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Platelet-Derived Growth Factor-A Overexpression Correlates with Atrial Fibrosis in the Patients with Atrial Fibrillation Secondary to Rheumatic Valvular Disease

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Abstract

Objective: To investigate the relationship between platelet-derived growth factor-A (PDGF-A) and atrial fibrosis in patients who have developed atrial fibrillation (AF) secondary to rheumatic valvular disease. **Methods**: 84 selected patients participated in the current study who have developed rheumatic heart disease and were going to have a cardiac surgical operation. In the current study, whole subjects were divided into two group, they were atrial fibrillation (AF) group (the quantity is thirty-nine) and sinus rhythm (SR) group (the quantity is forty-five). Before the operation, complete clinical data was available for the whole patients. During the operation, the right atrial tissue (0.3 - 0.5 mm^3) was disserted from every patient. Right atrial fibrosis was observed by Masson staining and the distribution of PDGF-A in right atrium specimen was observed by immunohistochemistry. RT-PCR techniques were applied to admeasure the mRNA expressions of PDGF-A in patients' atrial tissue. At the same time, western-Blot techniques were employed to admeasure the protein expressions of PDGF-A. **Results**: In baseline clinical characteristics, in both AF group and SR group, there was no apparently difference between them ($P > 0.05$); compared with SR group, the diameters of left atrium and right atrium in AF group were apparently increased ($P < 0.05$). The results of Masson staining revealed that the atrial tissue fibrosis was clearer in AF group, and collagen volume fraction in the AF group was evidently exceeding SR group ($P < 0.05$). The expressions of PDGF-A's mRNA and protein from right atrial tissue in the AF group were evidently greater than SR group ($P < 0.05$). The mRNA and protein expressions of PDGF-A and the right atrium diameter go hand in hand. **Conclusion**: Atrial

remodeling plays an important role in patients with valvular atrial fibrillation; PDGF-A in patients with AF was highly expressed in the right atrial, and was closely related with atrial fibrosis.

Keywords

Atrial Fibrillation, Platelet-Derived Growth Factor-A, Collagen Volume Fraction, Atrial Fibrosis

1. Introduction

The most common supraventricular arrhythmia in the world is Atrial fibrillation (AF), which is characterized by rapid and irregular activation of the atrium [1] [2].

Several cardiovascular disorders predispose to AF, such as valvular heart disease [3], coronary artery disease, congestive heart failure [4] [5], and hypertension. The most important histopathological change in atrial fibrillation is atrial fibrosis which involves a disproportionate excessive accumulation of extracellular matrix [6] between muscle fibers and around blood vessels. Atrial fibrosis underlies atrial structural remodeling [6] [7] and reportedly contributes to the development and maintenance of AF [8] [9]. Although, it is not clear for the precise pathophysiological mechanisms, it has been proposed that extracellular matrix-modulating enzymes, cytokines, growth factors, and components of the fibrinolytic system play considerable roles in AF.

Platelet-derived growth factor-A (PDGF-A), a member of the PDGF/vascular endothelial growth factor family [10], is highly expressed in the myocardium throughout development and adulthood [11]. PDGF-A solely binds to PDGF receptor-α (PDGFR-α), and subsequently activates several intracellular signaling cascades, then stimulates growth, differentiation and migration of cells [10] [12]. It is known that PDGF-A and PDGFR-α are essential for the development of support cells in the vasculature, and involved into tissue fibrosis. However, it is not well investigated whether they contribute to atrial fibrosis. In order to determine whether PDGF-A participates in atrial fibrosis associated with AF, we investigated the expression and distribution of PDGF-A in patients with and without AF.

2. Materials and Methods

2.1. Patient Enrollment and Data Collecting

84 consecutive patients with rheumatic heart disease (RHD) who were going to undertake cardiac surgery were enrolled in this study at First Affiliated Hospital of Xi'an Jiaotong University, Xijing Hospital and Shanxi Provincial People's Hospital in 2012. All patients consecutively underwent simple mitral valve replacement surgery.

Baseline demographics, physical examination, routine laboratory testing, echocardiography, and additional clinical data were available for all patients before surgery.

84 selected patients were divided into two group, they were atrial fibrillation (AF) group (the sample size is thirty-nine) and sinus rhythm (SR) group (the sample size is forty-five). The subjects were considered eligible to be enrolled into the AF group if they had obvious AF history and had been documented by electrocardiogram with AF for more than 6 months. The SR group is composed of patients who is sinus rhythm and without history of atrial fibrillation.

Before surgery, no patients received any type of angiotensin receptor blockers (ARB) or angiotensin-converting enzyme inhibitors (ACEI) for at least six months, and none of the subjects had taken anti-inflammatory drugs previously more than two weeks before the study. In the present study, we excluded the patients as follows: 1) patients who exceeded 65 years old or had a history of cancer; 2) patients with complicated diabetes, renal or liver failure; 3) patients with hyperthyroidism, hypertension, autoimmune disease; 4) patients with heart failure over New York Heart Association (NYHA) III or left ventricular ejection fraction (LVEF) less than 40%, and other heart diseases; 5) patients who suffered rheumatic fever in active stage.

Before they take part in this investigation, every selected patient has signed informed consent forms that obtained from every selected patient or their family members. All informed consent forms and the procedure protocols were authorized by the Ethics Committee of the First Affiliated Hospital of Xi'an Jiaotong University. The investigation tallies with the principles outlined in the Declaration of Helsinki.

2.2. Human Cardiac Tissue Collection and Storage

All patients underwent cardiac surgical operation with moderate hypothermia (33˚C - 34˚C) and the right atrium (RAA) tissue (0.3 - 0.5 mm^3) was disserted during the surgery. Each piece of tissue was cut immediately into three parts. One was dropped into liquid nitrogen and then stored in the –80˚C for RT-PCR and Western blot. Samples for immunofluorescence were promptly implanted into Tissue OCT-Freeze compound, flash frozen, and cut into 10-μm sections using Cryostat at optimal cutting temperature. All tissue sections were stored at –80˚C until they were used for immunostaining and analysis. The third was fixed in 4% paraformaldehyde solution for 12 - 24 hours, imbedded in paraffin with Masson's trichrome staining.

2.3. Masson's Trichrome Staining and Collagen Volume Fraction Assay

The specimens fixed in 4% paraformaldehyde were subjected to alcoholic dehydration and embedded in paraffin. 4 μm serial sections were sliced and subjected to Masson's trichrome staining to highlight collagen fibers. Collagen volume fraction (CVF) assay was performed in tissue sections of the right atrium. Mas-

son's trichrome a was obtained from Boster Biological Engineering Corporation (Wuhan, China). Two slides of each sample were randomly selected and observed under polarization microscope. Six different vessel-free fields (×200) of each slide were captured, and the images were analyzed using Image Pro Plus 6.0 (IPP 6.0) software. Collagen volume fraction was showed as the percentage of area of positive collagen staining in the total area of the image. The following formula was used to calculate the fibrosis score: collagen fiber area/total view area × 100%.

2.4. Detection of Positive for PDGF-A by Immunofluorescence Staining

The right atrial tissue samples were fixed in 4 per centum paraformaldehyde for 24 hour, afterwards in 30 per centum sucrose at 4˚C, until these tissues sank. After embedded in embedding reagent for frozen sections, 6-μm sections were obtained onto polylysine-coated slides. These slides were treated with acetone at 4˚C for 15 min and then with PBS (phosphate buffered solution). After treatment with 0.5% Triton X-100 at 37˚C for 30 minute, sections were hatched with 10% normal goat serum for 45minute at 37˚C. Subsequently, these sections were treated with foremost antibody (PDGF-A: 1:200; vimentin: 1:2500) at 4˚C overnight and secondary antibody (FITC conjugated goat anti-mouse antibody: 1:200; rhodamine red conjugated goat anti-rabbit antibody: 1:200; DAPI: 1:2000) at 37˚C for 30 min. After washing in PBS 5 times (5 min for each), mounting was done with anti-quencher, and sections were observed under a fluorescence microscope and photographed (×200). ImageJ image analysis software was employed to analyze and merge these photographs. Negative controls were obtained by omitting the incubation with primary antibodies.

2.5. Detection the Expressions of mRNA of PDGF-A and Type I, III Collagen by RT PCR

In brief, frozen human right atrial tissue samples were thawed and homogenized on ice, then overall RNA was isolated using RNA-simple Total RNA Kit (Tiangen Biotechnology, China) according to the corporation's directions and quantified. Then, using ReverTra Ace qPCR RT Kit (Tiangen Biotechnology) and according to the manufacturer's instructions, RNA was reversely transcribed into cDNA successfully. Amplification of cDNA was done on thermal cycler (Applied Biosystems Step One Plus System). The volume of the reaction mixture was 20 μl, according to merchant description, amplification was executed successfully (SYBR® Premix Ex Taq™ II PCR kit and Applied Biosystems Step One Plus System). The reaction conditions are as follows: pre-denaturation for 30 second at 95˚C and 40 cycles of 95˚Cfor 5 second and 60˚C for 30 second. The melt curve was applied to define the specificity of products. The supporting software was applied to analysis the Ct value of products. According to the following formula: ΔCt = Cttarget gene − Ctinternal reference, the ΔCt was **Table 1** Oligonucleotide probes calculated in two groups, respectively. The relationship between the

CT value of the target gene and the copies of this gene is negative, consequently, with the increase of the ΔCt, the gene expression decreased. Then, $2^{-\Delta\Delta Ct}$ method was applied to calculate the relative mRNA expression of target genes. Oligonucleotide probes were in Table 1.

2.6. Western Blotting Analysis

For Western blot analysis, frozen human right atrial tissue samples were used for protein isolation. Protein extraction was followed by the instruction of the total protein extraction kit (Apply gen Technologies Inc, China). Proteins (10 μg) was isolated by sodium dodecyl sulfate-polyacrylamide gel electrophoresis (SDS-PAGE) and transferred to nitrocellulose membranes (Stratagene, USA) followed by staining with Ponceau S solution (Sigma, USA). The membranes were blocked with 5% non-fat dry milk and then explored rabbit polyclonal anti-human PDEGF-A (Abcam, USA) antibodies. Horse radish peroxidase-conjugated rabbit anti-mouse or anti-rabbit IgG (1:5000, Santa Cruz Biotechnology) was used as secondary anti-body followed by incubation with ECL Western Blot Detection Kit (Amersham, The Netherlands). The amount of protein chosen was in the linear immunoreactive signal range. The immunoreactive signals were exposed to Kodak film for 5 min and analyzed with gelpro analyzer after analysis software (Bio-Rad, USA) normalized by the corresponding value of β-actin. Experiments were repeated three times and the mean was scored.

2.7. Statistical Analysis

All statistical analysis was made by the SPSS version 15.0. If they were normally distributed, continuous variables were indicated as mean ± standard deviation (x ± s). Comparisons of means were applied to independent t test between AF group and SR group. Groups for categorical variables were analyzed by chi-square. Correlation coefficients were assessed between PDGF-A and LAD, including the protein and mRNA expression of PDGF-A. A value of $P < 0.05$ was considered statistically significant.

The reporting of this study conforms to STROBE guidelines.

Table 1. Oligonucleotide probes.

The target genes	Primers	Base composition
PDEGF-A	sense	5'-ACGTCCGCCAACTTCCTGA T-3'
	antisense	5'-TCCGGATTCAGGCTTGTGGT-3'
Type I collagen	sense	5'-GCGACAGAGGCATAAAGGGT-3'
	antisense	5'-CCAGGGAGACCGTTGAGTC-3'
Type III collagen	sense	5'-GAGCTTCCCAGAACATCA-3'
	antisense	5'-ATTCCCCAGTGTGTTTCG-3'
GAPDH	sense	5'-CCTCCTGCACCACCAACT-3'
	antisense	5'-CTTCTGGGTGGCAGTGATG-3'

3. Results

3.1. Clinical Characteristics

Patients' characteristics are shown in Table 2. As a whole, most characteristics were similar between two groups. Left and right atrial diameters, measured by echocardiography, were significantly larger in the AF group than the SR group. All drugs were stopped at least 12 h before surgery.

3.2. Collagen Content and Distribution

Representative examples of right atrial tissue stained with Masson's trichrome from each group are shown in Figure 1. Although the marked interstitial alterations were present because of all patients with RMVD, an apparent difference was observed between the two groups.

Comparison with the SR (Figure 1(a)) patients, there were abundant collagen fibers in the AF (Figure 1(b)) group. On the contrary, only a small amount of collagen fibers was observed in the SR group. The CVF in the AF patients (45.4% ± 2.33%) increased more drastically than that in the SR patients (12.9% ± 1.02%) ($P < 0.001$; Figure 1(c)), indicating some relationships between AF and fibrosis.

Table 2. Clinical characteristics.

	AF (n = 39)	SR (n = 45)	t/χ^2 values	P value
Basic data				
Age (years)	51.1 ± 9.0	48.1 ± 10.9	−1.405	0.164
Sex	24/15	19/26	3.083	0.086
male/female (n)				
SBP (mmHg)	112.4 ± 10.4	115.8 ± 14.3	1.242	0.218
DBP (mmHg)	71.7 ± 7.4	70.5 ± 11.5	−0.567	0.572
Laboratory examinations				
WBC (10^9/l)	6.29 ± 1.55	6.0 ± 1.91	−0.763	0.448
RBC (10^{12}/l)	4.55 ± 0.67	4.44 ± 0.51	−0.811	0.420
HB (g/l)	137.1 ± 16.6	135.3 ± 16.8	−0.482	0.631
CR (μmol/l)	92.47 ± 12.46	91.58 ± 12.43	−0.225	0.823
Echocardiographic parameters				
LVEF (%)	53.10 ± 5.90	54.47 ± 5.90	1.041	0.301
LAD (mm)	58.03 ± 13.93*	49.55 ± 17.17*	−2.420	0.018
RAD (mm)	26.88 ± 4.81*	24.32 ± 4.26*	1.721	0.038

SBP, systolic blood pressure; DBP, diastolic blood pressure; WBC, white blood cell; RBC, red blood cell; HB, hemoglobin; CR, serum creatinine; LVEF, left ventricular ejection fraction; LAD, left atrium diameter; RAD, right atrium diameter.

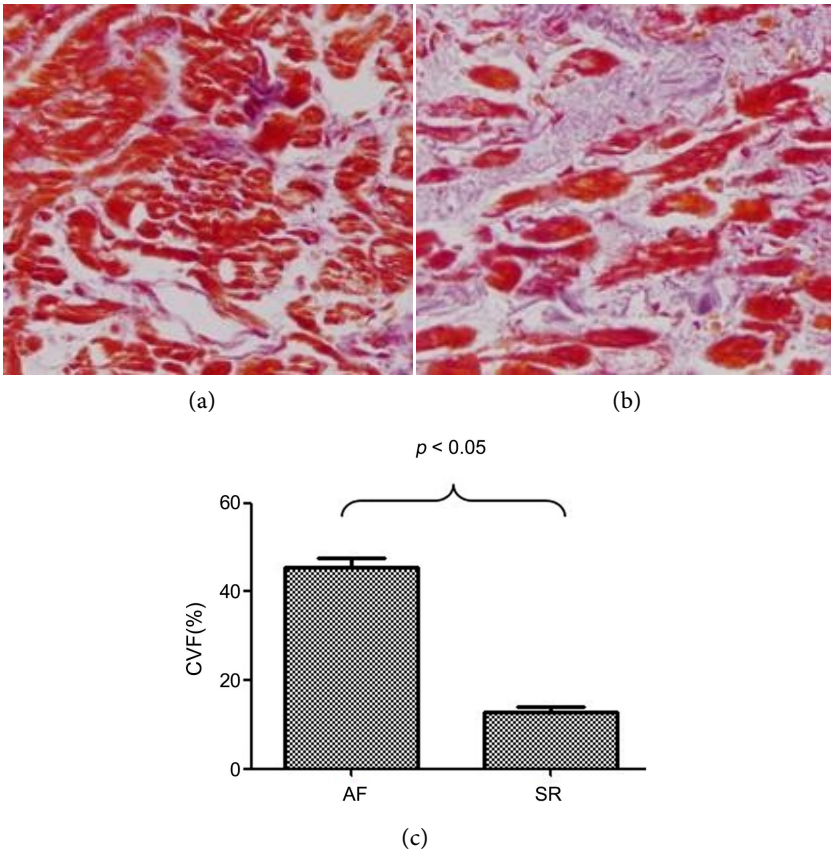


Figure 1. Representative examples of right atrial tissue stained with Masson's trichrome. (a): the intensity of collagen fibers with Masson's Staining in SR (original magnification × 200). (b): the intensity of collagen fibers with Masson's Staining in AF (original magnification × 200). (c): CVF was higher in AF group. AF, atrial fibrillation; SR, sinus rhythm.

3.3. mRNA Expressions of Type I Collagen and Type III Collagen

We performed qRT-PCR to test whether the type I collagen or type III collagen level increases in the AF patient's right atrial tissues. After RT-PCR, the Ct value and number of cycles were applied to depiction, and finally, the mRNA's amplification curve was acquired. The results demonstrated that the way has good repeatability and consistent amplification efficiency. $2^{-\Delta\Delta Ct}$ method was applied to represent the relative expressions of target genes. The results displayed that the expression of mRNA of type I collagen was 2.042 ± 0.177 in the AF group, which was markedly higher than those in the SR group (0.988 ± 0.099), $P < 0.05$ (**Figure 2(a)**).

Compared with the SR group, the expression of type III collagen mRNA in the AF group was greater, but there was no marked difference between the AF group and SR group (1.228 ± 0.151 VS 1.067 ± 0.068, $P > 0.05$), (**Figure 2(b)**).

3.4. Increased PDGF-A Expression in the Right Atrial of Patients with AF

Representative sections of the immunofluorescent stained right atrial tissue from each group are shown in **Figure 3**. Although PDGF-A was expressed in both

groups, compared with the SR group, the level of the PDGF-A's protein was apparently higher in the right atrial myocardium of the AF group (Figure 4).

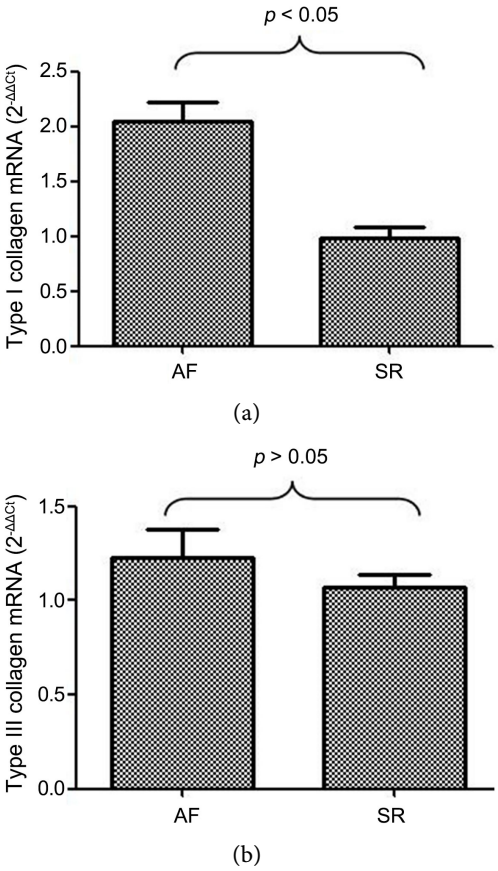
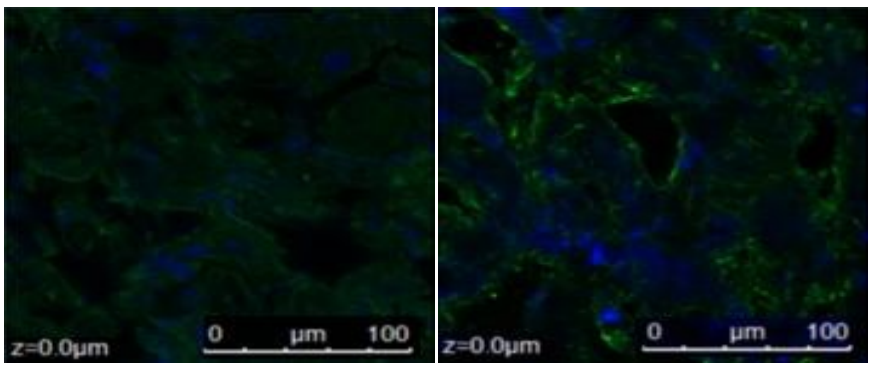


Figure 2. (a): mRNA expression of type I collagen in AF and SR groups; (b): mRNA expression of type III collagen in AF and SR groups. AF, atrial fibrillation; SR, sinus rhythm.



(a) (b)

Figure 3. Representative sections of the immunofluorescent stained right atrial tissue showed increased PDEGF-A in patients with AF compared with patients in SR. (a): Immunofluorescent stained expression of PDEGF-A in SR (original magnification × 200). (b): Immunofluorescent stained expression of PDEGF-A in AF (original magnification × 200). AF, atrial fibrillation; SR, sinus rhythm.

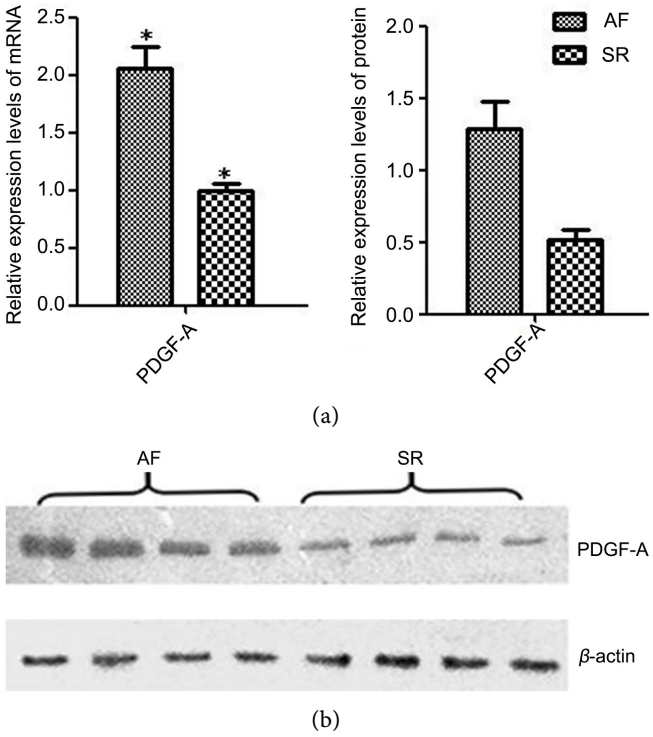


Figure 4. (a): Relative expression levels of mRNA and Protein in AF and SR groups; (b): Protein expression of PDGF-A in AF and SR groups by Western blotting; AF, atrial fibrillation; SR, sinus rhythm.

3.5. mRNA and Protein Expressions of PDGF-A

To test whether the PDGF-A increases in the right atrial tissues of the AF patients, we performed qRT-PCR and Western blotting. After real time PCR, the Ct value and number of cycles were applied to depiction, finally, the amplification curve of mRNA was acquired. The results demonstrated that the way has good repeatability and consistent amplification efficiency. $2^{-\Delta\Delta Ct}$ method was applied to indicates the relative expressions of target genes. The results displayed that the PDGF-A's mRNA expressions was 2.062 ± 0.184, the PDGF-A's protein expressions was 1.282 ± 0.193 in AF group, which were markedly higher than those in SR group (0.991 ± 0.062 and 0.517 ± 0.067, respectively; $P < 0.01$).

3.6. Positive Correlation between Protein and Gene Expression of PDGF-A and Type I Collagen

A strong positive correlation existed between mRNA of PDGF-A and Type I collagen; western blotting revealed a strong positive correlation between protein of PDGF-A and mRNA of Type I collagen (**Figure 5(a)**, $r = 0.75$; **Figure 5(b)**, $r = 0.72$, respectively).

4. Discussion

Numerous studies demonstrate that patients with chronic AF secondary to RMVD are very common, and in these patients, structural remodeling is very

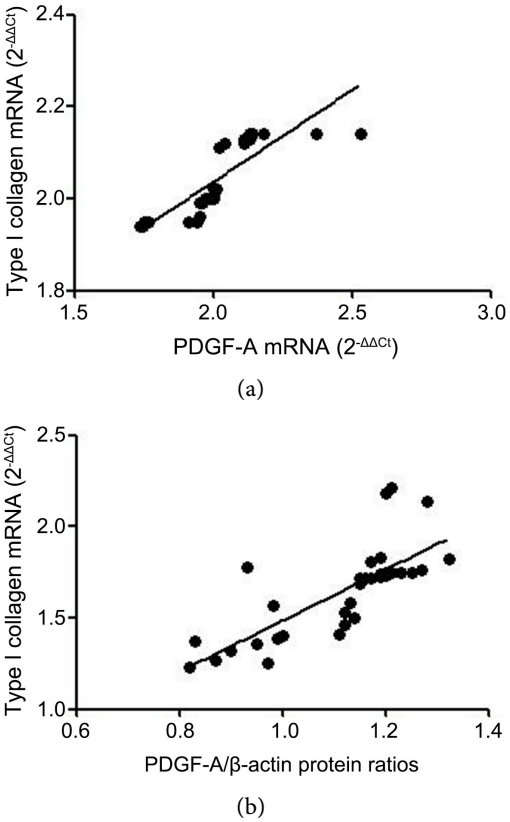


Figure 5. (a): positive correlation existed between mRNA of PDGF-A and Type I collagen in AF group; (b): positive correlation between protein of PDGF-A and mRNA of Type I collagen in AF group; AF, atrial fibrillation.

important for AF initiation and maintenance [13] [14]. Atrial fibrosis, as a hallmark of structural remodeling, has been implicated in tissue biopsies from AF patients [15]. Atrial fibrosis results from extracellularmatrix (ECM) accumulation of fibrillar collagen deposits [16]. Interstitial fibroblasts are differentiated to myo fibroblasts, which produce large amounts of collagen that replace degenerating myocardial cells. Expansion of ECM between cardiomyocytes may cause conduction delays and create alternate conduction pathways. These changes result in ectopicfoci and anisotropic conduction, creating nonuniform wave fronts that facilitate abnormal reentrant arrhythmias. Atrial fibrosis involves multiple factors such as the rennin-angiotensin system, TGFβ1, oxidative stress and in fl ammation [17], but the exact mechanisms responsible for the structural changes that accompany AF in patients with RMVD are unknown. It is clear that atrial fibrosis occurs as a result of underlying cardiac disease (e.g. RMVD) affecting atrial tissue, and this process of remodeling in turn acts as a substrate for the initiation and maintenance of AF [18] [19].

Fibrosis is thought to occur on both a tissue and cellular level, and our study aimed to investigate the signaling pathways responsible for atrial fibrosis.

PDGF is mainly released by platelet α particles, in addition to monocytes, smooth muscle cells, endothelial cells, etc. can synthesize and release PDGF. PDGF is an important cell-stimulating agent that can stimulate the division and proliferation of a variety of cells, and has chemotaxis on fibroblasts and smooth muscle cells. About 85% of the extracellular matrix in normal myocardial tissue is composed of extracellular collagen secreted by fibroblasts, of which type I collagen accounts for about 85% and type III collagen accounts for about 11%.

PDGF-A is a potent growth factor that plays important roles in the proliferation [20], migration and survival of interstitial cells. An increasing number of proof has proved that PDGF/PDGFR signaling pathway is associated with the pathological fibrosis of multiple organs. PDGF and its receptor system play an important role in the development of myocardial fibrosis. In addition, during myocardial fibrosis in salt-sensitive hypertensive rats, PDGFR-α acts at early stage, and PDGFR-α expressions increase in fibroblasts and myofibroblasts, suggesting that PDGF/PDGFR signaling pathway is involved in the myocardial fibrosis via stimulating fibroblasts to proliferate and transform into myofibroblasts and to secret massive collagens [21] [22]. PDGF-α receptor mRNA is upregulated in acutely rejecting cardiac allografts, and mRNA of PDGF-A is upregulated in chronically rejecting cardiac allografts [23]. PDGF-A markedly increased pro fibrotic TGFβ-1 mRNA and accelerated the formation of myocardial fibrosis, indicating that PDGF may also increase TGFβ-1 levels to the formation of fibrosis. Atrial fibrillation, characterized by atrial fibrosis, is a frequent arrhythmia, which increases the risk of stroke and heart failure [24] [25]. Injection of neutralizing PDGFR-α specific antibody alleviated atrial fibrosis [26] [27]. The present research demonstrates that atrial fibrosis is distinct in patients with AF secondary to RMVD, establishing the relationship between PDGF-A and atrial fibrosis. This result strongly suggests that PGDF-A may be a good target for antifibrotic therapy in the heart.

But due to this study is a human tissue experiment, only right atrial tissue was collected, research data from left atrial tissue was missed. So it cannot simultaneously explore the relationship between PDGF-A and left atrial fibrosis in patients with atrial fibrillation secondary to RMVD, and therefore the distribution and expression of PDGF-A, the atrial structural remodeling cannot be contrasted between the left and right atria.

5. Conclusion

There was significant atrial remodeling in patients with chronic AF secondary to RMVD26; PDGF-A in patients with atrial fibrillation was highly expressed in the right atrial, and was closely related to atrial fibrosis. PDGF-A may be up-regulated expression of type I collagen gene, which participated into atrial fibrosis. Present study is a human tissue experiment, only right atrial tissue is collected. So it cannot simultaneously explore the relationship between PDGF-A and left atrial fibrosis in patients with atrial fibrillation secondary to RMVD.

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Conflicts of Interest

The authors declare no conflicts of interest regarding the publication of this paper.

References

[1] Koniari, I., Artopoulou, E., Velissaris, D., *et al.* (2021) Biomarkers in the Clinical Management of Patients with Atrial Fibrillation and Heart Failure. *Journal of Geriatric Cardiology*, **18**, 908-951.

[2] Qi, M.M., Qian, L.L. and Wang, R.X. (2021) Modulation of SK Channels: Insight into Therapeutics of Atrial Fibrillation. *Heart, Lung & Circulation*, **30**, 1130-1139. https://doi.org/10.1016/j.hlc.2021.01.009

[3] Haim, M., *et al.* (2015) Prospective National Study of the Prevalence, Incidence, Management and Outcome of a Large Contemporary Cohort of Patients with Incident Non-Valvular Atrial Fibrillation. *Journal of the American Heart Association*, **4**, e001486. https://doi.org/10.1161/JAHA.114.001486

[4] Goya, W.S., Olia, P., Lucy, L., *et al.* (2021) Adult Height and Incidence of Atrial Fibrillation and Heart Failure in Older Men: The British Regional Heart Study. *IJC Heart & Vasculature*, **35**, Article ID: 100835. https://doi.org/10.1016/j.ijcha.2021.100835

[5] Zafrir, B., Lund, L.H., Laroche, C., *et al.* (2018) Prognostic Implications of Atrial Fibrillation in Heart Failure with Reduced, Mid-Range, and Preserved Ejection Fraction: A Report from 14964 Patients in the European Society of Cardiology Heart Failure Long-Term Registry. *European Heart Journal*, **39**, 4277-4284. https://doi.org/10.1093/eurheartj/ehy626

[6] Hiroko, I., Shigeru, M., Yoshiki, S. and Yoshiki, S. (2021) Cardiac Fibrosis Models Using Human Induced Pluripotent Stem Cell-Derived Cardiac Tissues Allow Anti-Fibrotic Drug Screening *in Vitro*. *Stem Cell Research*, **54**, Article ID: 102420. https://doi.org/10.1016/j.scr.2021.102420

[7] Zou, R., Zhang, D., Lv, L., *et al.* (2019) Bioinformatic Gene Analysis for Potential Biomarkers and Therapeutic Targets of Atrial Fibrillation-Related Stroke. *Journal of Translational Medicine*, **17**, Article No. 45. https://doi.org/10.1186/s12967-019-1790-x

[8] Li, Y.C., Zhang, R.J., Hu, N.W. and Li, N.S. (2021) Atrial Fibrosis Underlying Atrial Fibrillation (Review). *International Journal of Molecular Medicine*, **47**, Article No. 9. https://doi.org/10.3892/ijmm.2020.4842

[9] Koniari, I., Artopoulou, E., Velissaris, D., *et al.* (2021) Atrial Fibrillation in Patients with Systolic Heart Failure: Pathophysiology Mechanisms and Management. *Journal of Geriatric Cardiology*, **18**, 376-397.

[10] Li, N.N., Yin, Q. and Du, R.Z. (2021) Research Progress of Platelet Derived Growth Factor in Cardiovascular Diseases. *Medical Review*, **27**, 3388-3393.

[11] Michael, B. and Kristian, P. (2018) PDGF Family Function and Prognostic Value in Tumor Biology. *Biochemical & Biophysical Research Communications*, **503**, 984-990.

[12] Wang, F.J. (2021) PDGFR *a* Progress of Its Role in Cardiac Development and Heart Disease. *Journal of Practical Medicine*, **38**, 184-187+190.

[13] Zhao, Q., Yang, W.T., Li, X.D. and Shan, Z.L. (2022) Research Progress on the Relationship between microRNA and Atrial Fibrillation and Atrial Fibrosis. *Journal of PLA Medical College*, 1-5.

[14] Lau, D.H., Schotten, U., *et al.* (2017) Pathophysiology of Paroxysmal and Persistent Atrial Fibrillation: Rotors, Foci and Fibrosis. *Heart Lung & Circulation*, **26**, 887-893. https://doi.org/10.1016/j.hlc.2017.05.119

[15] Ismail, A., Maciver, D.H., Garratt, C.J., *et al.* (2015) Effects of Persistent Atrial Fibrillation-Induced Electrical Remodeling on Atrial Electro-Mechanics—Insights from a 3d Model of the Human Atria. *PLOS ONE*, **10**, e0142397. https://doi.org/10.1371/journal.pone.0142397

[16] Mariana, F.M., Isabel, M., Diana, M., *et al.* (2020) Atrial Matrix Remodeling in Atrial Fibrillation Patients with Aortic Stenosis. *Revista portuguesa de cirurgia cardio-toracica e vascular*: *Orgao oficial da Sociedade Portuguesa de Cirurgia Cardio-Toracica e Vascular*, **27**, 250.

[17] Hu, J., Zhang, J.J., Li, L., *et al.* PU. 1 Inhibition Attenuates Atrial Fibrosis and Atrial Fibrillation Vulnerability Induced by Angiotensin-II by Reducing TGF-*β*1/Smads Pathway Activation. *Journal of Cellular and Molecular Medicine*, **25**, 6746-6759. https://doi.org/10.1111/jcmm.16678

[18] Liu, Z.Y., Wu, G.P., Lin, Y.Y., *et al.* (2022) Correlation between Microrna-328 and Atrial Remodeling in Patients with Atrial Fibrillation. *Journal of China Medical University*, 1-5.

[19] Sethi, N.J., Nielsen, E.E., Safi, S., *et al.* (2018) Digoxin for Atrial Fibrillation and Atrial Flutter: A Systematic Review with Meta-Analysis and Trial Sequential Analysis of Randomised Clinical Trials. *PLOS ONE*, **13**, e0193924. https://doi.org/10.1371/journal.pone.0193924

[20] Que, D.D. and Song, X.D. (2021) Role of Atrial Remodeling in Atrial Fibrillation. *Journal of Practical Electrocardiology*, **30**, 404-407.

[21] Leask, A. (2015) Getting to the Heart of the Matter: New Insights into Cardiac Fibrosis. *Circulation Research*, **116**, 1269-1276. https://doi.org/10.1161/CIRCRESAHA.116.305381

[22] Wang, L.X., Xiao, Y., Yuan, Y., *et al.* (2017) Imatinib Attenuates Cardiac Fibrosis by Inhibiting Platelet-Derived Growth Factor Receptors Activation in Isoproterenol Induced Model. *PLOS ONE*, **12**, e0178619. https://doi.org/10.1371/journal.pone.0178619

[23] Tuuminen, R., Nykanen, A.I., Krebs, R., *et al.* (2009) PDGF-A, -C, and -D but Not

PDGF-B Increase TGF-beta1 and Chronic Rejection in Rat Cardiac Allografts. *Arteriosclerosis, Thrombosis, and Vascular Biology*, **29**, 691-698. https://doi.org/10.1161/ATVBAHA.108.178558

[24] Chung, S.-C., Sofat, R., Acosta-Mena, D., *et al.* (2021) Atrial Fibrillation Epidemiology, Disparity and Healthcare Contacts: A Population-Wide Study of 5.6 Million Individuals. *The Lancet Regional Health—Europe*, **7**, Article ID: 100157. https://doi.org/10.1016/j.lanepe.2021.100157

[25] Nedios, S. (2021) Screening for Atrial Fibrillation: Does Cardiovascular Disease Reduce the Probability of Detection? *International Journal of Cardiology*, **339**, 54-55. https://doi.org/10.1016/j.ijcard.2021.07.003

[26] Liao, C.-H., Akazawa, H., Tamagawa, M., *et al.* (2010) Cardiac Mast Cells Cause Atrial Fibrillation through PDGF-A-Mediated Fibrosis in Pressure-Overloaded Mouse Hearts. *The Journal of Clinical Investigation*, **120**, 242-253. https://doi.org/10.1172/JCI39942

[27] Pan, J.Q., Zhang, D.X., Shou, X.L., *et al.* (2016) Features of Atrial Structure in Patients with Valvular Atrial Fibrillation: A Multicenter Study. *Journal of Xi'an Jiaotong University* (*Medical Sciences*), **37**, 77-107. (in Chinese)