Angiotensin-Converting Enzyme Inhibition and Angiotensin II Type 1 Receptor Blockade Prevent Cardiac Remodeling in Pigs After Myocardial Infarction : Role of Tissue Angiotensin II

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Role of Tissue Angiotensin II

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Background—The mechanisms behind the beneficial effects of renin-angiotensin system blockade after myocardial infarction (MI) are not fully elucidated but may include interference with tissue angiotensin II (Ang II).

Methods and Results—Forty-nine pigs underwent coronary artery ligation or sham operation and were studied up to 6 weeks. To determine coronary angiotensin I (Ang I) to Ang II conversion and to distinguish plasma-derived Ang II from locally synthesized Ang II, ^{125}I-labeled and endogenous Ang I and II were measured in plasma and in infarcted and noninfarcted left ventricle (LV) during ^{125}I-Ang I infusion. Ang II type 1 (AT_1) receptor–mediated uptake of circulating ^{125}I-Ang II was increased at 1 and 3 weeks in noninfarcted LV, and this uptake was the main cause of the transient elevation in Ang II levels in the noninfarcted LV at 1 week. Ang II levels and AT_1 receptor–mediated uptake of circulating Ang II were reduced in the infarct area at all time points. Coronary Ang I to Ang II conversion was unaffected by MI. Captopril and the AT_1 receptor antagonist eprosartan attenuated postinfarct remodeling, although both drugs increased cardiac Ang II production. Captopril blocked coronary conversion by >80% and normalized Ang II uptake in the noninfarcted LV. Eprosartan did not affect coronary conversion and blocked cardiac Ang II uptake by >90%.

Conclusions—Both circulating and locally generated Ang II contribute to remodeling after MI. The rise in tissue Ang II production during angiotensin-converting enzyme inhibition and AT_1 receptor blockade suggests that the antihypertrophic effects of these drugs result not only from diminished AT_1 receptor stimulation but also from increased stimulation of growth-inhibitory Ang II type 2 receptors. **(*Circulation*. 2000;102:1556-1563.)**

Key Words: angiotensin ■ inhibitors ■ receptors ■ myocardial infarction ■ hypertrophy

Angiotensin-converting enzyme (ACE) inhibitors prevent cardiac remodeling (ie, ventricular hypertrophy and chamber dilatation) after myocardial infarction (MI). This occurs, in part, independent of their blood pressure–lowering effect,[1] suggesting interference with tissue angiotensin II (Ang II). However, ACE inhibitors do not always lower cardiac Ang II[2–4] because of (1) their pharmacokinetic properties (short half-life and lack of tissue penetration),[3,4] (2) ACE upregulation during prolonged ACE inhibition,[5] or (3) alternative converting enzymes, such as chymase.[6] Furthermore, previous Ang II measurements in infarcted hearts[4,7,8] have not taken into account the fact that cardiac Ang II is partly derived from the circulation. Plasma Ang II accumulates in the heart through Ang II type 1 (AT_1) receptor–mediated endocytosis,[2,9] and changes in AT_1 receptor density after MI[10,11] may influence this process.

AT_1 receptor antagonists have opened new avenues in the investigation of the importance of cardiac Ang II after MI. In general, these drugs increase rather than decrease Ang II levels,[12] and this may lead to activation of unoccupied growth-inhibitory[13] Ang II type 2 (AT_2) receptors. Currently, the effect of AT_1 receptor antagonism on the Ang II content of the infarcted heart is unknown. Furthermore, it is still controversial whether AT_1 receptor antagonism prevents the development of cardiac hypertrophy and increases survival after MI.[14,15]

Therefore, the aim of the present study was to investigate changes in cardiac Ang II content and origin (local synthesis versus uptake from plasma) after MI in pigs, to compare the effects of ACE inhibition and AT_1 receptor antagonism on these changes, and to evaluate whether the effects of both renin-angiotensin system (RAS) blockers on cardiac angio-

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tensin content are related to their effects on postinfarct remodeling.

Methods

Animals

Experiments were performed in accordance with the *Guide for the Care and Use of Laboratory Animals* (NIH Publication 86-23, revised 1985). Forty-nine 2- to 3-month-old Yorkshire×Landrace pigs of either sex entered the study. Adaptation of animals to laboratory conditions started 1 week before surgery.

Surgical Procedure

Animals were sedated with ketamine (30 mg/kg IM), anesthetized with thiopental (10 mg/kg IV), intubated, and ventilated with a mixture of O_2 and N_2O, to which 0.2% to 1.0% (vol/vol) isoflurane was added.[16] Anesthesia was maintained with midazolam (2 mg/kg+1 mg/kg per hour IV) and fentanyl (10 μg/kg per hour IV). The chest was opened via a left intercostal space, and a fluid-filled polyvinylchloride catheter was inserted into the aortic arch for hemodynamic monitoring and blood sampling.[16] Subsequently, the pericardium was opened, the proximal left circumflex coronary artery (LCXCA) was dissected out, and a suture was placed around the LCXCA. In 35 animals, the LCXCA was permanently ligated (MI group), whereas in 14 animals, the suture was removed (sham group). The pericardium was closed, and the aortic catheter was tunneled subcutaneously to the back. The chest was closed, and the animals were allowed to recover. Animals received analgesia (0.3 mg buprenorphine IM) for 2 days and antibiotic prophylaxis (25 mg/kg amoxicillin and 5 mg/kg gentamycin IV) for 5 days.[16]

Experimental Groups

Animals were followed for 1, 3, or 6 weeks. Of the 3-week MI animals, 6 received captopril (25 mg PO BID), and 7 received the AT_1 receptor antagonist eprosartan (400 mg PO BID, a gift of Dr P.K. Weck, SmithKline Beecham, Collegeville, Pa). This dose of eprosartan blocks Ang II–induced pressor responses by >95% (n=3). Treatment started 12 to 24 hours after LCXCA ligation and was continued for 3 weeks.

Echocardiography

At the end of the follow-up period, animals were sedated with ketamine, and 2D echocardiographic recordings of the left ventricular (LV) short axis at midpapillary level were obtained (Sonos 5500, Hewlett-Packard) and stored for offline analysis. LV end-diastolic cross-sectional area (EDA) and end-systolic cross-sectional area (ESA) were determined, and ejection fraction (EF) was calculated as (EDA−ESA)/EDA×100%.

Infusion of ^{125}I-Ang I

After echocardiography, pigs were anesthetized and prepared for hemodynamic monitoring, administration of ^{125}I-angiotensin I (Ang I), and blood and tissue sampling.[9,17] After baseline measurements were collected, the animals were subjected to a 60-minute infusion of ^{125}I-Ang I (≈4×10^6 cpm/min) into the LV cavity. ^{125}I-Ang I and ^{125}I-Ang II reach steady-state levels in plasma and cardiac tissue within 10 and 60 minutes, respectively.[9]

Blood and Tissue Sampling

During follow-up, arterial blood samples were collected in the morning from awake animals for measurement of norepinephrine, epinephrine, atrial natriuretic peptide (ANP), N-terminal ANP, Ang I, and Ang II.[2,18,19] During ^{125}I-Ang I infusion, arterial and coronary venous blood samples were collected from anesthetized animals for measurement of endogenous and ^{125}I-labeled Ang I and II.[2,9] With the ^{125}I-Ang I infusion still running, the heart was stopped by fibrillation, the LV and right ventricle (RV) were separated and weighed, and 0.5- to 1-g samples were rapidly obtained from noninfarcted anterior LV wall, lateral LV wall (containing the infarct area and border zone after MI), interventricular septum, and RV wall. Samples were immediately frozen in liquid nitrogen and stored at −70°C.

Biochemical Measurements

Norepinephrine, epinephrine, ANP, and N-terminal ANP were measured as described before.[18,19] Endogenous and ^{125}I-labeled Ang I and II were measured, after SepPak extraction and high-performance liquid chromatography separation, by γ counting and radioimmunoassay, respectively.[2,9]

Data Analysis

Fractional conversion and degradation of ^{125}I-Ang I in the coronary vascular bed, ie, the percentage of arterially delivered ^{125}I-Ang I that is converted to ^{125}I-Ang II or degraded to other angiotensin metabolites during coronary passage, were calculated as described previously.[20] To quantify cardiac Ang I and II synthesis, tissue levels of Ang I and II were corrected for uptake from plasma by using the steady-state plasma and tissue levels of ^{125}I-Ang I and II.[2] Cardiac tissue ^{125}I-Ang I was undetectable in nearly all animals. To prevent underestimation of the contribution of plasma Ang I to tissue Ang I, in situ–synthesized tissue Ang I was quantified under the assumption that tissue ^{125}I-Ang I equals 5% of the steady-state plasma levels of ^{125}I-Ang I.[2] Differences between sham and MI animals and differences between treated and untreated animals were tested by 2-way ANOVA or multivariate ANOVA, followed by the Student *t* test. Statistical significance was accepted at $P<0.05$. Data are expressed as mean±SEM.

Results

Mortality

All sham animals survived the follow-up period, but 2 sham animals died during ^{125}I-Ang I infusion because of technical failure. Eight MI animals died within 12 hours after induction of MI, and 2 died during eprosartan treatment on days 7 and 15.

Hemodynamic and Neurohumoral Characteristics

One day after surgery, mean arterial pressure was lower and heart rate was higher in MI pigs than in sham pigs (Figure 1). Mean arterial pressure partially recovered in the MI group during the first week but remained below sham levels during follow-up. Plasma norepinephrine and epinephrine levels were similar in MI and sham animals, whereas plasma ANP and N-terminal ANP were higher in MI animals during the entire follow-up period. Captopril and eprosartan did not affect any of the hemodynamic or neurohumoral parameters (data not shown).

Remodeling

LV EDA and ESA were increased at 1 week after MI, so that EF was lower in MI animals (Table 1). EF recovered during the follow-up period, although LV EDA and ESA remained increased compared with sham values. Captopril and eprosartan blunted the increases in LV dimension, thereby slightly increasing EF.

After MI, the surviving myocardium hypertrophied, as reflected by the increased LV and RV weights and the ratios of LV and RV weight to body weight (Table 1). Captopril and eprosartan attenuated LV and RV hypertrophy.

Hemodynamics During Anesthesia

Hemodynamics did not differ between MI and sham animals, except for LV end-diastolic pressure and pulmonary arterial

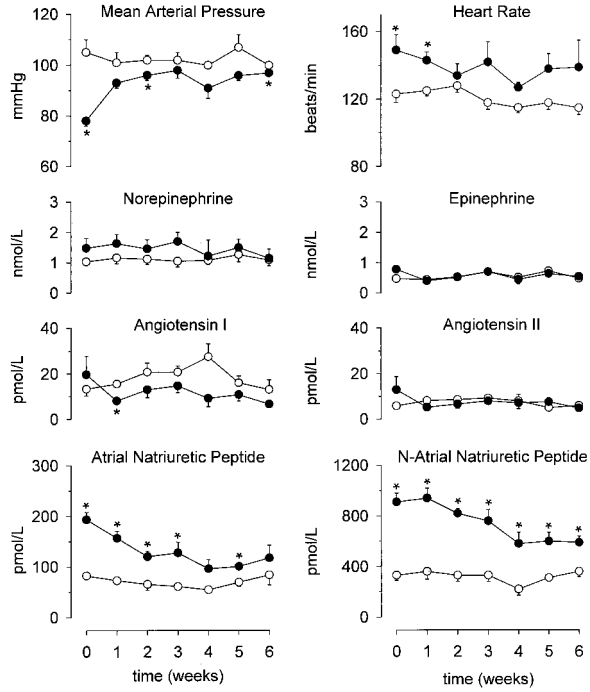

Figure 1. Mean arterial blood pressure, heart rate, and arterial plasma levels of neurohormones in sham (○) and MI (●) animals. N-atrial natriuretic peptide indicates N-terminal atrial natriuretic peptide. *$P<0.05$ vs sham.

pressure, which were higher ($P<0.05$) in MI pigs at 3 weeks (Table 1). Neither captopril nor eprosartan affected hemodynamics.

Angiotensin Levels in Plasma

In animals in the awake state, plasma Ang I and II levels (Figure 1) were somewhat higher than those levels in anesthetized animals (Table 2). However, the changes produced by MI and RAS blockade in animals in the awake state paralleled those in anesthetized animals. The Ang I and II levels in arterial and coronary venous plasma in MI animals were marginally higher (P=NS) than those in corresponding sham animals at 1 week. At 3 and 6 weeks, the levels were similar in both groups. Captopril increased plasma Ang I 5- to 10-fold but did not alter plasma Ang II in MI animals. The plasma Ang II/Ang I ratio decreased by ≈80% during captopril treatment, indicating effective ACE inhibition. Eprosartan increased plasma Ang I and II in MI animals 5- to 10-fold, without altering the plasma Ang II/Ang I ratio. Neither the steady-state ^{125}I-Ang I and II levels nor the ^{125}I-Ang II/Ang I ratios in arterial and coronary venous plasma differed between sham and MI animals at any time point. Captopril reduced the ^{125}I-Ang II levels and the ^{125}I-Ang II/Ang I ratio at both sampling sites, whereas eprosartan did not alter the steady-state plasma ^{125}I-Ang I and II levels.

Ang I Metabolism in Coronary Vascular Bed

Coronary ^{125}I-Ang I–Ang II conversion and ^{125}I-Ang I degradation in sham animals (Table 2) were not different from those reported in noninstrumented normal pigs.[20] MI did not alter Ang I conversion or degradation at any time point. Captopril inhibited conversion but did not affect degradation. Eprosartan affected neither conversion nor degradation.

Angiotensin Levels in Tissue

Ang I and II levels in the noninfarcted myocardium were higher in MI animals than in sham animals at 1 week but not at 3 and 6 weeks (Figures 2 and 3). In sham animals, Ang II levels were similar at all myocardial sites, whereas in MI animals, Ang II levels were lowest in the center of the infarct region and intermediate in the border zone. Both captopril

TABLE 1. Hemodynamics and Morphology

	1 wk		3 wk				6 wk	
Parameter	Sham (n=4)	MI (n=4)	Sham (n=5)	MI (n=6)	MI+Captopril (n=6)	MI+Eprosartan (n=5)	Sham (n=3)	MI (n=4)
MAP, mm Hg	95±4	90±3	93±4	96±6	85±4	89±2	94±6	100±3
Heart rate, bpm	115±5	124±11	101±6	103±8	103±5	113±8	118±9	130±8
Cardiac output, L/min	2.2±0.1	1.9±0.2	3.0±0.2	2.7±0.1	2.8±0.3	2.2±0.3	2.8±0.2	3.5±0.2
LV dP/dt_{max}, mm Hg/s	2343±214	2221±301	2053±138	1706±78	1872±162	1690±127	1940±104	2053±99
LVEDP, mm Hg	7±2	10±2	8±2	14±2*	11±1	11±3	7±1	9±2
PAP, mm Hg	21±4	31±1	16±3	28±2*	25±4	24±3	23±5	31±3
EDA, cm^2	8.1±0.1	13.6±0.3*	9.7±1.4	12.8±1.5	11.8±1.2	11.6±0.7	10.0±0.3	15.8±4.2
ESA, cm^2	4.1±0.5	10.4±0.6*	4.6±0.8	8.6±1.6*	6.9±1.0	7.0±1.3	5.7±0.8	9.7±2.8
EF, %	50±6	24±3*	53±4	33±9*	42±3*	42±8	43±6	40±4
BW, kg	23.7±0.5	23.5±1.2	30.0±1.4	28.7±1.1	28.3±1.4	28.4±0.6	36.5±1.5	40.1±1.5
LVW, g	75.1±1.4	90.6±1.1*	82.7±6.2	102.2±5.2*	88.2±3.2†	82.5±4.4†	102.1±1.0	131.3±12.2*
RVW, g	30.7±2.7	32.5±2.8	27.3±1.0	40.1±3.1*	31.9±1.5*†	30.3±2.7†	27.8±2.5	52.9±4.7*
LVW/BW, g/kg	3.17±0.11	3.90±0.22*	2.75±0.16	3.57±0.14*	3.17±0.25†	2.91±0.16†	2.80±0.09	3.32±0.42
RVW/BW, g/kg	1.30±0.10	1.39±0.12	0.91±0.04	1.39±0.06*	1.14±0.06*†	1.06±0.08†	0.77±0.10	1.34±0.16*

Values are mean±SEM. MAP indicates mean arterial pressure; LVEDP, LV end-diastolic pressure; PAP, pulmonary arterial pressure; BW, body weight; LVW, LV weight; and RVW, RV weight.

*$P<0.05$ vs sham; †$P<0.05$ vs untreated MI.

TABLE 2. Plasma Angiotensin Levels, Ang II/Ang I Ratios, and Coronary Ang I Metabolism

	1 wk		3 wk				6 wk	
Parameter	Sham (n=4)	MI (n=4)	Sham (n=5)	MI (n=6)	MI+Captopril (n=6)	MI+Eprosartan (n=5)	Sham (n=3)	MI (n=4)
Aorta								
Ang I, pmol/L	4.5±1.7	8.4±4.3	4.9±1.6	2.9±1.0	24.7±8.5*†	19.0±4.7*†	2.1±0.3	3.0±1.0
Ang II, pmol/L	1.4±0.1	6.6±2.9	3.6±1.2	1.9±0.5	4.2±1.6	16.5±3.4*†	1.1±0.1	2.1±0.7
Ang II/I ratio	0.54±0.22	0.82±0.08	0.62±0.08	0.73±0.04	0.16±0.05*†	0.92±0.09	0.50±0.12	0.49±0.20
^{125}I-Ang I, 10^3 cpm/L	546±76	501±52	420±28	545±106	620±63	563±73	369±28	459±50
^{125}I-Ang II, 10^3 cpm/L	263±19	255±33	232±46	286±62	100±15*†	385±15	186±12	352±81
^{125}I-Ang II/I ratio	0.50±0.05	0.51±0.05	0.56±0.12	0.57±0.14	0.17±0.03*†	0.73±0.09	0.51±0.01	0.74±0.13
Coronary vein								
Ang I, pmol/L	2.6±0.8	7.1±3.2	4.7±1.1	2.5±1.0	21.8±7.6*†	15.8±3.9*†	1.9±0.3	3.1±0.8
Ang II, pmol/L	1.1±0.2	4.8±2.0	2.8±0.8	1.4±0.3	4.5±1.4	18.0±3.2*†	1.3±0.2	1.9±0.6
Ang II/I ratio	0.58±0.22	0.63±0.08	0.49±0.08	0.66±0.10	0.18±0.06*†	0.96±0.16	0.65±0.25	0.59±0.13
^{125}I-Ang I, 10^3 cpm/L	257±35	323±35	203±31	267±74	455±50	225±28	193±25	250±26
^{125}I-Ang II, 10^3 cpm/L	277±23	268±38	223±48	289±52	97±11*†	346±16	223±33	349±58
^{125}I-Ang II/I ratio	1.12±0.08	0.82±0.05	1.14±0.16	1.22±0.20	0.21±0.04*†	1.67±0.26	1.17±0.17	1.21±0.17
Coronary ^{125}I-Ang I metabolism								
Fractional conversion, %	24±1	17±1	20±4	25±2	4±1*†	26±4	26±4	24±1
Fractional degradation, %	29±2	20±4	31±6	31±4	23±1	31±5	22±4	22±2

Values are mean±SEM.
*$P<0.05$ vs sham; †$P<0.05$ vs untreated MI.

and eprosartan increased Ang I and II 2- to 3-fold at all tissue sites in MI animals (Figures 2 and 3), without altering the tissue Ang II/Ang I ratios significantly (data not shown).

^{125}I-Ang I was undetectable in all regions, indicating that virtually all tissue Ang I had been produced in situ (Figure 2). ^{125}I-Ang II accumulated in cardiac tissue, reaching steady-state levels in the noninfarcted myocardium that were comparable to or higher than those in arterial plasma (Figure 4). ^{125}I-Ang II levels in the infarcted and border-zone lateral LV wall were ≈20% to 30% and ≈60% to 70% of those in arterial plasma. Blockade of cardiac ^{125}I-Ang II accumulation by eprosartan (Figure 4) indicates that this process is AT_1 receptor–mediated. There were no changes in ^{125}I-Ang II accumulation in the RV wall or in the infarcted or border-zone lateral LV wall during the follow-up period. In sham animals compared with normal noninstrumented animals, ^{125}I-Ang II accumulation in the noninfarcted anterior LV wall and the interventricular septum was increased at 1 week but not at 3 and 6 weeks,[9] whereas it was elevated in MI animals at 1 week and 3 weeks but not at 6 weeks. Captopril abolished the increased ^{125}I-Ang II accumulation in the noninfarcted myocardium of MI animals at 3 weeks. After the tissue ^{125}I-Ang II levels were used to correct for uptake of circulating Ang II, it appeared that the increase in cardiac Ang II levels in MI animals at 1 week was mainly due to uptake from plasma, whereas the increases in cardiac Ang II levels in captopril- and eprosartan-treated animals were due to Ang II generated in situ from locally synthesized Ang I (Figure 3).

Discussion

The present study demonstrates that after MI, the Ang II levels in the noninfarcted LV are transiently increased because of enhanced AT_1 receptor–mediated uptake of circulating Ang II. Captopril and eprosartan normalized or blocked this uptake and attenuated postinfarct remodeling but increased local cardiac Ang II production. These results suggest that the antihypertrophic effects of RAS blockade result not only from diminished AT_1 receptor stimulation but also from increased stimulation of growth-inhibitory AT_2 receptors.

LCXCA ligation caused LV and RV hypertrophy within 1 to 3 weeks. MI was accompanied by a transient but severe decrease in mean arterial pressure, which, via stimulation of renal renin release, may have caused the modest (nonsignificant) rise in plasma angiotensin levels in the first week. The hemodynamic and neurohumoral profile of the MI animals indicates that permanent LCXCA occlusion, which results in 15% to 25% infarction of the porcine LV, was associated with mild-to-moderate LV dysfunction, requiring minimal sympathetic activation to maintain cardiovascular homeostasis. This contrasts with an atrial pacing model in pigs, which led to severe heart failure and significant neurohumoral activation within 3 weeks.[21] Interestingly, in the latter model, LV AT_1 receptor density was decreased, whereas after MI, LV AT_1 receptors are usually upregulated.[10,22] The present findings are in agreement with such upregulation, inasmuch as the amount of plasma-derived ^{125}I-Ang II sequestered by the noninfarcted LV myocardium via AT_1 receptor–mediated internalization at 1 week and 3 weeks after MI was twice as high as that in a previous study in normal pigs.[9] However, similar increases were observed in sham animals at 1 week after surgery, suggesting that this procedure, possibly through the induction of a fibrogenic response,[22] is partly responsible for the increase in AT_1 receptor density. It must be realized that ^{125}I-Ang II uptake in the heart is mediated exclusively via

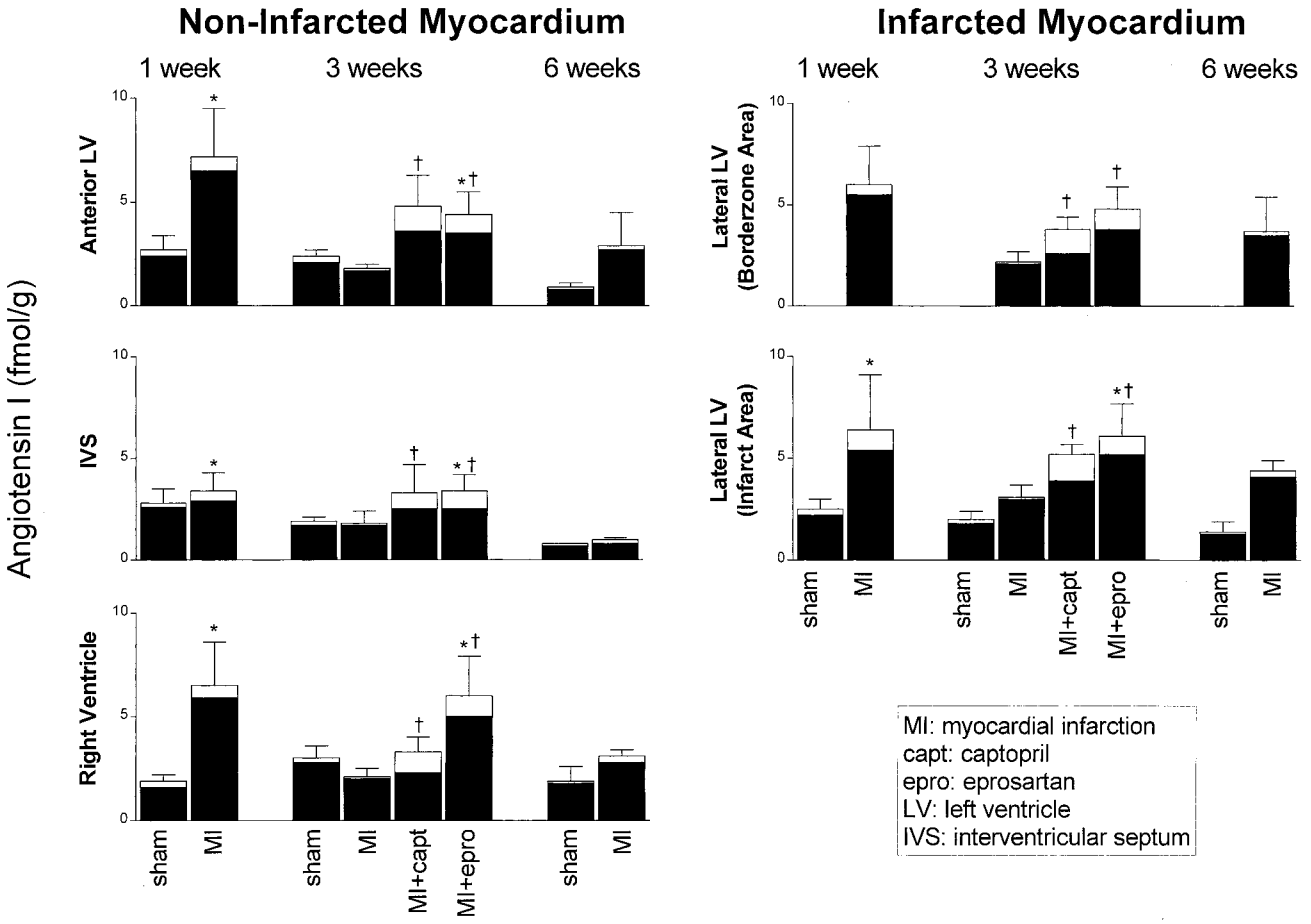

Figure 2. Cardiac Ang I levels. Black area in each bar represents amount of locally synthesized Ang I. *$P<0.05$ vs sham; †$P<0.05$ vs untreated MI.

AT_1 receptors, because AT_2 receptors do not internalize Ang II.[9,23] Furthermore, ^{125}I-Ang II formation from ^{125}I-Ang I that has diffused into the interstitial space does not contribute significantly to the steady-state tissue levels of ^{125}I-Ang II.[9,24]

The increase in AT_1 receptor–mediated uptake of circulating Ang II is responsible for the transient rise in Ang II levels in the noninfarcted LV myocardium, suggesting that circulating Ang II is among the factors initiating the development of LV hypertrophy, even when plasma Ang II is only marginally increased. This observation extends previous studies demonstrating that elevation of circulating Ang II, either through infusion of Ang II or through renovascular hypertension, induces biventricular hypertrophy within 2 weeks.[25] Increased ^{125}I-Ang II levels in the noninfarcted LV myocardium were no longer observed at 6 weeks, nor were they present in captopril-treated animals at 3 weeks. This, in combination with the notion that eprosartan fully blocked the uptake of circulating ^{125}I-Ang II at all myocardial locations, suggests that RAS blockade prevents the development of LV hypertrophy, at least in part, by attenuating or blocking the AT_1 receptor–mediated uptake of circulating Ang II.

In addition to their effects on cardiac ^{125}I-Ang II uptake, both captopril and eprosartan increased plasma and cardiac Ang I. Because cardiac renin is derived from the circulation, both under normal circumstances and after MI,[26–28] these increases most likely reflect the rise in renal renin release that normally accompanies RAS blockade. It is currently unknown whether enzymes other than renin (eg, cathepsin D[29]) contribute to Ang I generation in infarcted hearts. Similarly, the origin of cardiac angiotensinogen after MI has not been fully elucidated, although recent data suggest that most angiotensinogen in infarcted hearts,[27] as in normal hearts,[26] is plasma-derived.

In the eprosartan-treated pigs, plasma and cardiac Ang II rose in parallel with Ang I, whereas in the captopril-treated pigs, Ang II did not change in the circulation but increased in the heart. Consequently, the Ang II/Ang I ratio, a measure of ACE activity, was decreased in plasma but not in the heart after captopril treatment.

The rise in cardiac Ang II synthesis with captopril may have several causes. First, captopril may not have entered the heart in sufficient quantities. This seems unlikely, because in an earlier study, we observed a clear reduction in the cardiac Ang II/Ang I ratio after 3 days of treatment with the same dose of captopril.[2] It is also unlikely in view of the favorable cardiac effects of captopril in humans after MI.[1] Second, cardiac Ang II synthesis might occur at intracellular sites[24] that cannot be reached by ACE inhibitors. Third, the initial captopril-induced blockade of cardiac tissue Ang I–Ang II conversion[2] may have been compensated for during long-

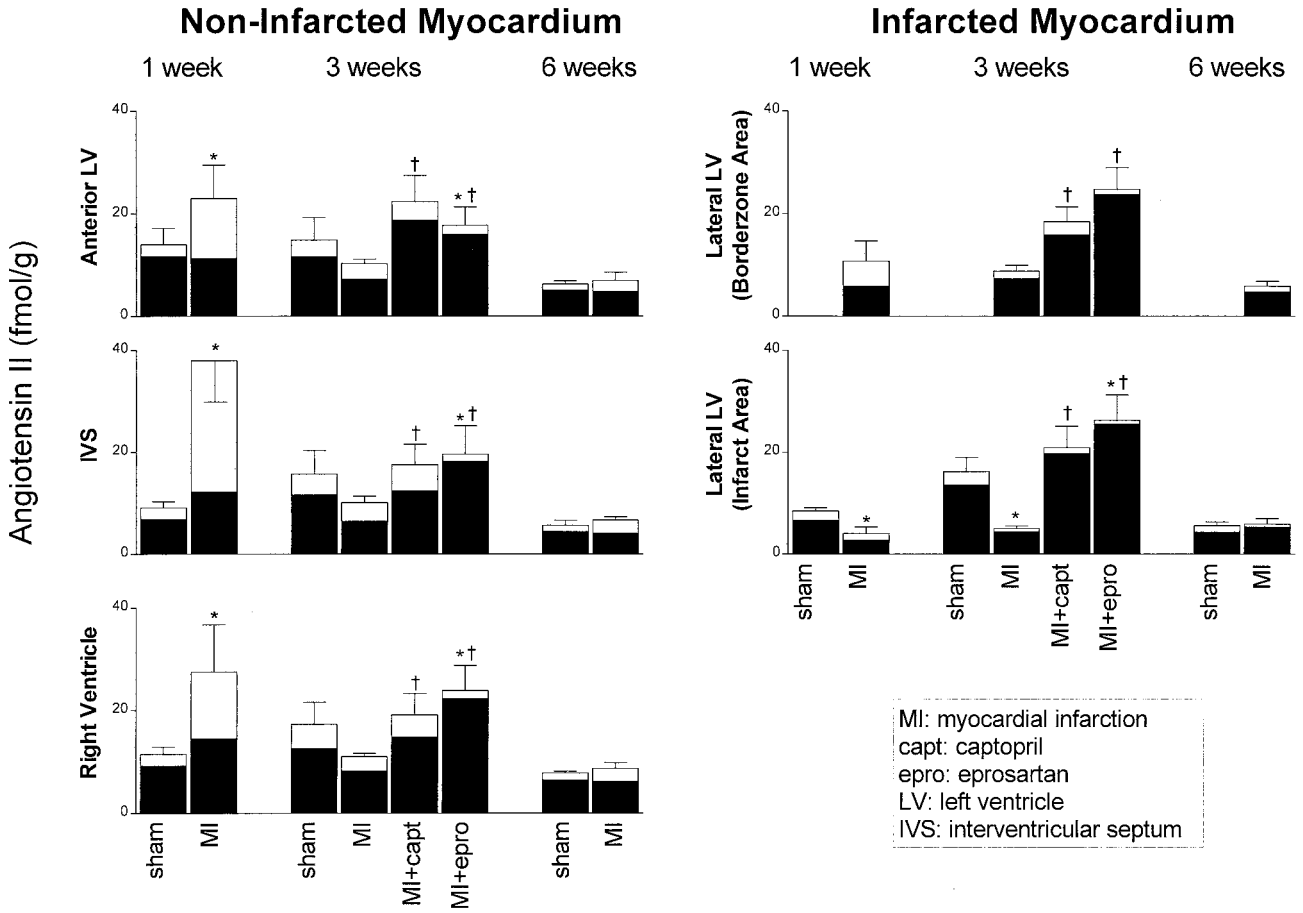

Figure 3. Cardiac Ang II levels. Black area in each bar represents amount of locally synthesized Ang II. *$P<0.05$ vs sham; †$P<0.05$ vs untreated MI.

term treatment with this drug, either through an upregulation of ACE[5] or because alternative converting enzymes such as chymase[6] have come into play. Our findings do not provide evidence for changes in cardiac or coronary ACE activity, because neither the cardiac Ang II/Ang I ratio nor coronary Ang I–Ang II conversion was altered in MI animals. Although MI-induced ACE upregulation has been described,[22,30] transgenic rats overexpressing cardiac ACE 40-fold have normal cardiac Ang II levels.[31] Moreover, we failed to observe ACE gene insertion/deletion polymorphism–related differences in vascular Ang I–Ang II conversion despite profound effects of this polymorphism on plasma and tissue ACE levels.[32] Thus, elevated cardiac ACE levels, if present in MI pigs, do not necessarily result in elevated Ang II levels or Ang II/Ang I ratios but may explain why captopril decreases the cardiac Ang II/Ang I ratio less effectively. Furthermore, chymase is present in the porcine heart,[33] and its concentration increases after MI.[34]

Does the rise in cardiac Ang II production during RAS blockade have a physiological function? If caused by chymase, one must realize that chymase is present in the cytosol of mast cells and in the extracellular matrix,[6] whereas ACE is located on the cell membrane, in proximity to AT_1 receptors.[22,35] Consequently, Ang II generated by chymase may couple less efficiently to AT_1 receptors than Ang II generated by ACE.[36]

AT_2 receptor antagonism abolishes the beneficial effects of AT_1 receptor blockade in MI rats.[37] This raises the possibility that the rise in Ang II in the present study results in stimulation of growth-inhibitory AT_2 receptors.[13,23] The AT_2 receptor density is increased in infarcted and failing hearts,[10,11] and because the net effect of Ang II depends on the AT_1/AT_2 receptor ratio,[13] it is indeed conceivable that growth inhibition occurs not only during AT_1 receptor antagonism but also during ACE inhibition, because the latter prevents the rise in AT_1 receptor density after MI (the present study) and is possibly accompanied by chymase-dependent Ang I–Ang II conversion at sites distant from AT_1 receptors.[35,36]

Enhanced AT_2 receptor stimulation, together with diminished AT_1 receptor stimulation, might also explain why captopril and eprosartan prevented RV hypertrophy in MI animals. Both drugs minimally affected the increase in pulmonary arterial pressure that, in combination with the elevated RV Ang II levels at 1 week after MI, may have contributed to this hypertrophy.

Interestingly, the lowest ^{125}I-Ang II accumulation and local Ang II production were observed in the infarct area. The latter finding could indicate that in this area, possibly secondary to the reduced blood flow, renin uptake was diminished in parallel with the diminished ^{125}I-Ang II uptake. Alternatively, and perhaps more likely in view of the unaltered local Ang I

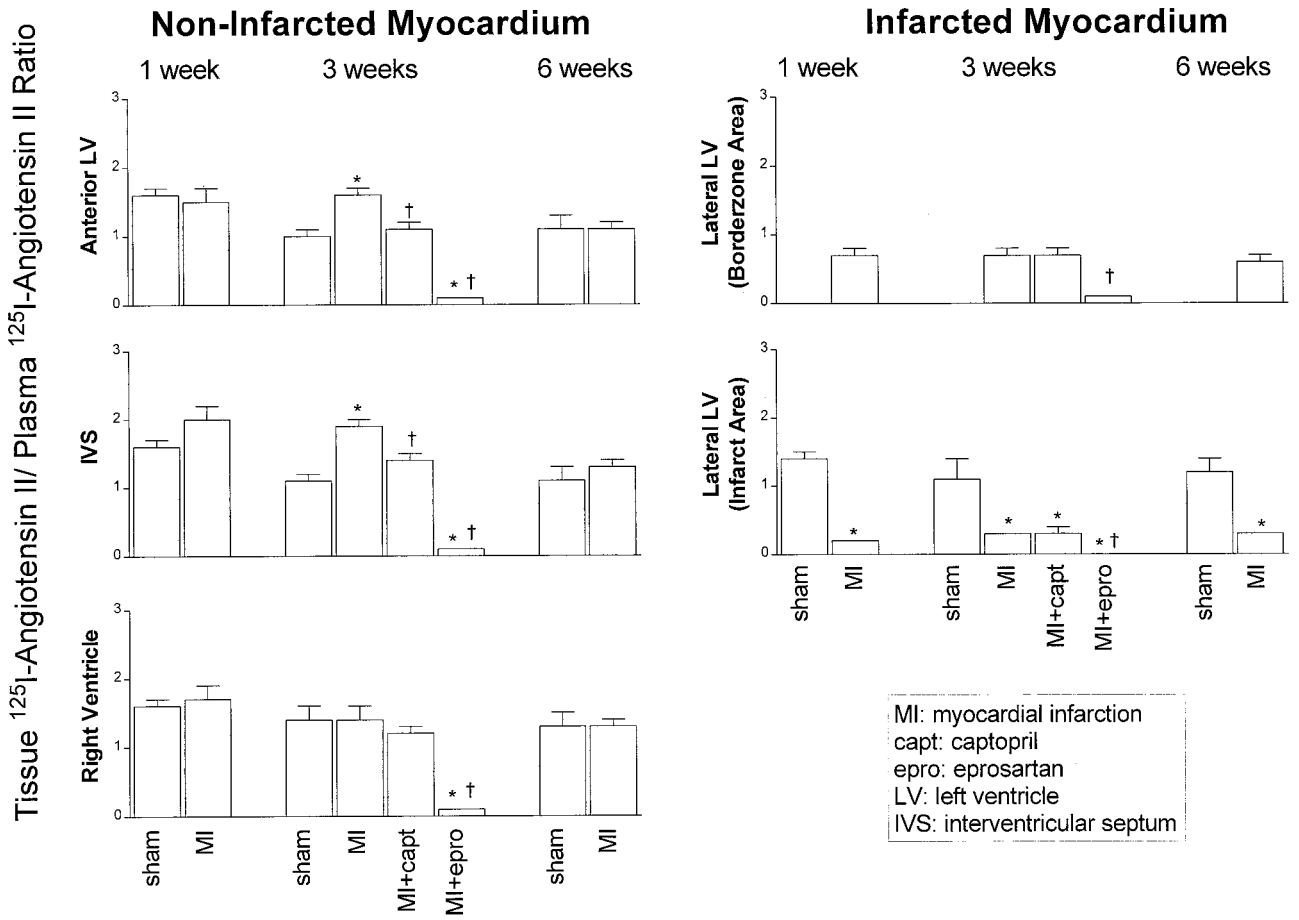

Figure 4. Tissue/plasma concentration ratios of ^{125}I-Ang II. *$P<0.05$ vs sham; †$P<0.05$ vs untreated MI.

production in this area, the low ^{125}I-Ang II and Ang II levels might be the consequence of increased local Ang II degradation.

In summary, MI results in a transient upregulation of AT_1 receptors in spared noninfarcted myocardium, which will cause enhanced sequestration of plasma Ang II even in the absence of changes in the circulating RAS. RAS inhibitors prevent the rise in plasma Ang II sequestration, either by interfering with myocardial AT_1 receptor upregulation or by blocking these receptors. Furthermore, these inhibitors increase tissue Ang II production, which through stimulation of cardiac AT_2 receptors may minimize postinfarct remodeling.

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