Activation of Cardiac Aldosterone Production in Rat Myocardial Infarction

Effect of Angiotensin II Receptor Blockade and Role in Cardiac Fibrosis

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Background—This study analyzed the regulation and the role of the cardiac steroidogenic system in myocardial infarction (MI).

Methods and Results—Seven days after MI, rats were randomized to untreated infarcted group or spironolactone- (20 and 80 mg · kg⁻¹ · d⁻¹), losartan- (8 mg · kg⁻¹ · d⁻¹), spironolactone plus losartan-, and L-NAME– (5 mg · kg⁻¹ · d⁻¹) treated infarcted groups for 25 days. Sham-operated rats served as controls. In the noninfarcted myocardium of the left ventricle (LV), MI raised aldosterone synthase mRNA (the terminal enzyme of aldosterone synthesis) by 2.0-fold and the aldosterone level by 3.7-fold. Conversely, MI decreased 11β-hydroxylase mRNA (the terminal enzyme of corticosterone synthesis) by 2.4-fold and the corticosterone level by 1.9-fold. MI also induced a 1.9-fold increase in cardiac angiotensin II level. Such cardiac regulations were completely prevented by treatment of the infarcted heart with losartan. The MI-induced collagen deposition in noninfarcted LV myocardium was prevented by 1.6-fold by both low and high doses of spironolactone and by 2.5-fold by losartan. In addition, norepinephrine level was unchanged in infarcted heart but was attenuated by both losartan and spironolactone treatments.

Conclusions—MI is associated with tissue-specific activation of myocardial aldosterone synthesis. This increase is mediated primarily by cardiac angiotensin II via AT₁-receptor subtype and may be involved in post-MI ventricular fibrosis and in control of tissue norepinephrine concentration. (Circulation. 1999;99:2694-2701.)

Key Words: myocardial infarction • aldosterone • angiotensin • collagen

In addition to the classic adrenal biosynthetic pathway, steroid hormone production has been evidenced in extra-adrenal tissues, namely brain1 and vessels.2 The existence of a steroidogenic system has also been suggested in cardiac tissue.3 Moreover, the gene expression of terminal enzymes of corticosterone and aldosterone synthesis, 11β-hydroxylase (11β-OHase) and aldosterone synthase (aldo-synthase), respectively, and production of both steroids have recently been demonstrated within rat heart,4 confirming the potential for steroid metabolism in cardiac tissue.

Mineralocorticoid receptor (MR) and 11β-hydroxysteroid dehydrogenase, which confers mineralocorticoid selectivity to aldosterone target tissues, have been detected in heart.5,6 Aldosterone excess triggers cardiac fibrosis5,6 and electrolyte imbalance.9 Aldosterone also regulates myocardial norepinephrine uptake.10 Taken together, these reports emphasize the potential role of cardiac aldosterone in the regulation of tissue function.

A selective activation of cardiac angiotensin II (Ang II)–generating pathway has been reported in the rat model of chronic myocardial infarction (MI).1,12,13 ACE inhibitors and Ang II AT₁-receptor subtype receptor antagonists reduce the post-MI cardiac remodeling.1,14,15 Because the plasma renin-angiotensin system is not consistently activated after MI,11,15 part of the beneficial effects of ACE inhibitors and AT₁-receptor subtype receptor blockade have been attributed to inhibition of cardiac Ang II–related action.

We hypothesized that the tissue aldosterone system may be also activated in MI and may contribute to the pathophysiology of this disease state. Thus, the present study aimed to (1) analyze the regulation of this cardiac steroidogenic system in rat MI; (2) determine whether cardiac Ang II, atrial natriuretic factor (ANF), and nitric oxide (NO), ie, the major stimuli of adrenal aldosterone biosynthesis,16,17,18 contribute to such regulations; and (3) assess the role of cardiac aldosterone in the regulation of postinfarction ventricular fibrosis and myocardial norepinephrine level.

This study provides the first evidence that MI is associated with an increase in myocardial aldosterone production, which may be involved in post-MI ventricular fibrosis and in control of tissue norepinephrine concentration.
Methods

Animals
The study, which was conducted in accordance with both institutional guidelines and those formulated by the European Community for experimental animal use (L358-86/609/EEC), used 3-month-old male Wistar rats (Iffa Credo, Lyon, France). Left ventricular (LV) infarction was produced by ligation of the left anterior descending coronary artery as previously described.19 Sham-operated animals were treated similarly, except that the ligature around the coronary artery was not tied. Seven days after coronary ligation, animals were randomly divided into 6 groups (n=6 per group), each group receiving one of the following treatments: (1) low dose of spironolactone (aldosterone receptor blockade, 20 mg·kg⁻¹·d⁻¹ in drinking water, Sigma), (2) high dose of spironolactone (80 mg·kg⁻¹·d⁻¹), (3) losartan (Ang II receptor blockade, 8 mg·kg⁻¹·d⁻¹ in drinking water, Merck Industries), (4) spironolactone and losartan (20 and 8 mg·kg⁻¹·d⁻¹, respectively, in drinking water), (5) N⁶-nitro-L-arginine methyl ester (L-NAME, NO synthase inhibitor, 5 mg·kg⁻¹·d⁻¹ in drinking water), and (6) untreated MI. Noninfarcted and nontreated sham-operated animals served as a control group.

Experimental Protocol
After 25 days of treatment, rats were anesthetized by intraperitoneal injection of ketamine (100 mg/kg), xylazine (10 mg/kg), and atropine (0.05 mg/kg). Functional parameters were then measured in closed-chest anesthetized rats. An ultraminiature catheter pressure transducer, inserted into the LV, was attached to a transducer control unit (model TC50, Millar instruments, Inc) connected to the data acquisition unit (MP100, Biopac Systems, Inc). Arterial and LV systolic pressures and the first derivatives for the maximal rate of LV pressure development (+LV dP/dt) and maximal rate of relaxation (−LV dP/dt) were then measured. At the end of the recording period, blood was collected for measurements of hormone concentration and renin activity. Subsequently, hearts were perfused through the aorta with an ice-cold NaCl 0.9% buffer to wash out plasma components. The pulmonary artery and vena cava were opened to drain the blood out of the heart and to avoid volume overload. After cessation of beating, hearts were excised and immediately dropped into an ice-cold NaCl 0.9% buffer. Transverse sections of heart were then embedded in mounting medium and frozen in liquid nitrogen–cooled ice-cold NaCl 0.9% buffer to wash out plasma components. The septum, including the border zone, was then removed from the remaining heart. In sham-operated animals, corresponding parts of the heart were discarded. The LV, including the septum, was separated from the right ventricle in the remaining heart and stored, as were the adrenal glands, at −70°C until use. All the experiments were then performed on the noninfarcted LV myocardium of each infarcted heart and on the corresponding part of the LV in sham-operated rats.

Total RNA Extraction
Total RNA was extracted from hearts and adrenal glands according to the Trizol reagent protocol (Life Technologies). The yields of total RNA extracted were similar in sham-operated rats and MI and in treated and untreated MI.

Quantification of 11β-OHase and Aldo-Synthase mRNAs by RT-PCR

Quantitative RT-PCR Protocol
Reverse transcription–polymerase chain reaction (RT-PCR) was performed as previously described.4 Quantifications were done with a synthetic cRNA as a standard corresponding to the 11β-OHase PCR product ligated with a 100-bp insert (PvuII/XhoI fragment of pBluescript II SK phagemid). After RT-PCR, we obtained a PCR product of 446 bp (Figure 1A).

Steroids, Ang II, and Norepinephrine Assay
Cardiac levels of steroids, Ang II, and unconjugated norepinephrine were assessed as previously described.4,20,21 The protein concentration of cardiac homogenate was determined according to the method of Bradford,22 with BSA used as standard.

Differentiation of 11β-OHase and Aldo-Synthase PCR Products
PCR products were size-differentiated with XhoI, which hydrolyzed the aldose-synthase PCR product into 2 fragments of 177 and 169 bp. XhoI did not affect the PCR products of 11β-OHase or internal standard as previously described.4

PCR products were separated on a 5% polyacrylamide gel, and radioactive signals were analyzed with a computer-based imaging system (Bas 1000, Fuji Medical Systems).
Quantification of MR and Glucocorticoid Receptor mRNAs by Ribonuclease Protection Assay

Specific antibodies against rat MR are not available, and binding studies are complicated by the binding of aldosterone and glucocorticoids to heterologous receptors. As a consequence, we have chosen to use the MR and glucocorticoid receptor (GR) mRNA levels as an indirect measure of the MR and GR regulation. Ribonuclease protection assay was performed as previously described. Protected products were separated on a 5% polyacrylamide-urea gel, and radioactive signals were analyzed with the Bas 1000. The values obtained were normalized by those obtained for GAPDH mRNA.

ANF Gene Expression

Northern blot analysis of ANF was performed with 10-μg samples of RNA as previously described. The radioactive signals were analyzed with the Bas 1000.

Determination of Infarct Size and Collagen Morphometry

Transverse myocardial sections (5 μm thick) were stained with collagen-specific Sirius red stain (0.5% in saturated picric acid). Each field was digitized on a Macintosh Performa 5320 by a gray-level camera (Hamamatsu) mounted on a light microscope (Leica). Infarct size was determined by planimetric measurement with digital image analysis software (Optilab, Graftek). To calculate infarct size, the ratio of scar length to total length of each slice was measured and expressed as a percentage, as previously described. Collagen was then quantified at 100 magnification with Optilab software. Perivascular areas were not included in this analysis.

Statistical Analysis

Results are expressed as mean±SE. One-way ANOVA was used to compare each parameter in the 6 experimental groups. Post hoc Student’s t test comparisons were then performed to identify which group differences accounted for the significant overall ANOVA. A value of P<0.05 was considered significant. Significance was adjusted for the number of comparisons by Bonferroni correction. Comparisons were then considered not significant unless the corresponding probability value was <0.004.

Results

Infarct Size, Cardiac Hypertrophy, and Physiological Data

None of the animals died during the post-MI 25-day treatments. Mean infarct size and body weight did not differ significantly between the experimental groups (Table 1). MI induced a slight cardiac hypertrophy, as demonstrated by the increase in ratio of heart weight to body weight by 1.2-fold. Only treatments with losartan prevented such cardiac hypertrophy. Aortic systolic and diastolic pressures, LV systolic pressure, and HR, heart rate. n=6 for each group. Values are mean±SEM.

Sham indicates sham-operated rat; Spi(1), spironolactone 20 mg·kg⁻¹·d⁻¹; Spi(2), spironolactone 80 mg·kg⁻¹·d⁻¹; Los, losartan-treated infarcted rats; BW, body weight; HW, heart weight; AoSP and AoDP, aortic systolic and diastolic pressures; LVSP, LV systolic pressure; and HR, heart rate. n=6 for each group. Values are mean±SEM. *P<0.05 vs sham-operated rat; †P<0.05, ‡P<0.01 vs MI; §P<0.05 MI+Spi(1) vs MI+Spi(2).

Cardiac Steroidogenic System in MI

Cardiac Gene Expression

In the noninfarcted area of the LV, MI induced a 2.0-fold increase in aldo-synthase mRNA level and a 2.4-fold decrease in 11β-OHase mRNA level (Figure 1). Losartan, but not spironolactone, completely prevented such changes. L-NAME also induced a 1.5-fold decrease in aldo-synthase gene expression, but without reaching statistical significance.

In the infarcted area of the LV, aldo-synthase gene expression was undetectable, whereas 11β-OHase (2017±214 mol/μg of total RNA) was significantly downregulated compared with noninfarcted LV myocardium (9867±1147 mol/μg total RNA, P<0.001).

Cardiac Steroid Production

In noninfarcted LV myocardium, MI enhanced aldosterone production by 3.7-fold (Figure 2A). In contrast, corticosterone level fell by 1.9-fold (Figure 2B). Only treatments with losartan prevented these regulations. As for aldo-synthase mRNA levels, the L-NAME–induced 1.4-fold decrease in aldosterone levels did not reach statistical significance. However, L-NAME treatment raised corticosterone level significantly, by 2- and 4-fold, compared with sham-operated and untreated MI rats, respectively. In addition, the level of deoxycorticosterone (DOC, the precursor of both aldosterone and corticosterone) was unchanged in the experimental groups (Figure 2C).

**TABLE 1.** Infarct Size and Anatomic and Physiological Parameters

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<tr>
<th></th>
<th>Sham</th>
<th>MI</th>
<th>MI+Spi(1)</th>
<th>MI+Spi(2)</th>
<th>MI+Los</th>
<th>MI+Spi(1)+Los</th>
<th>MI+L-NAME</th>
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<tr>
<td>BW, g</td>
<td>414±8</td>
<td>433±7</td>
<td>423±21</td>
<td>427±11</td>
<td>409±13</td>
<td>392±13</td>
<td>417±20</td>
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<td>HW, mg</td>
<td>1071±21</td>
<td>1300±63*</td>
<td>1212±62</td>
<td>1190±88</td>
<td>958±36</td>
<td>952±14*</td>
<td>1290±34*</td>
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<td>HW/BW, mg/g</td>
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<td>2.86±0.20</td>
<td>2.79±0.17</td>
<td>2.35±0.28†</td>
<td>2.42±0.14†</td>
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<td>AoSP, mm Hg</td>
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<td>77±3†</td>
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<td>139±3*</td>
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<td>84±6</td>
<td>84±8</td>
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<td>73±3</td>
<td>104±4*</td>
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<td>LVSP, mm Hg</td>
<td>135±20</td>
<td>131±5</td>
<td>118±6</td>
<td>109±10</td>
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<td>145±11</td>
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<td>+LVdP/dt, mm Hg/s</td>
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<td>5564±371</td>
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<td>4981±667</td>
<td>6207±282</td>
<td>5625±684</td>
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<td>−LVdP/dt, mm Hg/s</td>
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<td>5951±583</td>
<td>4923±328</td>
<td>5263±484</td>
<td>4599±579</td>
<td>6277±581</td>
<td>5171±599</td>
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<td>HR, bpm</td>
<td>437±12</td>
<td>475±14</td>
<td>432±21</td>
<td>411±43</td>
<td>426±20</td>
<td>422±12</td>
<td>417±23</td>
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In the infarcted area of the LV, MI resulted in an almost complete suppression of DOC (<50±20 pg/mg of protein), corticosterone (500±60 pg/mg of protein), and aldosterone (<10±4 pg/mg of protein) levels compared with the noninfarcted part of the LV (P<0.001).

**Cardiac MR and GR mRNA Levels**

MR and GR gene expressions were unaffected in untreated MI (123±30% and 109±21%), low-dose spironolactone– (113±26% and 106±23%), high-dose spironolactone– (96±21% and 104±11%), losartan- (101±16% and 96±17%), losartan and spironolactone– (122±13% and 111±7%), and L-NAME– (129±14% and 104±19%) treated infarcted rats, respectively, compared with 100% in sham-operated animals (data not shown).

**Adrenal Steroidogenic System in MI**

**Adrenal Gene Expression**

As shown in Figure 3, MI did not change adrenal aldosterone gene expression. Only treatments of infarcted rats with high doses of spironolactone and L-NAME affected aldosterone mRNA levels. No significant differences in 11β-HOase gene expression were evidenced among experimental groups.

**Adrenal Steroid Production**

Similarly, the plasma level of aldosterone was unchanged by MI but raised in infarcted rats treated with the high dose of spironolactone and decreased in those treated with L-NAME. Treatment with L-NAME was also associated with a 1.6-fold rise in corticosterone levels. Plasma DOC concentration was unaffected in experimental groups (Table 2).

**Angiotensin II Level**

**Cardiac Ang II Level**

In noninfarcted LV myocardium, MI was associated with a 1.9-fold increase in cardiac Ang II content. Only treatments with losartan prevented the MI-induced rise in tissue Ang II level (Figure 4A).

In the infarcted area of the LV, the same patterns of regulation were evidenced (Figure 4B). However, cardiac Ang II levels were 1.8-fold higher in the infarcted area than in noninfarcted LV myocardium of infarcted heart (P<0.01).

**Plasma Ang II Level**

In contrast to cardiac level, plasma Ang II and renin levels were not different between the sham-operated, untreated infarcted, low-dose spironolactone–treated, and L-NAME–treated infarcted rat groups (Table 2). Conversely, plasma
Ang II and renin activity levels were increased in losartan-, losartan plus spironolactone–, and high-dose spironolactone–treated infarcted rats, in agreement with previous studies.24,25

**ANF Gene Expression in Noninfarcted LV Myocardium**

As shown in Figure 5, ANF mRNA was induced in the noninfarcted area of the LV of infarcted rat hearts, as previously described.11 Moreover, treatment of infarcted rats with the high dose of spironolactone decreased, whereas treatment with L-NAME increased, LV ANF gene expression.

**Interstitial Fibrosis in Noninfarcted LV Myocardium**

Sirius red staining revealed a 2.8-fold increase in myocardial fibrosis in the noninfarcted part of the LV of infarcted hearts compared with sham-operated animals (Figure 6A and 6B). Interestingly, collagen accumulation in infarcted heart was 1.6-fold attenuated by chronic aldosterone receptor blockade, whatever the dose of spironolactone used. Interstitial fibrosis was prevented a further 2.5- and 2.4-fold by chronic AT1 receptor blockade alone or associated with a low dose of spironolactone, respectively.

**Norepinephrine Level in Noninfarcted LV Myocardium**

Cardiac norepinephrine level was unaffected by MI and by treatment of infarcted hearts with L-NAME. In contrast, both losartan and spironolactone (low and high doses) treatments decreased cardiac norepinephrine concentration (Figure 7).

**Discussion**

The major findings of this study are the following: i) aldo-synthase mRNA and aldosterone levels were raised whereas that of 11β-OHase and corticosterone were decreased in the noninfarcted area of LV; ii) MI is also associated with a rise in cardiac Ang II level; iii) these regulations were prevented by AT1 -subtype receptor blockade using losartan, but not by MR antagonist spironolactone; iv) in contrast, adrenal aldo-synthase and 11β-OHase gene expressions and plasma steroid levels were unaffected by MI and v) both spironolactone and losartan prevented myocardial fibrosis associated with MI and attenuated cardiac norepinephrine level.

**TABLE 2. Plasma Hormone Data**

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<tr>
<th></th>
<th>Sham</th>
<th>MI</th>
<th>MI + Spi(1)</th>
<th>MI + Spi(2)</th>
<th>MI + Los</th>
<th>MI + Spi(1)+Los</th>
<th>MI + L-NAME</th>
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<tr>
<td>Aldosterone, pg/mL</td>
<td>231±19</td>
<td>226±38</td>
<td>263±23</td>
<td>400±15†</td>
<td>212±53</td>
<td>331±83</td>
<td>106±17†</td>
</tr>
<tr>
<td>Corticosterone, ng/mL</td>
<td>29±2</td>
<td>27±2</td>
<td>27±5</td>
<td>19±2</td>
<td>26±3</td>
<td>29±3</td>
<td>46±5†</td>
</tr>
<tr>
<td>DOC, pg/mL</td>
<td>334±409</td>
<td>276±487</td>
<td>334±110</td>
<td>206±95</td>
<td>275±191</td>
<td>286±234</td>
<td>355±930</td>
</tr>
<tr>
<td>Ang II, ng/mL</td>
<td>1.8±0.4</td>
<td>1.4±0.2</td>
<td>1.9±0.6</td>
<td>4.9±0.2†</td>
<td>9.1±1.6†</td>
<td>6.7±0.9†‡</td>
<td>1.4±0.3</td>
</tr>
<tr>
<td>Plasma renin activity, pg · mL⁻¹ · h⁻¹</td>
<td>14±4</td>
<td>12±3</td>
<td>19±4</td>
<td>28±2†</td>
<td>36±7†</td>
<td>31±8†</td>
<td>11±3</td>
</tr>
</tbody>
</table>

Abbreviations as in Table 1. n=6 for each group. Values are mean±SEM.

*P<0.05 vs sham-operated rat; †P<0.05, ‡P<0.01 vs MI.

**Figure 4.** Angiotensin II content in the noninfarcted myocardium (A) and infarcted myocardium (B) of rat LV. Values are mean±SEM, n=6 per group. *P<0.05, **P<0.01 versus sham-operated rat and P<0.05, P<0.01 versus MI.

**Figure 5.** Northern blot analysis shows total RNA in the noninfarcted myocardium of rat LV hybridized with ANF (upper panel) and 18S (lower panel) oligonucleotide probes. Same as Figure 1.
Activation of Cardiac Steroidogenic Pathway
In this study, we evidence a tissue-specific upregulation of aldosterone-generating pathway in noninfarcted LV myocardium. In contrast, corticosterone production was decreased. Because both steroids are synthesized from a common precursor DOC, this suggests that MI modified the cardiac steroids biosynthetic pathway by increasing aldosterone synthesis at the expense of corticosterone production.

Accumulating evidences indicate that myocardial Ang II–generating pathway is also activated in MI. Indeed, increased cardiac expression of angiotensinogen, ACE and AT1 receptor proteins, ACE activity and Ang II content have been previously described in infarcted hearts. We confirm and extend these previous studies because we evidence that cardiac Ang II level was enhanced in the noninfarcted LV myocardium. Ang II is an important regulator of aldosterone biosynthesis and secretion in adrenal cortex as in heart. Ang II also modulates the corticosterone-generating pathway. However, the effect of Ang II on 11β-OHase mRNA and corticosterone levels is still controversial because Ang II has been shown to decrease, increase or not change 11β-OHase gene expression in adrenals. Nevertheless, in our study all changes associated with MI were prevented by Ang II AT1-subtype receptor blockade independently of variations in hemodynamic parameters. Because plasma renin-angiotensin system is not activated in MI and cardiac Ang II level is enhanced, our results support the conclusion that the MI-induced increase in tissue Ang II level triggers the rise in cardiac aldosterone production and the decrease in cardiac corticosterone synthesis.

Such a role of tissue Ang II on local aldosterone synthesis has been previously demonstrated in adrenals. Conversely, despite the marked increase in cardiac Ang II, the cardiac steroidogenic pathway was completely attenuated in the infarcted area. Because both ald-o-synthase and 11β-OHase are mitochondrial enzymes, it is likely that the lack of oxygen and the structural mitochondrial damage in such area blocked these enzymatic reactions.

NO is required for the action of both Ang II and ACTH on aldosterone production in isolated rat adrenal capsular tissue. In the same way, using a NO synthase inhibitor, we evidenced that NO may participate to the MI-induced regulation of cardiac aldosterone production. In contrast, ANF was not involved in these regulations because changes in cardiac aldosterone production were independent on cardiac ANF mRNA level. Nevertheless, ANF has a known inhibitory effect on adrenal aldosterone secretion. Therefore, the changes in ANF gene expression evidenced in both high dose of spironolactone and L-NAME treated infarcted hearts may also participate to cardiac aldosterone level variations.

Role of Cardiac Aldosterone
Cardiac Fibrosis
Chronic MR blockade is effective in preventing myocardial fibrosis of noninfarcted area of LV, independently of
blood pressure changes. Because plasma aldosterone level is unchanged and cardiac aldosterone production increased in MI, we may postulate that spironolactone effect is related to blockade of cardiac MR and consequently to blockade of cardiac aldosterone action. Several studies evidence a stimulatory effect of aldosterone on cardiac collagen synthesis. Although the mechanism of aldosterone-induced cardiac fibrosis remains unclear, these reports support the hypothesis that the increase in myocardial aldosterone level may be involved in post-MI ventricular fibrosis. Both doses of spironolactone attenuate cardiac fibrosis to the same extent. It is then likely that the enhanced concentration of spironolactone may be counterbalanced by the increase in both cardiac and plasma aldosterone levels associated with such treatment. Spironolactone, whatever the dose, is less effective than losartan in attenuating collagen accumulation after MI. Indeed, chronic AT1-receptor blockade totally prevents cardiac hypertrophy and fibrosis in noninfarcted LV myocardium as previously described. We also demonstrate that the beneficial effect of losartan on cardiac remodeling is related to both AT1-subtype receptor blockade and tissue Ang II levels reduction, in agreement with previous studies. In contrast, spironolactone attenuates cardiac fibrosis independently of changes in myocardial Ang II content. The antifibrotic effect of spironolactone is therefore related to blockade of aldosterone action and not to changes in tissue Ang II levels.

**Cardiac Norepinephrine Level**

This study also evidence that cardiac aldosterone regulates cardiac norepinephrine level. The presumed mechanism of such an effect involves the reduction by aldosterone of norepinephrine uptake. Therefore, blockade of aldosterone action using spironolactone increases norepinephrine uptake contributing then to the decrease in cardiac norepinephrine concentration. Indeed, norepinephrine is rapidly inactivated and metabolized on uptake by cardiac cells. Unexpectedly, despite the increase in both cardiac Ang II and aldosterone levels, cardiac norepinephrine concentration was unchanged in untreated MI. Such lack of variation in norepinephrine level may be related to structural changes and to loss of neuronal activity in infarcted myocardium. Nevertheless, the aldosterone-induced regulation of cardiac norepinephrine levels may contribute to the norepinephrine-related arrhythmogenic effects. In this view, spironolactone decreased ventricular arrhythmias in patients with heart failure.

In conclusion, we provide the first evidence that MI is associated with tissue-specific activation of cardiac aldosterone-generating pathway. Such increase in myocardial aldosterone level is mediated by cardiac Ang II via AT1-subtype receptor and may be involved in post-MI ventricular fibrosis and in control of tissue norepinephrine level.

**Acknowledgments**

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**References**

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