Aldosterone Production Is Activated in Failing Ventricle in Humans

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Background—Recent reports have indicated that aldosterone is produced in extra-adrenal tissues in animals. The present study was designed to examine whether aldosterone is produced in human heart.

Methods and Results—Plasma levels of aldosterone, BNP, and angiotensin-converting enzyme were measured in anterior interventricular vein (AIV), coronary sinus (CS), and aortic root (Ao), respectively, in 20 patients with left ventricular systolic dysfunction (LVSD), 25 patients with LV diastolic dysfunction (LVDD), and 23 control subjects. Aldosterone levels were significantly higher in AIV and CS than Ao in LVSD (98 ± 6 versus 72 ± 9 pg/mL, P < 0.001, and 97 ± 11 versus 72 ± 9 pg/mL, P < 0.001, respectively) and LVDD (87 ± 10 versus 71 ± 9 pg/mL, P < 0.01, and 84 ± 10 versus 71 ± 9 pg/mL, P < 0.01, respectively) groups, but no differences were observed in levels for these sites in the control group. Levels of ACE activity and BNP were also higher in AIV than Ao in both LV dysfunction groups. The difference in aldosterone levels between AIV and Ao and those in BNP and angiotensin-converting enzyme had a significant positive correlation with LVEDP and a significant negative correlation with LV ejection fraction in the LVSD group.

Conclusions—Production of aldosterone, angiotensin-converting enzyme, and BNP are activated in failing human ventricle in proportion to severity. (Circulation. 2001;103:72-77.)

Key Words: aldosterone ■ angiotensin-converting enzyme ■ angiotensin ■ B-type natriuretic peptide ■ ventricles

Aldosterone plays an important role in the pathophysiology of heart failure.1-2 This substance promotes retention of sodium and loss of potassium, activates the sympathetic nervous system and myocardial and vascular fibrosis, and causes baroreceptor dysfunction.1-5 Traditionally, aldosterone has been thought to be produced solely by adrenal cortex in response to angiotensin II, making it an important component of the circulating renin-angiotensin-aldosterone system.1

Recently, some workers reported that aldosterone is produced also in extra-adrenal tissues, including heart and blood vessels, in animals.6-10 However, whether aldosterone is produced in human heart, particularly in failing heart, is unknown.

Coronary sinus drains blood from the heart as a whole, and the anterior interventricular vein (AIV), which lies in the anterior interventricular groove, drains blood from the anterior left ventricle (LV).11 Therefore, the difference in hormone level between AIV and aortic root reflects the level of hormone from the LV, and that between the coronary sinus and aortic root reflects hormone level from the whole heart.

By the use of this method, we showed that production of A-type (ANP), and B-type (BNP) natriuretic peptides was activated in the LV in patients with heart failure.12-15 We also showed by use of this method that angiotensin-converting enzyme (ACE) activity from LV is increased in patients with LV dysfunction (LVD).15

The present study was designed to examine whether aldosterone is produced in addition to ACE and BNP in hearts of patients with LVD. We measured plasma levels of aldosterone together with those of BNP and serum ACE.

Methods

Subjects

The study population consisted of 45 patients with LVD who underwent cardiac catheterization at our institution. Patients were divided into systolic dysfunction (20 patients; 14 men and 6 women, mean age, 56.0 ± 3.0 years; and age range, 33 to 72 years) and isolated diastolic dysfunction (25 patients; 16 men and 9 women, mean age, 58.0 ± 3.0 years; and age range, 20 to 83 years) groups. Underlying cardiac disorders were dilated cardiomyopathy in 12 patients, old myocardial infarction in 6, and hypertensive heart disease in 2 in the systolic dysfunction group and hypertrophic...
cardiomyopathy in 13, hypertensive heart disease in 5, mitral regurgitation in 5, and aortic valvular regurgitation in 2 in diastolic dysfunction group. We defined LV systolic dysfunction (LVSD) as LV ejection fraction (LVEF) <55% and isolated LV diastolic dysfunction (LVDD) as LV end-diastolic pressure (LVEDP) >12 mm Hg, with LVEF >55% in the present study. Patients with severe heart failure were excluded from the study. All medications, including ACE inhibitors, angiotensin II receptor antagonists, aldosterone receptor blockers, nitrates, Ca antagonists, adrenergic \(-\)antagonists, and diuretics, were withheld in all patients for \(4\) days before study.

The control group comprised \(23\) patients (\(15\) men and \(8\) women; mean age, \(60.0 \pm 3.0\) years; age range, \(23\) to \(78\) years) in whom diagnostic cardiac catheterization, including coronary angiography and left ventriculography, was performed. Twenty control patients had atypical chest pain with normal coronary angiograms, and \(3\) patients had stable angina pectoris. None had myocardial infarction, cardiac hypertrophy, other heart muscle diseases, electrolyte disturbance, renal impairment (serum creatinine \(>2.0\) mg/mL), or hypertension.

The study protocol was in agreement with the guidelines of the ethics committee at our institution, and written informed consent was obtained from each patient before study, including consent for withholding of medication.

**Cardiac Catheterization Study**

Cardiac catheterization was performed in the morning, with patients in a fasting state. Hemodynamic measurements, including pulmonary artery pressure, pulmonary capillary wedge pressure, right atrial pressure, and cardiac output, were performed with a Swan-Ganz catheter inserted into the femoral or subclavian vein. Cardiac output was determined by use of the thermodilution technique in triplicate. After right heart catheterization was completed, a \(6\)F Goodale-Lubin catheter was placed in the coronary sinus through a brachial vein. The catheter was then advanced into the AIV fluoroscopically by use of a guidewire.\(^{12-15}\) Position of catheter tip in AIV was confirmed by injection of contrast dye medium. Patients in whom at least the proximal half of the AIV was not visualized were excluded from study. A Judkins catheter was placed at the root of the aorta by way of the femoral artery. Blood was sampled within \(2\) minutes at the aortic root, AIV, and coronary sinus. Care was taken to draw blood samples slowly from the AIV. Initial parts of the sample, including those forcibly drawn, were discarded, because forcible drawing of blood from the AIV resulted in spurious levels of hormone, probably because backflow from the coronary sinus occurred and contaminated the AIV.\(^{11,12}\)

Systemic arterial pressure, heart rate, and LVEDP were measured, and coronary arteriography and left ventriculography were performed. LVEF was determined by left ventriculograms.

**Hormonal Analysis**

Plasma levels of aldosterone were measured in duplicate by commercially available radioimmunoassay kits (SPAC-S aldosterone kit; Dainabot Inc.).\(^{16}\) The minimal detectable quantity of aldosterone was \(25\) pg/mL. Intra-assay and interassay coefficients of variation were 4.7% and 4.5%, respectively. Serum ACE activity was measured in duplicate by colorimetry using commercially available kits (ACE color; Fujirebio Inc.)\(^{17}\) Intra-assay and interassay coefficients of variation by this method were 6.7% and 8.3%, respectively. Plasma levels of BNP were measured with a specific immunoradiometric assay for human BNP (Shionoria BNP kit; Shionogi) as reported previously.\(^{18}\) Minimal detectable quantity of human BNP is \(4\) pg/mL. Intra-assay and interassay coefficients of variation were 5.3% and 5.9%, respectively.

**Statistical Analysis**

All values are expressed as mean \(\pm\)SE. Statistical significance was defined as a probability value <0.05. Unpaired \(t\) test or 1-way ANOVA was used to analyze results of hemodynamic or hormonal measurements.\(^{19}\) Hormonal levels at the aortic root, AIV, and coronary sinus within each group were compared with 2-way ANOVA with repeated measurements followed by Scheffé’s test. Correlation of plasma levels of aldosterone and BNP as well as serum levels of ACE activity with hemodynamic parameters were examined by use of linear regression analysis.

**Results**

**Cardiac Catheterization Data**

The Table shows cardiac catheterization data for patients with LVSD and LVDD and control subjects. LVEDP, LV end-diastolic volume index, and LV end-systolic volume index were significantly increased, and mean arterial blood pressure, cardiac index, and LVEF were significantly decreased in patients with LVSD versus control subjects. Mean arterial blood pressure and LVEDP were significantly higher in the LVDD group than in the control group.

**Comparison of Hormonal Levels**

Figure 1 shows plasma aldosterone levels, serum ACE activity, and plasma BNP levels in aortic root, AIV, and coronary sinus in LVSD and LVDD groups versus control subjects.

In the LVSD group, plasma levels of aldosterone were significantly higher in AIV and coronary sinus than aortic root (\(98 \pm 10\) versus \(72 \pm 9\) pg/mL, \(P <0.001\), and \(97 \pm 11\) versus \(72 \pm 9\) pg/mL, \(P <0.001\), respectively), whereas no difference existed in levels between AIV and coronary sinus. Plasma aldosterone levels in the LVSD group were significantly higher in AIV (\(98 \pm 10\) versus \(58 \pm 6\) pg/mL; \(P <0.001\) and coronary sinus (\(97 \pm 11\) versus \(62 \pm 7\) pg/mL; \(P <0.01\)) than in the control group.

In the LVDD group, plasma levels of aldosterone were significantly higher in AIV and coronary sinus than aortic root (\(87 \pm 10\) versus \(71 \pm 9\) pg/mL, \(P <0.01\), and \(84 \pm 10\) versus \(71 \pm 9\) pg/mL, \(P <0.01\), respectively), whereas levels were not different between AIV and coronary sinus. Plasma levels of aldosterone in the LVDD group were significantly higher in AIV (\(87 \pm 10\) versus \(58 \pm 6\) pg/mL; \(P <0.01\)) and coronary sinus (\(84 \pm 10\) versus \(62 \pm 7\) pg/mL; \(P <0.01\)) than in controls. On the other hand, no significant differences were seen in

<table>
<thead>
<tr>
<th>Cardiac Catheterization Data</th>
<th>Heart Rate, bpm</th>
<th>Mean BP, mm Hg</th>
<th>CI, L (-) min (^{-1}) (\cdot) m (^{-2})</th>
<th>LVEF, %</th>
<th>LVEDP, mm Hg</th>
<th>LVEDVI, mL (\cdot) m (^{-2})</th>
<th>LVESVI, mL (\cdot) m (^{-2})</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control group (n=23)</td>
<td>69 (\pm) 2</td>
<td>94 (\pm) 5</td>
<td>3.0 (\pm) 0.6</td>
<td>78 (\pm) 1</td>
<td>8.4 (\pm) 0.5</td>
<td>74 (\pm) 2</td>
<td>19 (\pm) 1</td>
</tr>
<tr>
<td>LVSD group (n=20)</td>
<td>73 (\pm) 3</td>
<td>87 (\pm) 5</td>
<td>2.5 (\pm) 0.1</td>
<td>43 (\pm) 2</td>
<td>18 (\pm) 2</td>
<td>101 (\pm) 6</td>
<td>57 (\pm) 5</td>
</tr>
<tr>
<td>(P)</td>
<td>0.421</td>
<td>0.038</td>
<td>0.006</td>
<td>&lt;0.001</td>
<td>&lt;0.001</td>
<td>&lt;0.001</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>LVDD group (n=25)</td>
<td>69 (\pm) 2</td>
<td>102 (\pm) 5</td>
<td>2.8 (\pm) 0.1</td>
<td>76 (\pm) 1</td>
<td>15 (\pm) 1</td>
<td>78 (\pm) 5</td>
<td>28 (\pm) 5</td>
</tr>
<tr>
<td>(P)</td>
<td>0.997</td>
<td>0.034</td>
<td>0.106</td>
<td>0.408</td>
<td>&lt;0.001</td>
<td>0.403</td>
<td>0.098</td>
</tr>
</tbody>
</table>

CI indicates cardiac index; LDEDVI, LV end-diastolic volume index; and LVESVI, LV end-systolic volume index.
plasma aldosterone levels among aortic root, AIV, and coronary sinus (61±7, 58±6, and 62±7 pg/mL, respectively) in the control group.

In the LVSD group, serum ACE activity was significantly higher in AIV and coronary sinus than in aortic root (13.2±0.7 versus 11.9±0.6 pg/mL, P<0.001, and 13.1±0.7 versus 11.9±0.6 pg/mL, P<0.001, respectively), whereas levels were not significantly different between AIV and coronary sinus. ACE activity levels were significantly higher in AIV (13.2±0.7 versus 10.6±0.7 pg/mL; P<0.01) and coronary sinus (13.1±0.7 versus 10.8±0.8 pg/mL; P<0.01) compared with controls. In the LVDD group, serum ACE activities were not significantly different for every sampling point, and levels were not different from those of the control group. No significant difference existed in ACE activity among the aortic root, AIV, and coronary sinus (10.7±0.7, 10.6±0.7, and 10.8±0.8 pg/mL, respectively) in the control group.

In the LVSD group, plasma BNP levels were significantly higher in AIV and coronary sinus than aortic root (673±121 versus 232±58 pg/mL, P<0.001, and 694±127 versus 232±58 pg/mL, P<0.001), whereas levels were not different between AIV and the coronary sinus. Plasma BNP levels in the LVSD group were significantly higher in aortic root (232±58 versus 20±2 pg/mL, P<0.001), AIV (673±121 versus 74±13 pg/mL, P<0.001), and coronary sinus (694±127 versus 79±15 pg/mL, P<0.001) versus the control group. In the LVDD group, plasma levels of BNP were significantly higher in AIV and coronary sinus than aortic root (215±72 versus 78±9 pg/mL, P<0.001, and 200±65 versus 78±9 pg/mL, P<0.001, respectively), with levels not different between AIV and coronary sinus. Plasma levels of BNP in the LVDD group tended to be higher at every sampling point compared with those of the control group. In the control group, plasma levels of BNP were significantly higher in AIV and coronary sinus than aortic root (74±13 versus 20±2 pg/mL, P<0.001, and 79±15 versus 20±2 pg/mL, P<0.001), whereas levels were not different between AIV and coronary sinus.

**Correlation of Cardiac Hormones With Hemodynamic Parameters**

The difference in plasma aldosterone levels between the AIV and the aortic root (ΔAldo [AIV-Ao]), where Ao indicates
The difference in serum ACE activity (ΔACE [AIV-Ao]) had a significant negative correlation with LVEF and a significant positive correlation with LVEDP in the LVSD group (Figure 2, center) but no correlation with either LVEF or LVEDP in the LVDD group (Figure 3, center).

The difference in plasma BNP levels (ΔBNP [AIV-Ao]) had a significant negative correlation with LVEF and a significant positive correlation with LVEDP in the LVSD group (Figure 2, right). In the LVDD group, ΔBNP (AIV-Ao) had only a weakly significant correlation with LVEDP (Figure 3, right).

Correlations Among Cardiac Aldosterone, ACE Activity, and BNP
In the LVSD group, ΔAldo (AIV-Ao) had a significant positive correlation with ΔACE (AIV-Ao) but no correlation with ΔBNP (AIV-Ao) (Figure 4A). A significant positive correlation occurred between ΔACE (AIV-Ao) and ΔBNP (AIV-Ao) (Figure 4A).

In the LVDD group, ΔAldo (AIV-Ao) had a significant positive correlation with ΔACE (AIV-Ao), whereas it had no correlation with ΔBNP (AIV-Ao) (Figure 4B) as in the LVSD group. A significant positive correlation occurred between ΔACE (AIV-Ao) and ΔBNP (AIV-Ao) (Figure 4B).

Discussion
In the present study, plasma level of aldosterone was increased significantly between AIV and aortic root in failing human ventricle, particularly when failure was caused by systolic dysfunction. No significant difference occurred in plasma aldosterone level in controls, which indicates that aldosterone production is activated in failing human LV but not in normal human heart. However, the difference in aldosterone level between AIV and aortic root was modest, which suggests that the function of cardiac aldosterone may be mainly paracrine or autocrine. Aldosterone effects are mediated by binding of the hormone to its specific receptor, the mineralocorticoid receptor (MR), and both MR and MR-protecting 11β-hydroxysteroid dehydrogenase (11-HSD) have been shown to be expressed in human heart.20,21

The present study also showed that levels of serum ACE activity were increased significantly between AIV and aortic root in failing hearts with systolic dysfunction but not significantly in failing hearts with diastolic dysfunction or control hearts. These results are in agreement with those of our previous studies15 and indicate that ACE production also was activated in failing ventricles of humans with systolic dysfunction. However, the absolute differences in ACE activity between AIV and aortic root were small, which indicates that the biological function of cardiac ACE is mainly autocrine, paracrine, or both.

On the other hand, differences in BNP levels between AIV and aortic root were highly significant in failing ventricles with systolic dysfunction as well as failing ventricles with diastolic dysfunction and control hearts. This indicates that the heart is the main source of BNP production and the endocrine organ for circulating BNP. These results are in agreement with those of our previous studies.13–15,22

The exact mechanism for induction of aldosterone into the failing heart is not clear. Aldosterone synthesis is mainly stimulated by angiotensin II, the active peptide of renin-
angiotensin system, but its production is also controlled by potassium, adrenocorticotropic hormone, and natriuretic peptides, including ANP and BNP.23–28 In the present study, levels of aldosterone had a highly significant positive correlation with levels of ACE activity in failing ventricles. This result suggests that increased activity of local ACE, causing conversion of angiotensin I to angiotensin II, may stimulate production of aldosterone in failing hearts in a paracrine or autocrine manner. Indeed, Silvestre and coworkers10 recently showed that cardiac aldosterone is activated in rat heart with myocardial infarction and that this is mediated primarily by cardiac angiotensin II. They further showed that aldosterone synthase mRNA was elevated in infarcted rat ventricles.10

Cardiac aldosterone, ACE, and BNP had significant positive correlations with LVEDP and significant negative correlations with LVEF in patients with LVSD. These results are in agreement with those of our previous studies13–15,22 and suggest that increased wall tension or stretch in the dilated ventricles may be a main stimulus for activation of cardiac aldosterone as well as ACE and BNP production in ventricles with systolic dysfunction. On the other hand, no significant correlation was found between cardiac hormones and LVEDP except cardiac BNP, which had a weak correlation with LVEDP in patients with isolated LVDD. The results indicate that systolic dysfunction is more important than diastolic dysfunction for activation of cardiac aldosterone as well as ACE and BNP in failing hearts.

No significant correlation existed between levels of aldosterone and BNP in failing ventricles. This result probably is due to the fact that aldosterone production is stimulated by angiotensin II but suppressed by natriuretic peptides (ANP and BNP) in failing hearts.23–28 We and others have shown that ANP and BNP directly suppress aldosterone production.25–28

Clinical Implications
Aldosterone was originally thought to be important in the pathophysiology of heart failure only because of its ability to increase sodium retention and potassium loss. However, Weber et al4 and Young et al5 have shown that aldosterone promotes myocardial and vascular fibrosis independent of the hemodynamic effects. Aldosterone also causes direct vascular damage and baroreceptor dysfunction and prevents uptake of norepinephrine by the myocardium.4,29,30 Recently, Pitt and coworkers31 showed that blocking aldosterone with a low dose of spironolactone substantially reduced the risks of morbidity and mortality among patients with severe heart failure (RALES trial). Efficacy of an aldosterone blockade in their trial does not appear to be due entirely to prevention of sodium retention or potassium loss.

The present study shows that aldosterone production is activated in failing ventricles of humans. Concentration of aldosterone within the heart is reported to greatly exceed circulating concentrations.9 Thus, cardiac aldosterone may play an important role in the pathophysiology of heart failure, and aldosterone receptor antagonists may ameliorate heart failure by blocking the action of locally produced aldosterone in failing heart.

Conclusions
We conclude that the production of aldosterone, in addition to ACE and BNP, is activated in failing human ventricles in proportion to severity, particularly of systolic dysfunction.

References


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