs to the aspartic proteinase family which includes the enzymes pepsin, cathepsin D, gastricsin, chymosin, penicillopepsin and endothiapepsin. These enzymes have two aspartic acid residues in their active site that are essential for their catalytic activity. They show considerable similarity as evidenced by sequence homology[23,24] and X-ray crystal data[25–27]. Renin, unlike other aspartic proteinases, has no general protease activity and is inactive at acidic pH[28,29].

Renins from several different species have been isolated and purified (mouse[30], rat[31], dog[32], hog[33], man[34,35]). Since renin from the mouse submaxillary gland was the first renin to be completely purified and is the only renin that has been crystallized so far, it has been the most extensively investigated. The amino acid sequence is known for the renins from the mouse submaxillary gland[36,37] and the human kidney only[38,39].

Human renin has 340 amino acids. It has two glycosylation sites and is formed from a preprorenin with pre- and pro-segments of 20 and 46 amino acids respectively[40]. Human renin has not been crystallised. However, other aspartic proteinases have been and their X-ray crystal structures analysed (mouse submaxillary gland renin[41], pepsin[42], endothiapepsin[25], penicillopepsin[26], rhizopuspepsin[27]). Three dimensional models of human renin have been constructed based on the X-ray crystal structure of some of these other enzymes (Figure 2)[23,24,43]. These models reveal that the beta-carbonyl groups of the aspartates (Asp-38 and Asp-226) are coplanar, symmetrically arranged and held in a network of hydrogen bonds. The adjacent S and S′ subsites are well defined hydrophobic regions with walls composed of mixed beta-sheets. The entire active site appears as a long, deep cleft and can accommodate 7 amino acid units (S_4–S_3'). The enzyme also contains a mobile flap (Leu-79–Gly-90), which, when closed, lies across the cleft from the S_2 to the S_2' subsite and probably holds the substrate in the active site[44]. Renin has a bilobal structure with an approximate two-fold symmetry axis running along the cleft. This is consistent with the hypothesis that aspartic proteinases, including renin, have arisen from a common ancestral gene by duplication and subsequent divergence and fusion[39,45].

The catalytic mechanism of aspartic proteinases has been studied with pepsin, penicillopepsin and mouse submaxillary gland renin[46–50]. Results from these studies are consistent with a general acid-base hydrolysis mechanism, whereby water stereospecifically attacks the carbonyl of the scissile amide bond and the aspartates act as

FIGURE 2 Stereo view of a computer-generated model of human renin looking down the active site cleft. The mobile flap can be seen at the top center of the model, overhanging the cleft. The model can be viewed in 3-dimensions using stereoscopic glasses.

anchors and mediate the appropriate proton transfers. However, a mechanism involving an ammonium ion instead of water or covalent catalysis with an acyl enzyme intermediate cannot be excluded[51].

Renins from different species vary in their specificity for angiotensinogen from other species. Human renin is less selective than other mammalian renins; it cleaves angiotensinogen from other species whereas sub-primate renins do not cleave human angiotensinogen[19]. However, in all species angiotensinogen is the only known physiological substrate for renin. With human angiotensinogen as a substrate, human renin has a pH optimum of 5.5–6.0 and a K_m in the μM range[34,35].

Angiotensinogen is a glycoprotein with a molecular weight of approximately 50 000 to 60 000[19]. The first 10 amino acids at the N-terminal represent ANG I and are the same in most species. The amino-acid sequences on the C-terminal side of the cleavage site for renin (from position 11) differ in various species. Consequently, the peptide bond that is cleaved by renin is a Leu-Val bond in man[52] and a Leu-Leu bond in some other species (rat, horse)[19] (Table I). These differences in the amino acid sequence adjacent to the cleavage site may contribute to the species-specificity of the renin-angiotensinogen reaction. Although angiotensinogen is mainly synthesized in the liver, other tissues such as the brain have the capacity to synthesize it[13,53]. Since converting enzyme and renin have also been found at these sites, all of the components necessary for the local formation of ANG II are present in these tissues[11,13].

RENIN INHIBITORS

Structure and Activity in vitro

The first substances used to inhibit renin were natural products. Initially, antisera produced against crude renin extracts were used[54,55]. More recently, polyclonal and monoclonal antibodies against purified renin from the mouse[56], dog[57] and man[57–60] have been prepared. R-3-36-16, a mouse monoclonal antibody against human renin, is the most potent inhibitor of human renin described so far ($IC_{50} = 10^{-11}$ M)[60].

Another of the early renin inhibitors, pepstatin, also originated from a natural source[61]. Pepstatin is a microbial peptide that is a potent competitive inhibitor of most aspartic proteinases ($K_i = 0.05$ nM for pepsin) but a rather weak inhibitor of renin ($K_i = 10\,\mu$M for human renin) (Table I)[62]. The solubility and potency of pepstatin were improved by acetylation or addition of hydrophilic amino acids to the N-terminal[63].

Synthetic peptidic inhibitors of renin have been produced by two different approaches. One approach was based on the hypothesis that the prosegment of prorenin interacts with the active site of renin and inhibits enzymatic activity by preventing the access of angiotensinogen. Peptide analogues of fragments of the prosegment sequence were synthesized but they proved to be only weak inhibitors of renin (Table I)[64].

The other approach was to synthesize peptidic analogues of the N-terminal amino acid sequence of angiotensinogen. The first inhibitors were based on the sequence of horse angiotensinogen (Table I). The shortest peptide sequence that is cleaved by renin is the octapeptide extending from position 6 to 13. This peptide is also a weak competitive inhibitor of renin[65]. Shorter peptides are not cleaved and are also weak inhibitors[66]. Substitution of the amino acids at the scissile bond (positions 10 and 11)

TABLE I
Chemical structure and *in vitro* activity of renin inhibitors

	cleavage site ↓ 1 2 3 4 5 6 7 8 9 10 11 12 13 14	IC_{50} against renin (M) Human	Dog
RENIN SUBSTRATES			
Human angiotensinogen[52]	Asp-Arg-Val-Tyr-Ile-His-Pro-Phe-His-Leu——Val-Ile-His-Asn-R		
Horse angiotensinogen[19]	Asp-Arg-Val-Tyr-Ile-His-Pro-Phe-His-Leu——Leu-Val-Tyr-Ser-R		
NATURAL INHIBITOR			
Pepstatin[61,62]	Iva-Val-Val — Sta — Ala-Sta	1×10^{-5}	1×10^{-6}
SUBSTRATE ANALOGUE			
RIP[68]	Pro-His-Pro-Phe-His-Phe——Phe-Val-Tyr-Lys	[a]2×10^{-6}	
TRANSITION-STATE ANALOGUES			
H-77[69]	Pro-His-Pro-Phe-His-Leu—^{R}—Leu-Val-Tyr	1×10^{-6}	2×10^{-8}
H-142[69]	Pro-His-Pro-Phe-His-Leu—^{R}—Val-Ile-His	1×10^{-8}	1×10^{-5}
H-261[71]	Boc-His-Pro-Phe-His-Leu—^{OH}—Val-Ile-His	7×10^{-10}	2×10^{-8}
SR 42 128[76]	Iva-Phe-Nle — Sta — Ala-Sta	3×10^{-8}	5×10^{-8}
CGP 29 287[75]	Z-Arg-Arg-Pro-Phe-His — Sta — Ile-His-Lys(Boc)OMe	1×10^{-9}	2×10^{-8}
SCRIP[77]	Iva-His-Pro-Phe-His — Sta — Leu-Phe-NH_2	2×10^{-8}	1×10^{-8}
Compound 3[78]	Iva-His-Pro-Phe-His — Chsta — Leu-Phe-NH_2	2×10^{-10}	2×10^{-9}
Compound 6[79]	Boc-Phe-His-Leu—^{AA}—Leu-Ile-His-OMe	2×10^{-7}	
A 60956[80]	Boc-Phe-His-Cha—^{OH}—Sleu-R_1	7×10^{-9}	
Compound 4e[81]	Boc-Phe-His — Stoff — Ile-$NHCH_2R_2$	5×10^{-10}	

[a]: K_i instead of IC_{50}; Sta: statine; Nle: norleucine; R: $-CH_2NH_2-$; OH: $-CHOHCH_2-$; Chsta: (3*S*, 4*S*)-4-amino-5-cyclohexyl-3-hydroxypentanoic acid; AA: $CH(OH)CH_2NH$; Cha: 3-cyclohexylalanine; Sleu-R_1: $CH_2SCH_2CH(i\text{-Bu})NHCOCH_2CH_2C_6H_5$; Stoff: 2,2-difluorostatone; R_2: 2-pyridyl; Z, Boc, OMe, Iva: protection groups

of octapeptide analogues prevented cleavage and resulted in competitive inhibitors of greater potency[67]. The best compound from these early studies was a decapeptide, renin inhibitory peptide (RIP), which has an inhibitory potency against human renin in the μM range (Table I)[68].

As a subsequent approach, the peptides at the scissile bond were replaced by non-cleavable dipeptide analogues. These contained either a reduced peptide bond (CH_2NH)[69,70] or an hydroxyethylene ($CHOHCH_2$) group[71,72]. These structural elements are thought to act as "transition state" mimics since they resemble the tetrahedral conformation of the natural substrate at the scissile bond after it has bound to renin and is activated for cleavage[73]. The concept has resulted in inhibitors with a potency in the nM range such as H-142[69,70] and H-261[71] (human renin) and H-77[69,70] (canine renin) (Table I).

Statine, the unusual amino acid found in pepstatin, is also thought to act as a transition state mimic. Therefore, statine has been incorporated into peptide analogues to replace the amino acids at positions 10 and 11[74-76]. This resulted in inhibitors with potencies in the nM range such as statine-containing renin inhibitory peptide (SCRIP)[77], CGP 29 287[75] (Figure 3) and SR 42 128[76] (Table I). More recently, replacement of the isobutyl group in statine with a cyclohexylmethyl group was shown to enhance the potency of these inhibitors[78].

Current research in the transition state mimic inhibitors is concentrated on finding alternative dipeptide mimics and on reducing the molecular size and the number of peptide bonds in these compounds. Several new dipeptide mimics have been recently described[79-81]. Many of the inhibitors containing these new mimics have only two or three natural amino acids and inhibit human renin in the nM range (Table I). As the molecular size and the peptidic nature of renin inhibitors decrease, their bioavailability after oral administration should increase.

The species and enzyme specificity of renin inhibitors varies (Figure 3). For the transition state analogue inhibitors, specificity appears to depend on the amino acid sequence and on the type of mimic[69,72]. Since most of these inhibitors were designed to inhibit human renin, they usually inhibit primate renin but are much less potent against renin from subprimate species. Consequently, primate models have been developed for the *in vivo* evaluation of such specific inhibitors[58,68,82].

Activity in vivo

Antisera against renin were the first experimental probes used to inhibit the biological actions of this enzyme *in vivo*[54,55]. Active and passive immunization against renin blocked the pressor reponse to exogenous renin and lowered BP in various models of renin-dependent hypertension. More recently these early findings obtained with antisera raised against impure renin preparations have been confirmed using polyclonal and monoclonal antibodies produced against pure renin[56-58,60]. The mouse monoclonal antibody R-3-36-16 is a potent inhibitor of human renin *in vivo* as well as *in vitro*. In mildly sodium-depleted marmosets, a dose of 10 μg/kg i.v. induced an inhibition of plasma renin activity of over 90% and lowered blood pressure[60]. These effects persisted for at least 2 h after injection (Figure 4a).

Pepstatin was the first of the peptidic inhibitors to be evaluated *in vivo*. It was reported to lower BP in rats under various conditions where the activity of the RAS was stimulated[62,63,83]. However, the low potency and lack of specificity of pepstatin limit the interpretation of these experiments[61,84]. The primate-specific inhibitors,

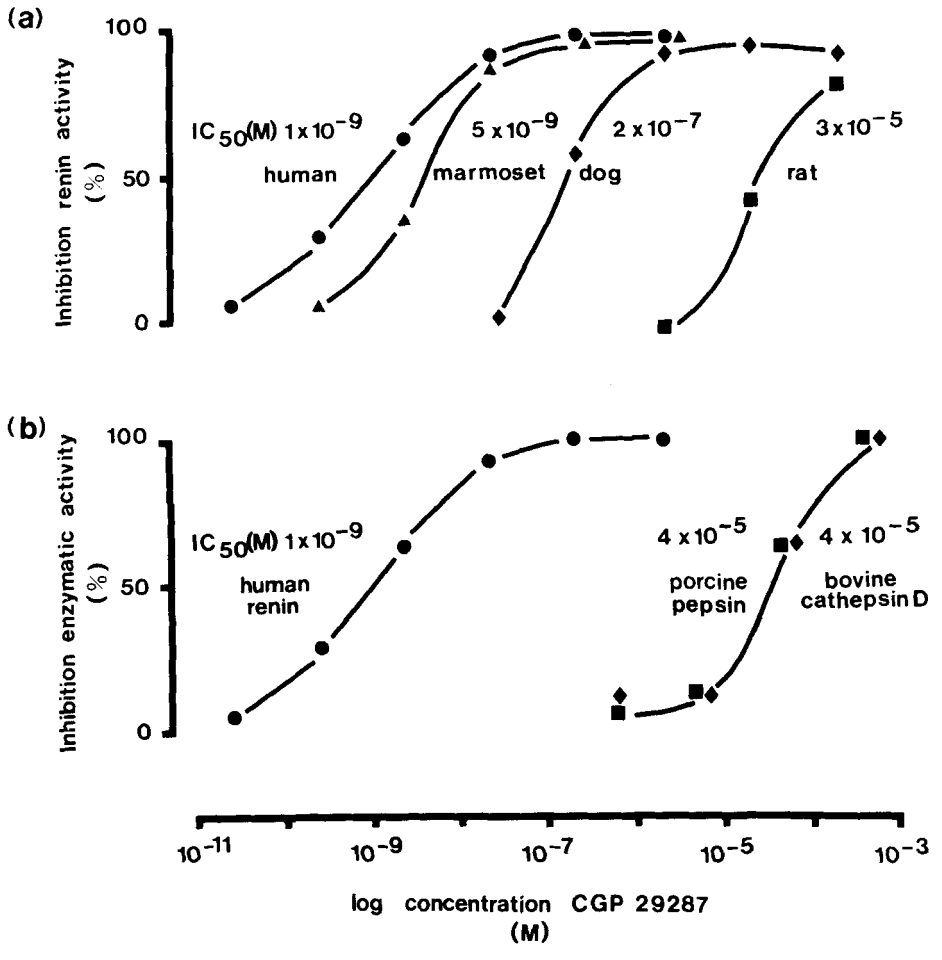

FIGURE 3 (a) Species specificity of the renin inhibitor CGP 29 287; inhibition of renin activity in human, marmoset, dog or rat plasma by CGP 29 287.(b) Enzyme specificity of the renin inhibitor CGP 29 287; inhibition of human renin, porcine pepsin or bovine cathepsin D activity by CGP 29 287. IC_{50} = inhibitory concentration, 50%. These results indicate that CGP 29 287 is a potent inhibitor of primate renin, is less active against renin from subprimate species, and only a weak inhibitor of other aspartic proteinases. (From Wood *et al.*[75] by permission of the American Heart Association Inc.).

RIP[68], H-142[82,85] and H-261[71], have been shown to induce a hypotensive response in sodium-depleted monkeys. Comparable results have been obtained in sodium-depleted dogs with H-77[86,87] and SCRIP[77]. In these studies BP remained lowered during infusion of H-77, H-142 or SCRIP over several hours. This indicates that an increase in renin release or an activation of other BP-regulating systems does not overcome the hypotensive effects induced by inhibition of renin. In addition, no rebound increase in BP was observed after stopping the infusions.

Because of their peptidic nature, RIP, H-77, H-142 and SCRIP have very short biological half-lives and must be administered by i.v. infusion. CGP 29 287 and

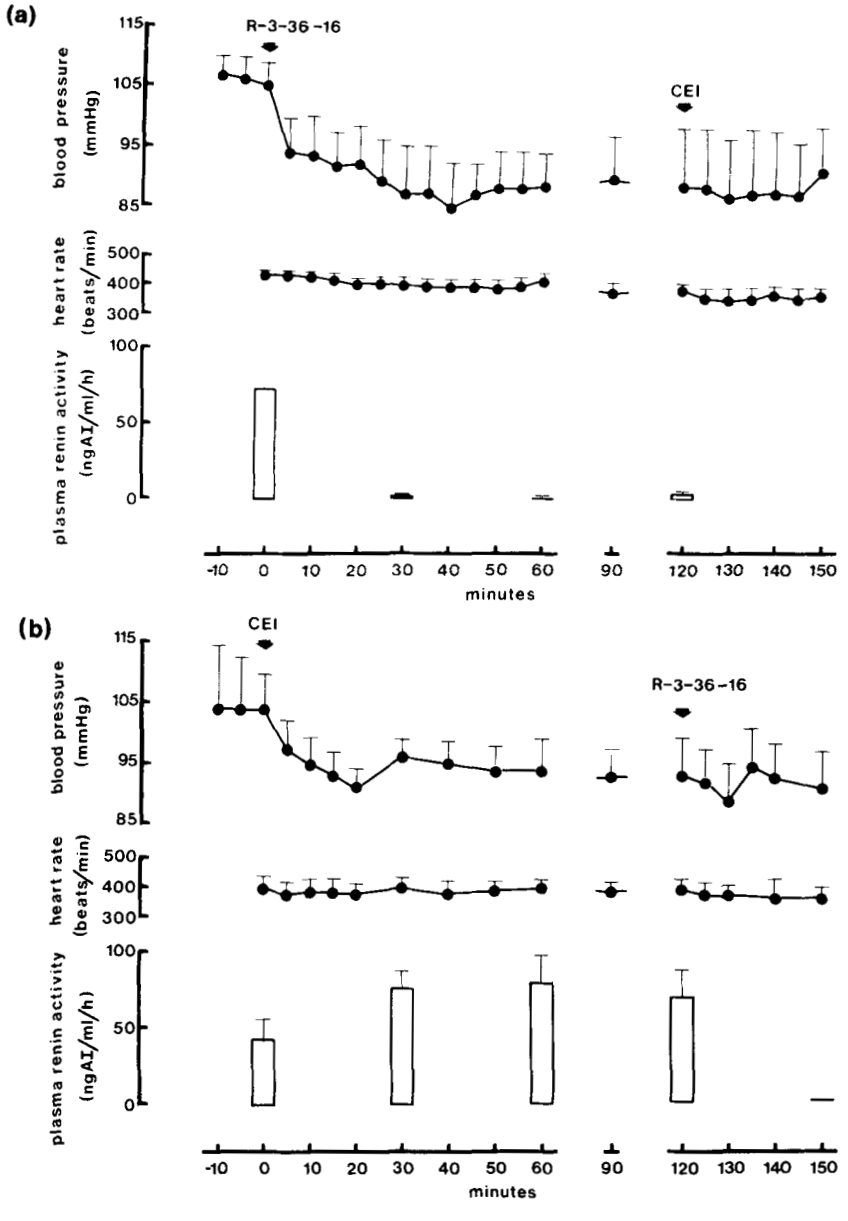

FIGURE 4 Blood pressure, heart rate and plasma renin activity in conscious marmosets pretreated with furosemide (5 mg/kg i.m.). (a) Effects of the converting-enzyme inhibitor (CEI), teprotide (2 mg/kg i.v.), injected 120 min after the monoclonal antibody against human kidney renin, R-3-36-16 (0.01 mg/kg i.v.). (b) Effects of R-3-36-16 (0.1 mg/kg i.v.) injected 120 min after the converting-enzyme inhibitor enalaprilat (2 mg/kg i.v.). The two types of agents induced a hypotensive response of similar magnitude. Prior administration of the renin inhibitor completely prevented the hypotensive response to the converting-enzyme inhibitor and vice versa. This study shows that the hypotensive response induced after renin or converting-enzyme inhibition appears to be entirely due to blockade of the renin-angiotensin system. Other mechanisms do not appear to contribute to the hypotensive response induced by the antibody or by the converting-enzyme inhibitors in this experimental situation. (From Wood *et al.*[60] by permission of the American Heart Association Inc.).

SR 42 128 were the first synthetic renin inhibitors that were effective after bolus i.v. injection. Doses of 1 mg/kg of CGP 29 287[75] and 3 mg/kg of SR 42 128[76,88] induced a complete inhibition of plasma renin activity and a reduction in BP which persisted for at least 1 h after bolus injection in sodium-depleted marmosets. The structural feature of CGP 28 287 that appears to be responsible for its increased half-life *in vivo* is the protection groups at both terminals. CGP 29 291, a renin inhibitor with the same structure as CGP 29 287 but without protection groups, has a much shorter duration of action[89]. CGP 29 287 was also effective after oral administration in a dose of

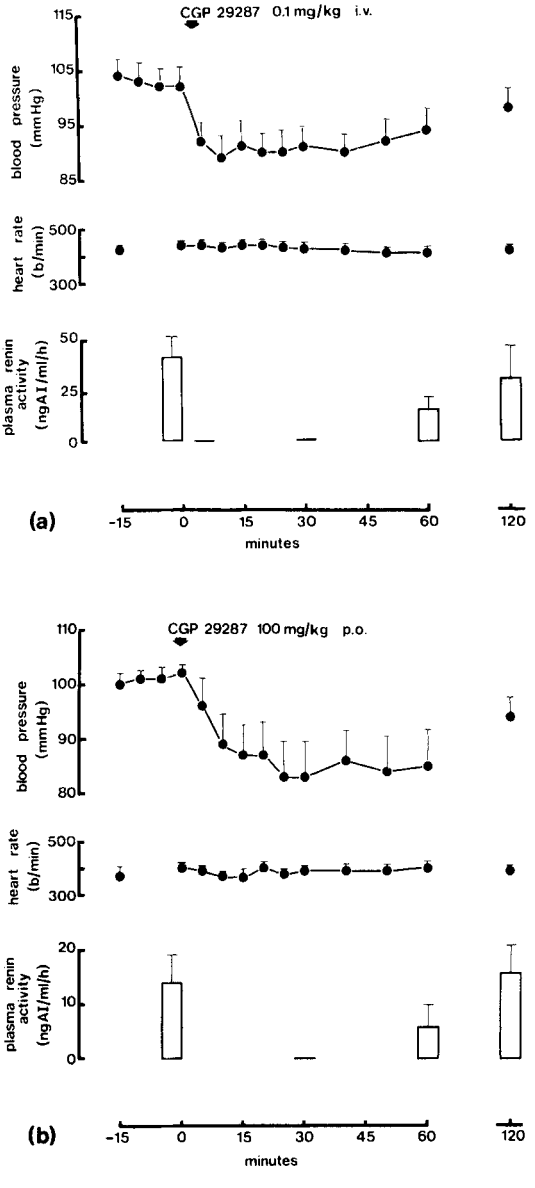

FIGURE 5 Effects of the renin inhibitor CGP 29 287 on blood pressure, heart rate and plasma renin activity in conscious marmosets, pretreatment with furosemide (5 mg/kg i.m.), after, (a) intravenous or, (b) oral administration. The large difference between the equieffective intravenous and oral doses indicates that the bioavailability of CGP 29 287 is very low. (From Wood *et al.*[75] by permission of the American Heart Association Inc.).

100 mg/kg[75] (Figure 5). However, the ratio between equieffective intravenous and oral doses suggests that the bioavailability of CGP 29 287 is very low (Figure 5).

The fall in BP induced by various renin inhibitors has been shown to be associated with a reduction in plasma renin activity[58,60,68,71,75,76,85,87] (Figures 4, 5 and 7) and a decrease in plasma concentrations of ANG I and ANG II[70,71,85,86]. A positive correlation between the hypotensive effects of the renin inhibitors and the pre-treatment plasma renin activity [58,87] or the decrease in plasma ANG II has been demonstrated[70,71] (Figure 6). These findings indicate that the fall in BP is a consequence of the inhibition of renin and the subsequent reduction in ANG II concentrations. In most of these studies the threshold doses for the inhibition of plasma renin activity and the decrease in BP were similar. Moreover, the time courses for the changes in plasma renin activity and in BP were similar (Figures 4, 5 and 7). However, in studies with SCRIP in sodium-depleted dogs the maximum hypotensive response occurred at doses that were substantially higher than those necessary to achieve full inhibition of plasma renin activity[77]. After stopping the infusion, BP recovered more rapidly than plasma renin activity. The reason for this dissociation is not clear. It may be due to a depressor effect of SCRIP unrelated to inhibition of renin. Alternatively, these results may indicate that inhibition of tissue renin might be a more important determinant of the hypotensive effect than blockade of plasma renin. Thus the distribution of renin

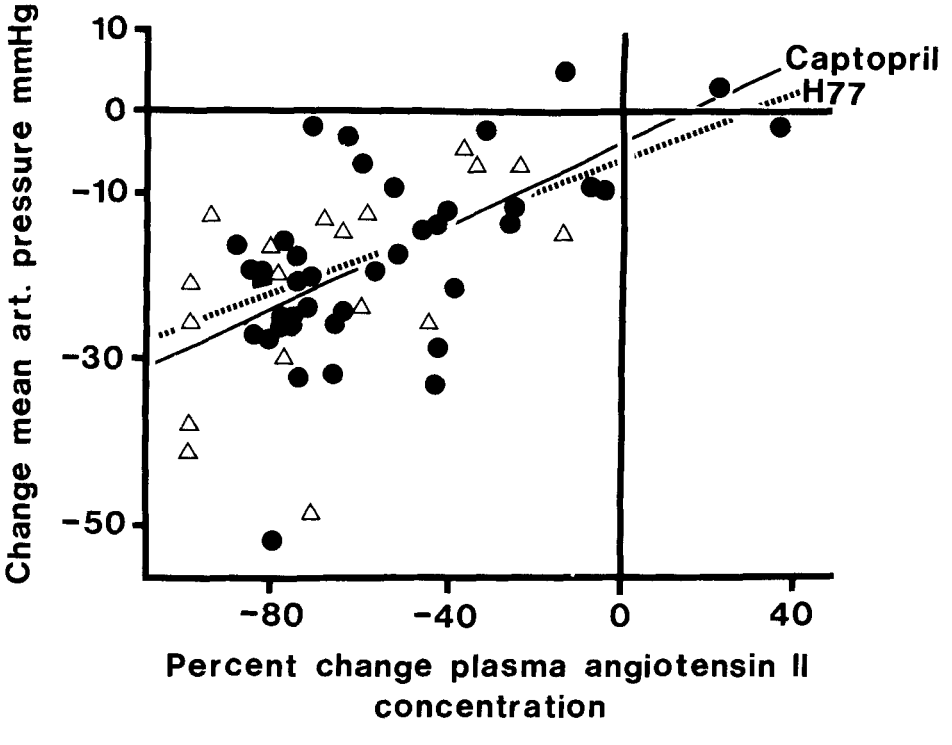

FIGURE 6 Relation between changes in the plasma concentration of angiotensin II and changes of mean arterial pressure in conscious sodium-depleted dogs given either the converting-enzyme inhibitor captopril (▲) or the renin inhibitor H-77 (●). This study shows that captopril and H-77 lower blood pressure to a similar extent for a given fall in the plasma concentration of angiotensin II. (Redrawn from Leckie *et al.*[70] by permission of Marcel Dekker Inc.).

inhibitors to sites of local ANG I formation might be a prerequisite for their hypotensive effectiveness.

In most of the animal studies with renin inhibitors no reflex tachycardia has been observed despite the fall in BP[58,60,76,84,88] (Figures 4a and 5). This is in contrast to the effects of many other depressor agents but is consistent with experimental and clinical findings with converting-enzyme inhibitors[1–5] (Figure 4b). Some tachycardia has been observed after high doses of RIP[68] and H-77[86,87]. This might be due to the rapid fall in BP induced by the higher doses or to an action of these particular compounds that is unrelated to the reduction in ANG II.

Comparative studies with renin and converting-enzyme inhibitors have been done to assess whether blockade of the RAS is the exclusive mechanism by which both types of agents act. In normotensive sodium-depleted animals, the maximum fall in BP induced by a renin inhibitor or a converting-enzyme inhibitor appears to be similar (Figures 4 and 7)[58,60,68,70,84]. After renin or converting-enzyme inhibition, the fall in BP has been shown to correlate with the pretreatment plasma renin activity[58,87] or the decrease in plasma ANG II concentrations[70,71]. The shape and intercept of these regression lines were identical after either treatment. Several studies have shown that the prior administration of a renin inhibitor entirely prevented the hypotensive response to a converting-enzyme inhibitor and *vice versa*[60,70,75,84] (Figure 4). Although a few studies have shown a small additional fall in BP when a converting-enzyme inhibitor was administered after a renin inhibitor[87,90], the majority of these studies do not support the proposition that converting-enzyme inhibitors lower blood pressure

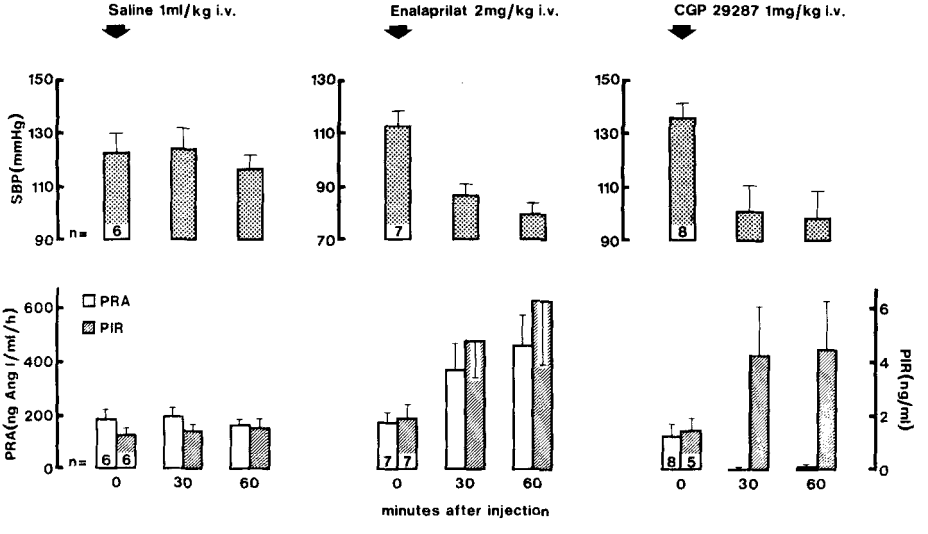

FIGURE 7 Systolic blood pressure (SBP), plasma renin activity (PRA, radioimmunoassay of ANG I after incubation of plasma), and plasma immunoreactive renin (PIR, direct immunosorbent assay) in conscious, severely sodium-depleted marmosets after i.v. injection of the converting-enzyme inhibitor enalaprilat or the long-acting renin inhibitor CGP 29 287. Both agents induced a comparable fall in SBP. PRA increased after enalaprilat but was almost completely inhibited after CGP 28 287. The number of renin molecules in the plasma, as indicated by the PIR values, increased to the same degree after both drugs. This indicates that blockade of the RAS by either converting enzyme or renin inhibition results in the same reactive stimulation of renin release. (From Hofbauer *et al.*[91] by permission of the American Heart Association Inc.).

after acute administration by mechanisms other than the blockade of the RAS. However, it cannot be excluded that after chronic administration or under other experimental conditions a difference between renin and converting-enzyme inhibition might be observed.

Converting-enzyme inhibitors and renin inhibitors induce opposite changes in plasma renin activity; it is increased after converting-enzyme inhibition and decreased after renin inhibition. Despite these opposite changes in plasma renin activity, the total number of renin molecules is increased by both treatments[77,91]. In a study with CGP 29 287 and the converting-enzyme inhibitor enalaprilat in marmosets, total plasma immunoreactive renin increased by a similar amount after acute treatment with either compound[91] (Figure 7). These findings indicate that renin release from the kidney responds in the same way to either renin or converting-enzyme inhibition and that the signal for release is the withdrawal of the negative feed-back inhibition by ANG II.

As well as having similar effects on BP and heart rate, inhibitors of renin and converting enzyme induce comparable changes in systemic and regional haemodynamics. CGP 29 287 and enalaprilat have been shown to increase renal blood flow and decrease renal vascular resistance to the same extent in normotensive marmosets after mild sodium-depletion[92] (Figure 8). The renin inhibitory monoclonal antibody R-3-36-16 induced similar changes. Enalaprilat and SCRIP have been shown to have similar systemic haemodynamic effects in dogs with acute left ventricular failure[93]. Both agents reduced total peripheral resistance and had no significant effect on cardiac output. Comparable antihypertensive effects of renin and converting-enzyme inhibitors have been demonstrated in monkeys[68,94] after acute renal artery con-

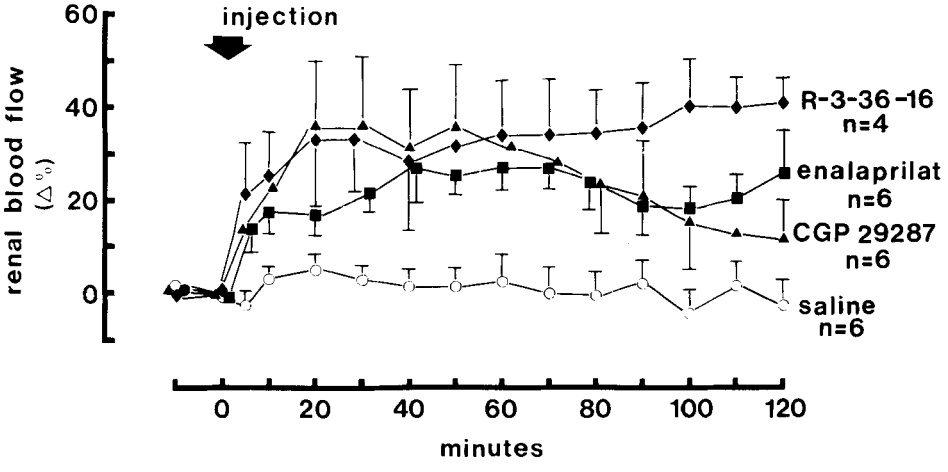

FIGURE 8 Effects of the converting-enzyme inhibitor enalaprilat (2 mg/kg i.v.), the renin inhibitor CGP 29 287 (1 mg/kg i.v.) or the renin monoclonal antibody R-3-36-16 (0.1 mg/kg i.v.) on renal blood flow in conscious marmosets pretreated with furosemide (5 mg/kg i.m.). At 30 min after injection the change in renal vascular resistance was of a similar magnitude for all of the three agents ($-28 \pm 3\%$, $-32 \pm 6\%$ and $-33 \pm 7\%$ respectively). These findings indicate that the renal vasodilatation induced after renin or converting enzyme inhibition is entirely due to a reduction in the renal vasoconstrictor action of endogenous angiotensin II.

striction. However, more studies are necessary to verify that renin inhibitors have an antihypertensive efficacy similar to converting-enzyme inhibitors.

THERAPEUTIC POTENTIAL OF RENIN INHIBITORS

The presently available data from animal studies indicate that renin inhibitors should have the same therapeutic potential as converting-enzyme inhibitors for the treatment of hypertension and congestive heart failure. Because of renin's unique substrate-specificity, renin inhibitors might have the clinical advantage of a lower incidence of side effects after chronic therapy. On the other hand, renin inhibitors might prove to be less effective than converting-enzyme inhibitors if mechanisms unrelated to the inhibition of the RAS are important for the antihypertensive efficacy of converting-enzyme inhibitors.

As yet, there have been few studies with renin inhibitors in man. Studies with the relatively weak inhibitor, RIP[95], and the more potent but short acting inhibitor, H-142[96], in normal volunteers or hypertensive patients confirmed the hypotensive action observed in experimental animals. However, in high doses, both compounds had some unexpected effects that are not seen after converting-enzyme inhibition. RIP induced precipitous decreases in BP and heart rate in salt-depleted normotensive subjects and in a patient with low renin essential hypertension. H-142 induced a significant increase in heart rate and plasma noradrenaline concentrations. These effects might be due to nonspecific effects of these particular compounds that are unrelated to renin inhibition. Studies with more potent and long acting inhibitors need to be done.

The mouse monoclonal antibodies against human renin have the greatest potency and the longest duration of action of any of the inhibitors of renin that have been produced so far. However, their potential to induce an allergic response in man probably precludes their clinical application. Theoretically, immunogenicity might be reduced by hybridization of the variable regions from mouse monoclonal antibodies with Fc-fragments of human origin[97] or by the production of monoclonal antibodies after *in vitro* immunization of human lymphocytes[98]. However, even if this were achieved, the clinical application of monoclonal antibodies would be limited by the necessity for parenteral application.

Several of the currently available synthetic renin inhibitors are potent and long acting after i.v. administration. However, as yet none of them are sufficiently active after oral administration. Until orally active renin inhibitors are available, it will not be possible to assess the therapeutic potential of renin inhibitors in long-term clinical studies.

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