Inducibility of Atrial Fibrillation Caused by Acute Increase of Atrial Pressure in Rat Diseased Heart With Chronic Atrial Dilation

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SUMMARY

Although acute atrial dilation facilitates the induction of atrial fibrillation (AF) in the normal heart, little is known about whether the induction of AF due to acute atrial dilation increases in the diseased heart. To clarify this, we compared the inducibility of AF by an acute increase of atrial pressure with and without chronic atrial dilation induced by volume- and pressure-overload in rats. Eight weeks after creating abdominal aortocaval shunt and aortic constriction rats (LVH rats, $n = 8$) or sham rats ($n = 8$), the hearts were perfused in Langendorff's manner. Right atrial (RA) pressure was increased from 2 cm H₂O to 10 cm H₂O by the height of the reservoir. Inducibility of AF was evaluated by 5 times burst pacing from the right atrium, and mean cycle length of AF (CL) and the atrial effective refractory period (AERP) were also measured. The inducibility of AF increased from $5 \pm 3\%$ at 2 cm H₂O to $50 \pm 5\%$ at 10 cm H₂O RA pressure in sham rats ($P < 0.01$), but not in LVH rats ($20 \pm 7\%$ to $25 \pm 6\%$, NS). Mean CL and AERP in LVH rats were longer than those in sham rats. In addition, the AERP decreased with an increase in RA pressure from 2 cm H₂O to 10 cm H₂O in sham rats, but not in LVH rats. The inducibility of AF caused by an acute increase of RA pressure did not increase in the diseased heart, suggesting that electrophysiological remodeling may play a role, at least in a compensated state, for the prevention of AF due to an acute increase of atrial pressure. (Int Heart J 2012; 53: 257-260)

Key words: Acute atrial dilation, Atrial fibrillation, Chronic atrial dilation, Abdominal aortocaval shunt, Aortic constriction

Chronic atrial dilation in the diseased heart is associated with an increased incidence of atrial fibrillation (AF). Atrial electrical and structural remodelings, ie, intra-atrial conduction disturbances, repolarization abnormality, and interstitial fibrosis, have been demonstrated to be the mechanisms by which atrial dilation leads to a substrate of AF in clinical and animal studies.^{1,2)}

On the other hand, acute atrial dilation in the normal heart has also been shown to facilitate the induction of AF.³⁾ This vulnerability of AF has been attributed to conduction slowing and reduction in the atrial effective refractory period (AERP) by activation of stretch-activated ion channels. However, it remains unclear whether the induction of AF due to acute atrial dilation increases in the diseased heart with chronic atrial dilation. We hypothesized that atrial electrical and structural remodelings in the diseased heart may alter the atrial electrophysiological properties due to acute atrial dilation, resulting in the induction of AF.

The aim of this study was to clarify whether the inducibility of AF caused by an acute increase of atrial pressure is altered in rats with chronic atrial dilation.

METHODS

A total of 16 male Wister rats aged 8 weeks were used in

this study. All animal experiments were conducted in accordance with the guidelines of the Fukushima Medical University Animal Research Committee. The research protocol was approved by the institutional review board.

Animal model: Left ventricular (LV) hypertrophy was induced by volume- and pressure-overload (LVH rats, $n = 8$). Briefly, after anesthesia with sodium pentobarbital (50 mg/kg intraperitoneally), an abdominal aorta and inferior vena cava shunt was made according to a previous method.⁴⁾ Next, surgical aortic constriction at the supra-renal arterial level was performed.⁵⁾ Identical operations were performed on control rats without making the fistulas and aortic constriction (sham rats, $n = 8$).

Echocardiographic assessment: Eight weeks after the operations, LV function [LV end-diastolic diameter (LVEDD), LV end-systolic diameter (LVESD), interventricular septal thickness (IVST), and LV ejection fraction by the Pombo method, with the following formula] was assessed by M-mode echocardiography in the long-axial view, and right atrial (RA) diameter (RAD) was measured in the apical 4-chamber view.

LV ejection fraction (%) = $100 \times [(LVEDD^3 - LVESD^3) / LVEDD^3]$

Langendorff-perfused heart preparation: Following echocardiographic examination, the chest was opened by a midline incision and a cannula was inserted retrogradely into the ascending aorta for retrograde perfusion of the heart as previously described.^{6,7)} The heart was then perfused with oxygenated

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modified Krebs-Henseleit solution containing (in mmol/L): NaCl 118, KCl 4.7, CaCl_2 2.5, KH_2PO_4 1.2, MgSO_4 1.2, NaHCO_3 2.5, glucose 5, and sodium pyruvate 5. The perfusate was continuously bubbled with 95% O_2 -5% CO_2 and it was maintained at 37°C. The coronary perfusion pressure was maintained at 90 mmHg and LV end-diastolic pressure was adjusted to 5 mmHg. The pulmonary artery and superior caval vein were ligated, and RA pressure was allowed to vary by the height of the reservoir connecting to the inferior caval vein cannula. One bipolar silver electrode was attached to the high right atrium for atrial pacing, and the other bipolar electrode was fixed at the low right atrium for recording an epicardial electrogram.

Experimental protocol: After the perfusion system was set up at 2 cm H_2O RA pressure, burst RA stimulation with 1200 beats/minute for 15 seconds was performed for induction of AF. The induced rhythm was defined as AF when a fast atrial irregular rhythm was maintained for more than 5 seconds. Inducibility of AF was defined as the induction rate by 5 times burst pacing. When an AF was induced, the cycle length of the AF was measured and averaged for 5 seconds from the start of the AF. Thereafter, the AERP was measured by the single early premature stimulus method using 8 regularly paced beats at cycle length = 200 msec. Ten minutes after the end of these experiments, the same protocol was repeated at 10 cm H_2O of RA pressure.

Statistical analyses: The data are presented as the mean $\pm$ SEM. The unpaired Student *t*-test was used for comparisons between rats with the LVH and the sham group. Comparisons of the inducibility of AF, the mean cycle length of AF, and AERP before and after increased RA pressure were performed using the paired Student *t*-test. Differences were considered significant at a level of $P < 0.05$.

RESULTS

Echocardiographic findings: The table shows the echocardiographic findings in LVH and sham rats. LVEDD, LVESD, IVST, and RAD were significantly increased in LVH rats ($P < 0.05$), indicating the development of LV volume- and pressure-overload induced LV hypertrophy. The LV ejection fraction was significantly lower in LVH rats ($P < 0.05$). However, LVH rats showed no signs of congestive heart failure.

Effect of acute increase of atrial pressure on AF inducibility: The inducibility of AF is shown in Figure 1. During a 2 cm H_2O of RA pressure, the inducibility of AF was significantly higher in LVH rats than in sham rats ($5 \pm 3\%$ versus $20 \pm 7\%$,

Figure 1. The inducibility of atrial fibrillation (AF) at each right atrial (RA) pressure (2 cm H_2O and 10 cm H_2O) in sham rats ($n = 8$) and LVH rats ($n = 8$).

Figure 2. Mean cycle length of AF at each RA pressure (2 cm H_2O and 10 cm H_2O) in sham rats ($n = 8$) and LVH rats ($n = 8$).

Figure 3. RA effective refractory period (AERP) at each RA pressure (2 cm H_2O and 10 cm H_2O) in sham rats ($n = 8$) and LVH rats ($n = 8$).

Table. Echocardiographic Findings

	Sham ($n = 8$)	LVH ($n = 8$)	<i>P</i>
LVEDD (cm)	0.54 $\pm$ 0.01	0.65 $\pm$ 0.01	< 0.05
LVESD (cm)	0.21 $\pm$ 0.02	0.37 $\pm$ 0.02	< 0.05
IVST (cm)	0.14 $\pm$ 0.01	0.22 $\pm$ 0.01	< 0.05
RAD (cm)	0.29 $\pm$ 0.01	0.39 $\pm$ 0.02	< 0.05
LVEF (%)	0.86 $\pm$ 0.02	0.80 $\pm$ 0.02	< 0.05

LVEDD indicates left ventricular end-diastolic diameter; LVESD, left ventricular end-systolic diameter; IVST, interventricular septal thickness; RAD, right atrial diameter; and LVEF, left ventricular ejection fraction.

$P < 0.05$). After the RA pressure increased to 10 cm H₂O, the inducibility of AF increased significantly in the sham rats ($5 \pm 3\%$ to $50 \pm 5\%$, $P < 0.01$), but not in the LVH rats ($20 \pm 7\%$ to $25 \pm 6\%$, NS).

Mean cycle length of AF: The mean cycle lengths of AF in both groups of rats are shown in Figure 2. In LVH rats, AF mean cycle length was longer than that in the sham rats at each RA pressure (83 ± 6 msec in sham rats, 110 ± 8 msec in LVH rats at 2 cm H₂O, $P < 0.05$, 73 ± 7 msec in sham rats, 102 ± 6 msec in LVH rats at 10 cm H₂O, $P < 0.05$). However, mean cycle length was not different with an increase in RA pressure in both groups of rats.

Atrial refractoriness: The AERPs in both groups of rats are shown in Figure 3. In the LVH rats, AERP was longer than that in the sham rats at each RA pressure (20 ± 4 msec in sham rats, 29 ± 4 msec in LVH rats at 2 cm H₂O, $P < 0.05$, 15 ± 4 msec in sham rats, 28 ± 3 msec in LVH rats at 10 cm H₂O, $P < 0.05$). In addition, AERP shortened with an increase in RA pressure in the sham rats (20 ± 4 msec to 15 ± 4 msec, $P < 0.05$), but not in the LVH rats (29 ± 4 msec to 28 ± 3 msec, NS).

DISCUSSION

The main findings obtained with chronic atrial dilation induced by volume- and pressure-overload LVH hearts are as follows. First, the induction of AF due to an acute increase of RA pressure increased in the sham rats, but not in the LVH rats. Second, AERP shortened with an increase in RA pressure in the sham rats, but not in the LVH rats. Third, AERP in the LVH rats was longer than that in the sham rats regardless of the degree of RA pressure. These results suggest that electrophysiological remodeling in LVH may play a role, at least in a compensated state, for the prevention of AF induction via rapid mechanical change.

Various animal studies evaluating the effects of acute atrial dilation on AF have used the normal heart. In this study, the induction of AF due to an acute increase of RA pressure increased in sham rats, which is consistent with a previous report.³⁾ It is generally accepted that a reduction in AERP by activation of stretch-activated ion channels appears to be of major importance in providing a substrate for AF due to acute atrial dilation in the normal heart.³⁾ In the present study, AERP also decreased with an increase in RA pressure in the sham rats.

On the contrary, the induction of AF caused by an acute increase of RA pressure in LVH rats did not increase in this study. In addition, a reduction in AERP due to an acute increase of RA pressure was not observed in LVH rats. In light of recent work on atrial stretch, the effects of chronic atrial dilation on atrial electrophysiology have become a primary focus. Verheule, *et al* have demonstrated that chronic atrial dilation due to mitral regurgitation in dogs increases in AERP and its dispersion contributes to the vulnerability of AF.²⁾ Morton, *et al* have shown that chronic RA stretch due to an atrial septal defect causes electrical remodeling with an increase in AERP in a clinical investigation.¹⁾ In the present study, AERP in the LVH rats was longer than that in the sham rats regardless of the degree of RA pressure, findings that are consistent with previous observations of chronic atrial dilation. Therefore, this is an interesting study for estimating the influences of an acute

increase of atrial pressure on the vulnerability leading to AF in the diseased heart with chronic atrial dilation. The mechanism of these changes in atrial electrophysiological properties from the normal to the LVH heart remains to be clarified. Satoh and Zipes previously showed that AERP in the thin RA free wall differs from the AERP of the thick crista terminalis, and the AERP in the thin wall increases more than that in the thick wall during acute atrial stretch in open chest dogs.⁸⁾ Taking these results into consideration, our data suggests that shortening in AERP differences by acute stretch may be one of the mechanisms for changing the inducibility of AF in LVH hearts.

We did not assess the atrial conduction velocity in this study, which is a relevant issue. According to the multiple wavelet theory,⁹⁾ the wavelength of the reentrant wave equals the product of AERP and conduction velocity. In the present study, mean cycle length of AF in the LVH rats was longer than in the sham rats at each RA pressure. Thus, we cannot rule out the possibility that the conduction velocity in the right atrium may be changed by an acute increase of RA pressure in the LVH heart. In addition, we did not evaluate ionic remodeling due to atrial dilation. A variety of ionic abnormalities, ie, decreased I_{to} and I_{Ca} and increased I_{K1} , have been reported in atrial myocytes from an experimental diseased heart.¹⁰⁾ Accordingly, ionic remodeling could be involved in these mechanisms.

An important limitation of the present study is that we measured variables under an open chest and anesthetized conditions in the rat model. Since the degree of acute atrial dilation using a Langendorff preparation may not mimic the degree of dilation observed in human atrium, we should take into consideration this was a conscious closed chest model when extrapolating the results of this study to a clinical setting.

In conclusion, this study provides evidence that an acute increase of atrial pressure alters the vulnerability of a diseased heart, which leads to AF. Our results suggest that electrophysiological remodeling in chronic atrial dilation may play a role, at least in a compensated state, for the prevention of AF due to an acute increase of atrial pressure.

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