Hemodynamic changes, atrial remodeling and deposits of atrial natriuretic peptide in patients with atrial fibrillation are associated with clinical outcome of the maze operation

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Hemodynamic changes, atrial remodeling and deposits of atrial natriuretic peptide in patients with atrial fibrillation are associated with clinical outcome of the maze operation

Directed by Professor Hui-Nam Pak

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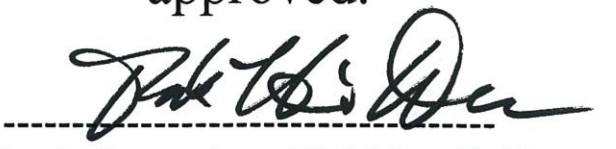
This certifies that the Doctoral Dissertation of Junbeom Park is approved.



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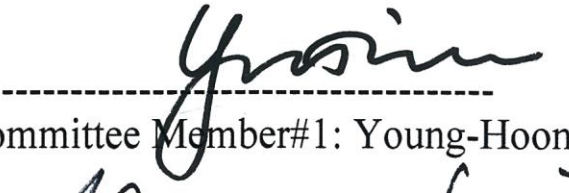

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ABSTRACT

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Background: Isolated atrial amyloid (IAA) deposition in the left atrium (LA) is associated with atrial fibrillation (AF), and the maintenance of AF results in remodeling of the atrial substrate. However, there is no direct evidence of an association between atrial deposition of atrial natriuretic peptide (ANP) and atrial amyloids in patients with AF, and the clinical implications of this after the maze operation have not been investigated.

Methods: Twenty left atrial appendage (LAA) tissues were acquired during mitral valve surgery and the maze operation. All tissues were stained with Congo red under polarized light to detect amyloids, and the area of amyloid deposits was quantified using Image Pro. ANP level was evaluated by Western blotting. And we quantified the protein expression of endocardial functions (endocardial nitric oxide [eNOS], phosphorylated eNOS [Phospho-eNOS]), intercellular adhesion molecule-1 (ICAM-1) and thrombotic factors (von Willebrand factor [vWF], a disintegrin and metalloproteinase with a thrombospondin type 1 motif, member 13 [ADAMTS13]) by Western blotting. Hemodynamic factors (pulmonary arterial pressure, PAP) were measured prior to the operation were analyzed according to the expression of ANP.

Results: 1. ANP+ patients had a lower systolic PAP (p=0.04) than ANP- patients, and the presence of amyloid deposits was also negatively correlated (r=-0.6, p=0.008) with systolic PAP. 2. The expression level of eNOS (p=0.043),

phospho-eNOS (p=0.013), vWF (p=0.031) and ICAM-1 (p=0.013) were lower in tissue with ANP expression (ANP+) than ANP- tissues. 3. More amyloid deposits were detected in ANP+ tissue than ANP- tissue (p=0.002). 4. Recurrence of AF after the maze operation was higher in ANP+ patients than ANP- patients (p=0.033), and all recurrence occurred within 150 days. ANP+ status was significantly associated with the recurrence of AF after maze (p=0.005) even after adjusting for age, sex, and LA volume for a mean of 362.3±473.8 days (maximum 1,565 days).

Conclusions: Atrial deposits of ANP showed a negative correlation with PAP, and reflected the presence of more atrial amyloid deposits. Deposition of ANP was strongly associated with decreased endocardial function and expression of intercellular adhesion molecule. Finally, expression of ANP was significantly associated with AF recurrence after the maze operation.

Key words: atrial natriuretic peptide, amyloid, endocardial nitric oxide, atrial fibrillation, maze

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I. INTRODUCTION

The spectrum of histological atrial alterations in patients with chronic persistent atrial fibrillation (AF) has not been completely elucidated. The structural basis of short- and long-term electrical remodeling has been described in different experimental models; although remodeling can result from AF, in the course of time, it can itself exacerbate the persistence and recurrence of AF.[1] In a previous study, we showed that the smooth muscle layer was closely related to the existence of AF, degree of atrial remodeling, and fibrosis in patients who underwent open heart surgery.[2] Increased atrial fibrosis has been reported in patients with AF, and these patients also have an activated angiotensin system.[3,4] Another structural change in the heart commonly observed with increasing age is amyloidosis, which is similar to atrial fibrosis. Recently, atrial amyloidosis has been shown to play an important role in atrial histological remodeling that occurs in patients with persistent atrial fibrillation.[5] The heart is most commonly affected by a strictly localized or organ-limited variant of atrial amyloidosis called isolated atrial amyloidosis (IAA).[6,7] IAA affects atrial conduction and increases the risk of AF, and is associated with increased synthesis and secretion of atrial natriuretic peptide (ANP).[8] The fibril protein deposited in IAA is ANP, which is a peptide hormone synthesized and secreted predominantly by atrial cardiomyocytes.[9,10] However, previous studies only showed an association

between histological characteristics and incidence of AF; clinical progression and outcomes were not reported. In addition, most studies related to AF included tissues from patients with mixed valve disease, which may obscure atrial remodeling by AF. We therefore focused on acquiring LA appendage tissue from patients with persistent AF and mitral valve disease during open heart surgery. We analyzed left atrial tissue changes and the clinical implications of these changes after maze surgery to treat AF.

II. MATERIALS AND METHODS

Study population

The study protocol adhered to the Declaration of Helsinki and was approved by the Institutional Review Board of Yonsei University Health System. All patients provided written informed consent. The study enrolled 20 patients who underwent mitral valve surgery (14 mitral valve replacements and 6 repairs) and the maze operation between December 2009 and April 2014 at Severance Hospital. All patients were diagnosed with persistent AF by documented EKG monitoring for more than 7 days. Thirteen patients had mitral regurgitation, and 7 had mitral stenosis. Full wall-thickness specimens were excised from the LA appendage. All myocardial specimens were fixed in 70% alcohol and 4% formalin solution-immediately after excision. Our exclusion criteria were as follows: 1) specimens with thickness less than that of the full wall; 2) follow-up loss after surgery of more than 12 months; 3) presence of aortic valve disease; 4) congenital cardiac disease; 5) taking anti-arrhythmic drugs (Class Ic or III); or 6) cardioversion performed.

Maze operation and measurements of intracardiac hemodynamics

All patients were prepared for mitral valve surgery and maze operation in overnight fasting status. For continuous cardiac output monitoring,

thermodilutional pulmonary artery catheter (Swan-Ganz, CCOmbo, Baxter Healthcare Co., Irvine, CA, USA) was inserted via the right internal jugular vein under local anesthesia. After that, anesthesia was induced with midazolam 0.05 mg/kg, sufentanil 1.5 μg/kg and rocuronium 50 mg, and maintained with continuous infusion of sufentanil 0.5 μg · kg–1 · h–1, vecuronium 8–10 mg/h and low dose of sevoflurane in oxygen (40–60%) with air. After vital sign was stabilized during monitoring of pressure curve, central venous pressure (CVP; from right atrium) and pulmonary arterial pressure (PAP; systolic, diastolic period) were measured. Cardiac ouput and cardiac index were measured by thermodilution method. All maze operations were performed by modified Cox III with cryoablation combined with LA appendage resection.

Tissue preparation and immunohistochemistry

Multiple 5-m-thick serial sections were used. Immunohistochemical staining was performed using an avidin-biotin peroxidase system (Dako). Paraffin-embedded tissue sections were deparaffinized, then washed with phosphate-buffered saline (PBS). A hydrogen peroxidase block (0.3% H2O2 in PBS) was placed on sections for 10 min, and slides were then washed in PBS. Slides were then incubated with primary antibodies for 90 min at room temperature (approx. 25℃). Antibodies against atrial natriuretic peptide (ANP, 5 g/ml, rabbit antihuman polyclonal; Abcam) were used to detect amyloid deposits (Figure 1-A). After incubation, slides were washed in PBS, and the appropriate secondary antibody (Dako) was placed on the sections for 30 min. The sections were again washed in PBS. Substrate-chromogen solution was applied for 5 min for DAB staining. Sections were then rinsed under running tap water for 5 min. Hematoxylin was added for 1 min followed by rinsing under running tap water. Specimens were then dehydrated in alcohol, mounted, and examined with light microscopy.

Quantification of fibrosis and amyloid deposits

The presence of amyloid deposits was demonstrated by the appearance of green birefringence from Congo red staining under polarized light (Figure 1-C, D). Sirius red staining was used to determine the presence and degree of fibrosis (Figure 1-B).

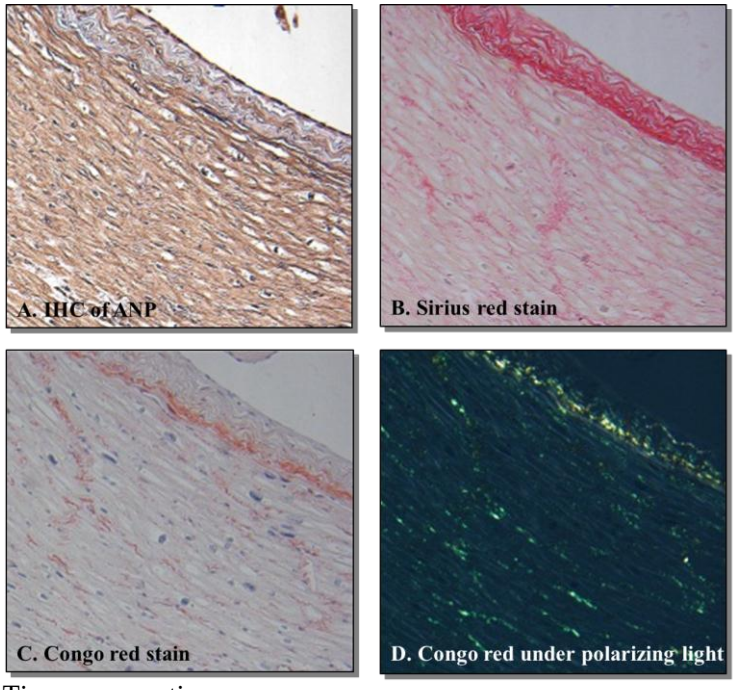
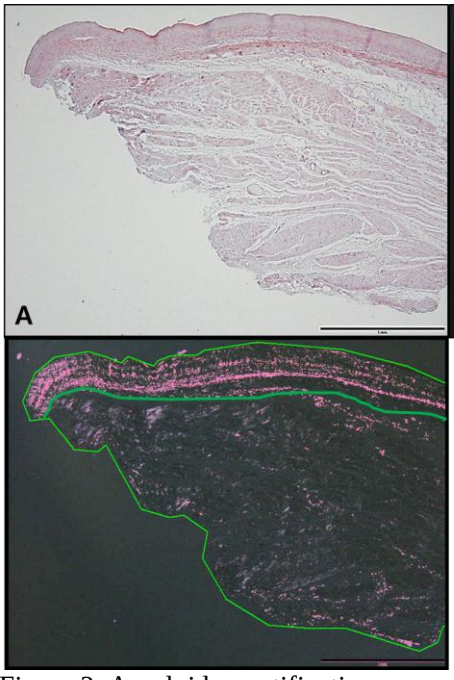


Figure 1. Tissue preparation

All tissues were stained with immunohistochemistry on ANP (A), Congo red (C) and viewed under polarizing light (D) for amyloid detection, and stained with Sirius red stain (B) to detect fibrosis

Virtual microscopic images were used to quantify the degree of fibrosis and amyloid deposition, and a single investigator who was blinded to the clinical information performed measurements. Fibrosis was quantified as the percent (%) area with fibrosis relative to the entire tissue section using Image Pro Plus

6.0 (Media Cybernetics, Silver Spring, MD, USA). Amyloid deposits were quantified as the percent area (%) of amyloid deposition relative to the entire tissue area. Tissue was divided into the endocardium and myocardium, and the amyloid deposition area in each layer was calculated using Image Pro (Figure 2).



Amyloid deposit (%)	Total	Endocardium	Myocardium
N	20	20	20
Mean ± SD	6.2±4.8	14.7±11.4	4.7±4.1
Median	5.5	11.4	3.3

Figure 2. Amyloid quantification

A. Congo red stain, B. Congo red stain under polarizing light. C. Percent area (%) of amyloid deposition to the entire tissue area was calculated. Tissue was divided into the endocardium and myocardium, and amyloid deposition in each layer was quantified from virtual microscopic images. D. Amyloid deposits were usually located more at endocardium than myocardial layer.

Western blotting analysis of atrial tissue

Proteins from LA appendage tissue were separated by 10% SDS-PAGE and then transferred to PVDF membranes (Amersham). After blocking for 1 h at room

temperature with 10% skim milk in TBS with Tween 20 (TBS-T 0.1%, tween 20), membranes were incubated overnight at 4℃ with monoclonal antibodies against vWF, ADAMTS13, eNOs, phosphorylated eNOs, ADMA, ICAM, angiotensin II, and ANP in antibody dilution buffer (5% bovine serum albumin in TBS with Tween 20). Membranes were washed three times with TBS-T, then incubated with alkaline phosphatase-conjugated anti-IgG (1:5000 dilution in 10% skim milk in TBS with Tween 20) for 1 h at room temperature. Membranes were washed five times with TBS-T and detected by ECL solution (1:1 ratio of substrate solution:enhancer solution). Glyceraldehyde-3-phosphate dehydrogenase (GAPDH) was measured as an internal control.

Echocardiographic evaluation of the heart

All patients underwent trans-thoracic echocardiography (TTE; Sonos 5500, Philips Medical System, Andover, MA, USA or Vivid 7, GE Vingmed Ultrasound, Horten, Norway) prior to maze operation. Chamber size (LA volume index [LAVI], LA dimension, wall thickness of LV, and LV mass index [LVMI]), transmitral flow velocity (E wave, A wave), and tissue Doppler images of the mitral annular septal area (peak diastolic velocity [Em], peak systolic velocity [Sm]) were acquired according to the American Society of Echocardiography guidelines.[11,12]

Post-maze management and follow-up

Patients visited an outpatient clinic regularly at 1, 3, 6, and 12 months and then every 6 months or whenever symptoms occurred after the maze operation. All patients underwent electrocardiography (ECG) at every visit and 24- or 48-hour Holter recording and/or event recording at 3, 6, and every 6 months following the 2012 HRS/EHRA/ECAS Expert Consensus Statement guidelines.[13] However, Holter monitor or event monitor recordings were obtained whenever patients reported palpitations to evaluate possible recurrence of arrhythmia. We

defined recurrence of AF as any episode of AF or atrial tachycardia of at least 30 sec in duration. Any ECG documentation of AF recurrence after 3 months was diagnosed as clinical recurrence, and as early recurrence if this occurred within 3 months. Total recurrence was considered clinical or early recurrence.

Data analysis

Normally distributed continuous variables were expressed as means±standard deviations (SD). Statistical significance of comparisons was assessed using paired t-tests and $\chi 2$ tests. Clinical association between ANP and area of amyloid deposits was analyzed by correlation analysis. Kaplan-Meier and Cox regression analysis were used to analyze AF-free survival after the maze operation. Variance inflation factors (VIF)≥10 were considered to indicate co-linearity and were excluded from multivariate linear regression. A p-value <0.05 was considered statistically significant.

III. RESULTS

Baseline characteristics

A total of 20 patients (58.0±1.7 years, 55% male) underwent the maze operation combined with mitral valve surgery. Among these 20 patients, 12 (60%) patients experienced AF recurrence after surgery (55% early recurrence, 25% clinical recurrence) over a mean follow-up duration of 362.3±473.8 days, and the mean recurrence duration was 21.4±42.8 days (Table 1). There was no significant difference in clinical characteristics (age, gender, body surface area [BSA], body mass index [BMI], CHADS2 score or echocardiography, p>0.05) according to the expression of ANP, with the exception of AF recurrence (p<0.05). Patients who expressed ANP as determined by Western blot analysis (ANP+) showed a higher recurrence rate of AF (87.5% vs. 41.7%, p=0.03) than non ANP-expressing (ANP-) patients (Table 1).

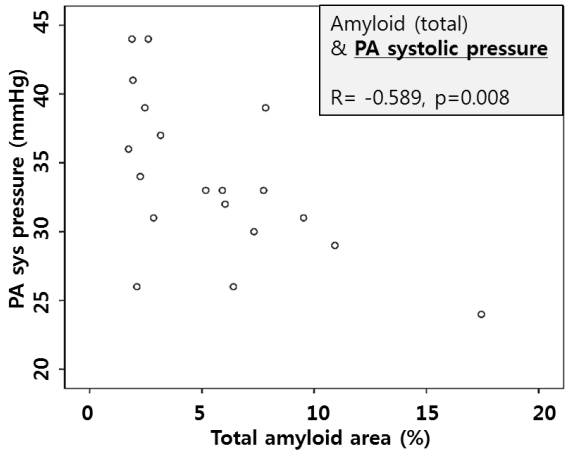
Table 1. Baseline characteristics of the study subjects

	All	ANP (-)	ANP (+)	p
N	20	12	8	
Age (years)	58.0±1.7	54.3±63.6	63.6±10.7	0.52
Male (n,%)	11 (55)	58	50	0.731
BSA (m^2)	1.6±0.2	1.7±0.2	1.6±0.2	0.358
BMI (kg/m^2)	23.3±3.4	23.8±3.4	22.5±3.6	0.415
CHADS2 score	0.9±1.1	0.6±0.9	1.3±1.4	0.207
CHA2DS2-VASc score	1.7±1.3	1.3±1.1	2.3±1.6	0.106
Echocardiography				
LA dimensions (mm)	57.6±7.8	59.8±8.1	54.1±6.5	0.112
LA volume index (ml/m^2)	90.1±25.8	96.7±56.9	80.2±21.8	0.168
EF (%)	65.1±7.8	64.8±6.4	65.5±10.0	0.857
E/E'	14.1±4.9	12.2±6.4	15.7±3.3	0.328
Clinical outcome				
Early recurrence (n,%)	11 (55)	5 (41.7)	6 (75.0)	0.158
Clinical recurrence (n,%)	4 (25)	2 (16.7)	2 (25.0)	0.384
Total recurrence (n,%)	12 (60)	5 (41.7)	7 (87.5)	0.03
Clinical recurrence duration (Days)	109.3±33.5	90	119.0±41.0	0.667
Recurrence duration (Days)	21.4±42.8	5.8±7.5	30.3±52.7	0.389
FU duration (Days)	362.3±473.8	507.6±569.1	146.0±103.9	0.053

Intracardiac hemodynamic change as mediator of atrial tissue remodeling

Figure 3 shows the association between PAP and atrial tissue change. As the deposits of atrial amyloid increased, systolic PAP was getting decreased (r=-0.589, p=0.008, Figure 3-A), and ANP (+) patients had lower systolic PAP (30.3±4.9 vs. 35.8±5.4mmHg, p=0.04) than ANP(-) patients (Figure 3-B).

A



B

	ANP (-)	ANP (+)	p
Cardiac output	4.7±2.5	3.4±0.7	0.238
Cardiac index	2.5±1.3	2.1±0.4	0.541
PA sys pressure (mmHg)	35.8±5.4	30.3±4.9	0.04
PA dia Pressure (mmHg)	19.3±3.0	17.9±3.6	0.382
PA mean pressure (mmHg)	25.8±4.4	23.3±5.0	0.264
CV pressure (mmHg)	10.8±3.0	9.9±2.0	0.442

Figure 3. Intracardiac hemodynamic change as mediators of atrial tissue remodeling. A. The negative correlation between amyloid deposit area and systolic pulmonary arterial pressure. B. ANP (+) tissue shows low systolic pulmonary arterial pressure

In direct comparison, the amount of ANP also showed a positive correlation with the area of amyloid deposition (Figure 4-A,B and C) and ANPs which was detected in immunohistochemistry was matched well with amyloid deposits.

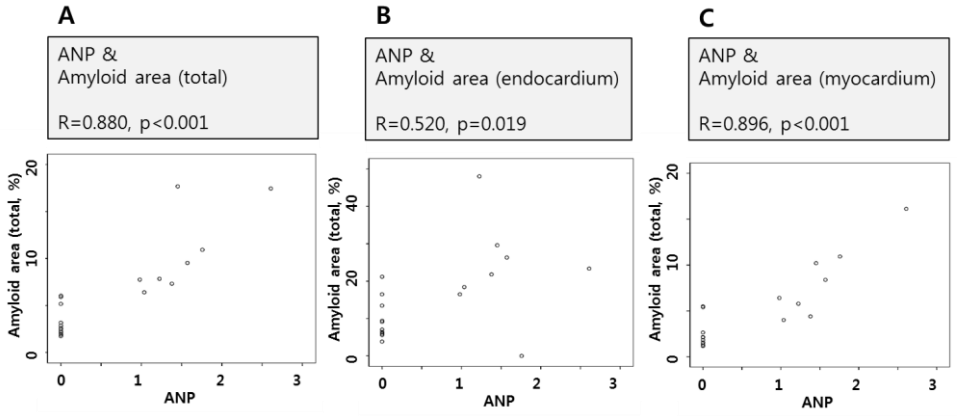


Figure 4. Positive correlation between ANP and area of amyloid deposits. ANP expression showed a positive correlation with amyloid deposition, regardless of layers (A,B,C).

And although the amyloid was deposited more in the endocardial layer (14.7±11.4%) than the myocardium (4.7±4.1%)(Figure 2-D), ANP+ tissue had more amyloid deposits than ANP- tissue regardless of cardiac layers (Table 2).

Table 2. Tissue characteristics according to the expression of ANP

	ANP (-)	ANP (+)	p
Congo red under polarized light			
Amyloid area (%, total)	3.2±1.6	10.6±4.5	0.002
Amyloid area (%, endocardium)	9.2±5.2	23.0±13.5	0.005
Amyloid area (%, myocardium)	2.3±1.5	8.3±4.1	0.004

Atrial tissue characteristics according to the expression of ANP

Table 3 and figure 5 show differences in atrial tissue according to the expression of ANP. ANP+ tissue was characterized by lower expression of endocardial nitric oxide synthase (eNOs; 0.1±0.2 vs. 0.4±0.4, p=0.43), phosphorylated

eNOs (pho-eNOs; 0.1±0.3 vs. 0.7±0.6, p=0.013) and asymmetric dimethylarginine (ADMA; 0.6±0.2 vs. 1.0±0.3, p=0.02) than ANP- tissue. Expression of intercellular adhesion molecule-1 (ICAM-; 0.6±0.2 vs. 1.0±0.3, p=0.013) and angiotensin II (0.6±0.1 vs. 1.0±0.3, p=0.003) was also lower in ANP+ tissue than ANP- tissue.

Table 3. Tissue characteristics according to the expression of ANP

	ANP (-)	ANP (+)	p
Western blot			
vWF	1.0±0.4	0.6±0.2	0.031
ADAMTS13	1.2±0.6	1.2±0.4	0.757
eNOs	0.4±0.4	0.1±0.2	0.043
Phospho eNOs	0.7±0.6	0.1±0.3	0.013
ADMA	1.0±0.3	0.6±0.2	0.02
ICAM	1.0±0.3	0.6±0.2	0.013
Angiotensin II	1.0±0.3	0.6±0.1	0.003

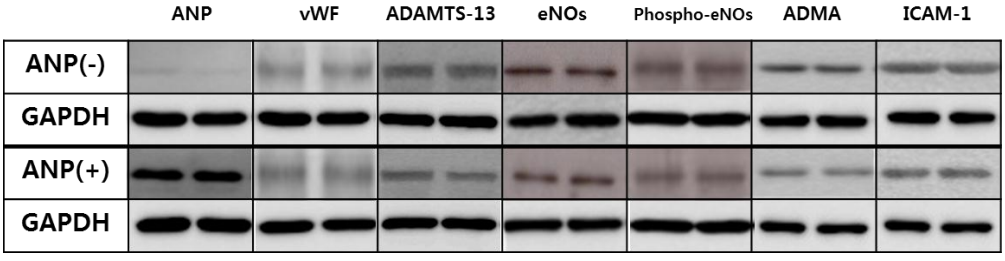


Figure 5. The quantification (Western blotting) of proteins which represent platelet aggregation (vWF, ADAMTS-13), endocardial function (eNOs, Phospho-eNOs, ADMA) and intercellular adhesion (ICAM-1) according to the expression of ANP.

Clinical implications of the presence of ANP in patients who underwent the maze operation

Multivariable Cox regression analysis showed that the presence of ANP was significantly associated with recurrence of AF after maze operation (HR 9.6, 95% CI 1.9-47.1, p=0.005) (Table 4).

Table 4. Multivariable Cox regression analysis of AF recurrence after maze

	HR	95% CI	p
Age	0.944	0.878-1.015	0.117
Male	0.214	0.049-0.946	0.042
LA volume index	1.022	0.992-1.052	0.148
The expression of ANP	9.582	1.947-47.148	0.005

In most ANP+ patients, AF recurred within 150 days, and there was a significant difference in clinical outcomes between the ANP+ and ANP- groups (log rank p=0.034) (Figure 6).

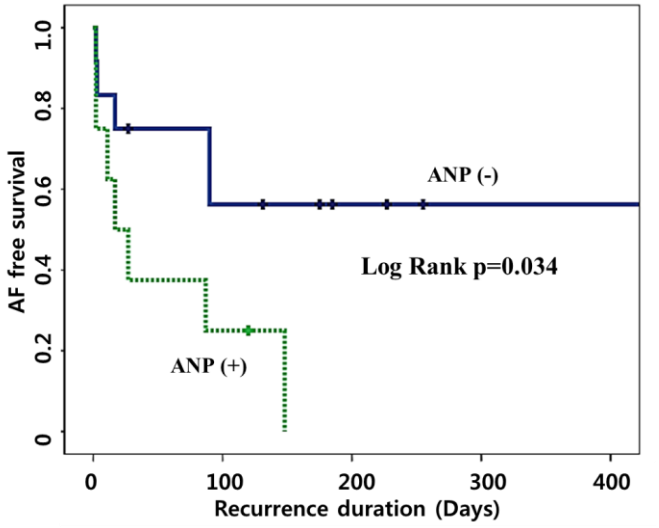


Figure 6. AF-free survival after the maze operation depends on the expression of ANP

IV. DISCUSSION

The expression of atrial ANP in patients with AF was strongly associated with atrial amyloid deposits, and reflected decreased endocardial function and decreased expression of intercellular adhesion. Intracardia hemodynamic change played key roles as mediators of atrial change by AF, and influenced clinical outcome after maze operation. The expression of ANP showed inverse correlation with angiotensin II, which reflected the formation of atrial fibrosis. These findings were consistent with previous study results, suggesting ANP(+) patients had less benefit from angiotensin converting enzyme (ACE) inhibitor.

Clinical meanings of atrial amyloid deposition and ANP expression

IAA usually occurs at sites were ANP is synthesized, because the cardiac and extracardiac signals that stimulate the synthesis and secretion of ANP may result in amyloidogenesis.[14,15] So none of the enrolled patients had clinical evidence of polyneuropathy, polyclonal gammopathy or a family history of amyloidosis, and all extracardiac causes of amyloid deposit were excluded. IAA belongs to the family of senile amyloidosis and these findings have many evidences in previous studies.[6,7,16,17] So the incidence of IAA has increasing tendency with age. Atrial amyloid deposits were also immunoreactive for ANP and thus interpreted as IAA.[17] These findings were consistent with our study. The ANP(+) patients showed older tendency than ANP(-) patients, amyloid deposits also coincided with the presence of ANP.[7] And the area of the amyloid deposits was related to the amount of ANP regardless of cardiac layers, although more amyloid was distributed in the endocardial layer than the myocardium. Moreover, the ANP+ tissue showed decreased endocardial function (eNOs, phosphorylated eNOs) and expression of ICAM. Interestingly, a previous study showed that IAA and atrial fibrosis had an inverse correlation, suggesting that these patients may not benefit from treatment with angiotensin converting enzyme (ACE) inhibitors to reduce the amount of atrial fibrosis.[8] Our study

results were also consistent; ANP+ atrial tissue showed decreased expression of angiotensin II compared to ANP- atrial tissue. Patients with higher levels of ANP expression, which corresponds to less atrial fibrosis deposits, may not benefit from ACE inhibitor treatment. Although we did not quantify angiotensin II in serum, only tissue, these findings may be the basis for the inverse correlation between IAA and atrial fibrosis.

Intracardiac hemodynamic change as mediators of atrial tissue remodeling

Atrial natriuretic peptide (ANP) plays roles as a strong vasodilator, which is secreted in atrium. And it is involved in maintaining homeostasis of body water and sodium concentration. So ANP inhibits the secretion of aldosterone from the adrenal gland and secretion of renin from the kidney and subsequently the production of angiotensin II in response to high blood pressure or increased volume status. Receptor-agonist binding complex of ANP reduces blood volume as enforcing natriuresis from kidney and therefore may affects cardiac output or systemic blood pressure. In our study, patients with ANP (+) were associated with low PAP compared with those with ANP(-). LA pressure increased more by AF with increased LV filling pressure by mitral valve disease, and it may affect to enforce secretion of ANP. Consequently, general effects of ANP may play inverse roles to intra-cardiac pressure and body volume status caused by the renin-angiotensin system. Moreover, the vasodilatic effects of ANP may dilate veins directly and decrease CVP related to reducing of ventricular preload and cardiac output. To summarize, ANP plays as a counter-regulatory system for the renin-angiotensin-aldosterone system, and has strong vasodilatic effects in response to increased intra-cardiac pressure and blood volume.

Atrial amyloid deposits and AF

Imaging studies [18-20] and studies of atrial tissue have shown significant associations between AF and atrial fibrosis. Studies using animal or human

tissue model have also demonstrated that AF alters atrial tissue,[21,22] and that atrial fibrosis plays a key role in the initiation or maintenance of AF.[22-24] Recently, we reported more advanced matrix fibrosis in the LA of patients with AF than those with sinus rhythm.[25] Deposits of atrial amyloids are also influenced by various factors affecting the heart, such as age, mitral valve disease, and AF. However, it is not known whether amyloid deposits cause AF or vice versa. Once amyloid is deposited in the atrium by any causes, it influences atrial conduction, myocardial contraction and relaxation. P-wave duration was significantly longer in patients with amyloid deposits, and amyloid deposits were an independent predictor of AF even after adjusting for age and sex.[8] In a study of 100 consecutive autopsied heart, the incidence of atrial tachyarrhythmia was higher in hearts with high grade IAA than those with low grade IAA.[26] Finally, AF promotes the deposition of amyloid, and amyloid deposition promotes AF, resulting in a vicious cycle.

Clinical implications of atrial amyloid deposits

Several studies have investigated the association between amyloid deposits and AF.[5,8] Indeed, deposits of amyloid in IAA, as determined by ANP expression, have been shown to be strongly associated with the incidence of AF and aggravate the progression of AF. However, these studies investigated the association between amyloid deposits and the incidence of AF in patients with various valve diseases that are characterized by advanced left atrial remodeling. So we examined patients with just isolated mitral valve disease to overcome this limitation of previous studies. We found that atrial amyloid deposits in patients with AF affected clinical outcomes after the maze operation. In addition, the expression of ANP was strongly associated with atrial tissue remodeling as assessed by expression of endocardial function, fibrosis, and interstitial adhesion. But, we did not find an association between changed endocardial function caused by atrial tissue remodeling and thrombogenesis in LAA. Both

vWF and ADAMTS13 were correlated negatively with the expression of ANP; an inverse correlation was not detected. ADAMTS 13 is usually known as a large protein involved in degrading large vWF multimers as vWF cleaving protese. This unusual finding may be due to the burning out of atrial tissue by strong hemodynamic factors of severe mitral valve disease.

Limitations

Our study had several limitations. First, although we included patients who underwent the maze with mitral valve surgery, we could not demonstrate that the atrial tissue changes were due to AF, because mitral valve disease also results in remodeling of LA tissue. ANP+ tissue showed lower expression in both vWF and ADAMTS13, and this finding was not consistent with a previous study that reported an inverse correlation between vWF and ADAMTS13 expression. Furthermore, the cohort of patients included in this study was small, so we could not clarify the association between atrial tissue changes and clinical recurrence rate except early recurrence.

V. CONCLUSION

Atrial amyloid deposits were strongly associated with the expression of ANP, accompanied by same intracardiac hemodynamic change. Deposits of ANP in the LA were also associated with decreased endocardial function and expression of intercellular adhesion molecules. In consequence, the expression of ANP was significantly correlated with AF recurrence after the maze operation.

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APPENDICES

ACE	angiotensin converting enzyme
ADAMTS13	a disintegrin and metalloproteinase with a thrombospondin type 1 motif, member 13
ADMA	asymmetric dimethylarginine
AF	atrial fibrillation
ANP	atrial natriuretic peptide
BMI	body mass index
BSA	body surface area
CVP	central venous pressure
ECG	electrocardiography
eNOS	endocardial nitric oxide
GAPDH	phosphate-buffered saline
IAA	isolated atrial amyloid
ICAM-1	intercellular adhesion molecule-1
LA	left atrium
LAA	left atrial appendage
LAVI	LA volume index
LVMI	LV mass index
PAP	pulmonary arterial pressure
PBS	phosphate-buffered saline
Phospho-eNOS	phosphorylated eNOS
SD	standard deviations
TTE	trans-thoracic echocardiography
VIF	Variance inflation factors
vWF	von Willebrand factor

ABSTRACT(IN KOREAN)

심방 세동 환자에서 Atrial natriuretic peptide 의 축적과 심방의 변성 및 혈역동학적 변화와 maze 수술 이후 임상적 예후와의 관련성

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박 준 범

배경: 고립성 심방 아밀로이드 축적 (IAA: Isolated atrial amyloid) 은 심방 세동 (AF: atrial fibrillation)과 관련되며, 심방 세동이 지속됨에 따라 심방의 기질적 변화를 초래한다. 그러나 심방 세동 환자에서 심방 조직의 기질적 변화와 심방 나트륨 이뇨단백 (ANP: atrial natriuretic peptide) 사이의 직접적 증거는 없었다. 그리고 심방의 기질적 변화 및 심방 나트륨 이뇨단백과 메이즈 (maze) 수술 이후의 임상적 유용성에 대한 연구는 없었다.

방법: 20명의 좌심방이 조직은 방실 판막 수술과 메이즈 수술과정에서 획득되었고, 모든 조직은 아밀로이드를 검출하기 위해 콩고(Congo red) 염색 하에 편광 현미경으로 관찰하였고, 아밀로이드 축적의 영역은 이미지 프로를 사용하여 정량화 되었다. 심방 나트륨 이뇨단백의 정량화는 웨스턴 블로팅 (Western blooting)을 사용하였다. 그리고 심내막 기능단백질 발현 (내막 산화 질소 [eNOS], 인산화-내막 산화 질소 [Phospho-eNOS]), 세포 간 부착

분자 (ICAM-1) 및 혈전 인자 (von Willebrand factor [vWF], a disintegrin and metalloproteinase with a thrombospondin type 1 motif, member 13 [ADAMTS13])을 정량화 하기 위해 웨스턴 블로팅을 사용하였다. 수술 전에 측정된 혈역동학적 수치들은 (폐동맥압)은 심방 나트륨 이뇨단백의 발현 여부에 따라 분석되었다.

결과: 1. ANP 발현 양성환자의 경우, 수축기 폐동맥압이 낮게 측정되었으며 (p=0.04), 아밀로이드의 축적과 수축기 폐동맥압은 음의 상관관계를 보여주었다 (r=-0.6, p=0.008). 2. eNOS (p=0.043), phosphor-eNOS (p=0.013), vWF (p=0.031) and ICAM-1 (p=0.013)의 경우 ANP가 발현된 조직에서 낮게 측정되었다. 3. 아밀로이드의 축적이 많을수록 ANP의 발현양도 높게 측정되었다 (p=0.002) 4. 메이즈 수술 이후 심방 세동의 재발은 ANP 발현 양성환자에서 높게 나타났으며 (p=0.033), 모든 재발은 150일 이내에 나타났다. ANP 발현 양성 여부는 나이, 성별, 심방의 부피를 보정한 이후에도 평균 362.3±473.8일 (최대 1,565일) 동안의 경과 관찰 기간 동안, 메이즈 수술 이후 심방 세동의 재발과 관련성을 보이고 있었다.

핵심되는 말 : 심방 나트륨 이뇨단백, 아밀로이드, 심내막 산화질소, 심방세동, 메이즈