Atrial Fibrillation and Atrial Fibrosis

Short Title: Myocardial fibrosis

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Abstract

Atrial fibrillation (AF) is the most common arrhythmia in humans. It affects 5% of the population over the age of 65 years and is projected to rise as the population ages.

Experimental data from animal models of AF shows that AF is associated with progressive structural and electrical remodeling of the atria. Atrial fibrosis alters atrial electrical conduction and excitability and provides a substrate for AF maintenance. However, whether fibrosis is causally related to AF or an epiphenomenon, and the precise mechanisms underlying atrial fibrosis remain unclear. A variety of signaling systems involving angiotensin II and related mediators, are centrally involved in atrial fibrosis. This article will review the role that atrial fibrosis plays in AF, the mechanisms of atrial fibrosis, and emerging therapeutic approaches to AF aimed at attenuating atrial fibrosis.

Keywords: Fibrosis, atrial fibrillation.
Atrial fibrillation (AF) is the commonest arrhythmia in humans (1) and accounts for significant morbidity and mortality. (2) It is a mechanistically complex arrhythmia for which available therapies remain suboptimal. For example, AF ablation is effective at eliminating the triggers for AF, particularly in patients with lone AF. (3,4) However, it is less effective in patients with persistent AF with underlying structural heart disease. (5) Anti-arrhythmic drug therapy is of limited efficacy because it targets only the electrical substrate of AF and is associated with side effects and pro-arrhythmia. (3) AF requires a trigger for initiation and favorable substrate for maintenance. It has been demonstrated that electrical, contractile and structural remodeling contribute towards AF maintenance in both animal models of AF and in clinical AF. (6) Fibrosis is the hallmark of atrial structural remodeling and results from an accumulation of collagen to replace degenerating myocytes. (7) In animal models of atrial pacing induced AF, electrical remodeling is potentially fully reversible after cessation of rapid pacing whereas structural remodeling regresses slowly and only partially. (6) Atrial fibrosis occurs as a common end point in a variety of settings, including senescence, (8) heart failure, (9) mitral valvular disease, (10) and myocardial ischemia. (11) It stands to reason therefore, that future therapies for AF should be aimed upstream at reversing or retarding the fibrotic process. The first aim of this review, therefore, is to highlight the role of atrial fibrosis in AF. The second aim is to elucidate the mechanisms of atrial fibrosis emphasizing atrial specific mechanisms of fibrosis. The final aim is to discuss potential new therapeutic options for AF that target atrial fibrosis.
Atrial Fibrillation and Atrial Fibrosis: Cause or Effect?

Atrial fibrosis is a common feature of clinical AF (12) and is associated with AF in a variety of experimental paradigms. (13) In humans, AF is secondary to underlying organic heart disease in 70% of patients, and lone AF in the rest. (14) Increased collagen deposition has been documented in lone-AF patients compared with sinus rhythm control subjects. (15) Fibrosis is also observed in AF patients with underlying structural heart disease, including mitral valve disease and cardiomyopathy. (16,17) In these patients, the extracellular matrix (ECM) volume and composition was found to correlate with AF persistence. (18) With increasing use of delayed gadolinium enhanced MRI to detect left atrial fibrosis in patients undergoing AF ablation, there is now an opportunity to evaluate the extent of atrial structural remodeling non-invasively. Oakes et al (18) reported that the extent left atrial fibrosis by MRI predicts AF recurrence after AF ablation. Mankhopf et al (19) performed a similar study and found that the degree of LA fibrosis predicts AF recurrence regardless of underlying co-morbidities. Taken together, these clinical studies highlight the association between atrial fibrosis and AF. However, the mechanisms by which fibrosis promotes AF remains unclear.

Mechanisms of Atrial Fibrillation in Fibrosis

To better understand the mechanisms of atrial fibrosis and AF, a review of experimental models of AF is instructive. Several large animal models of AF have been used for the study of AF. These include chronic rapid atrial pacing, chronic ventricular pacing to produce congestive heart failure, mitral regurgitation and ageing. (20-23) In general, atrial fibrosis is a prominent characteristic of the CHF, mitral regurgitation and
ageing models of AF whereas electrical rather than structural remodeling is more prominent in the rapid atrial pacing model. In dogs with CHF, (13) atrial fibrosis causes localized regions of conduction slowing and heterogeneity. This provides the basis for unidirectional block and macro-reentry. In addition to fibrosis, CHF also alters atrial ionic current and Ca2+ handling properties, (24,25) however, in this model, structural remodeling has been shown to be the main substrate for AF rather than electrical remodeling. (13) On the other hand, the rapid atrial pacing model is characterized by minimal atrial fibrosis but prominent electrophysiological remodeling including the downregulation of I\textsubscript{cal} and potassium currents, shortening of atrial refractory periods leading to promotion of AF. (6)

Multiple mechanisms of AF have been proposed, including multiple wavelet reentry, focal firing and stable “mother rotor” reentry. The mechanism of AF in the setting of atrial fibrosis remains unclear. High-density electrical mapping demonstrates that AF with structural remodeling is characterized by a stable high-frequency area as opposed to AF with electrical remodeling of atria which is characterized by multiple high-frequency wavelets. (21) However, multiple mechanisms may be present in the same model. In addition to stable reentry, focal activity is also present in dogs with CHF. Fenelon et al (26) performed biatrial mapping in dogs with CHF and showed that the majority of AF episodes had a focal mechanism and was successfully eliminated by focal radiofrequency catheter ablation. Okuyama et al (27) performed high-density mapping of the pulmonary veins during AF in dogs with CHF and found that focal activation originated from within pulmonary veins. Stambler et al (28) suggested that focal AF in the setting of CHF is caused by triggered activity produced by delayed
afterdepolarizations and initiated by intracellular Ca2+ overload. This triggered activity is promoted by alterations of calcium handling with CHF. (28) However, triggered activity may also promoted by atrial fibrosis due to alteration of sink-source relationships caused by the separation of myocyte bundles by interstitial fibrosis.

**Mechanisms of Atrial Fibrosis**

The precise mechanisms of atrial fibrosis and whether there are atrio-ventricular differences in susceptibility to fibrosis remain unclear. Nevertheless, it appears that the atrium is more susceptible to atrial fibrosis than the ventricle. There are currently three interrelated pathways involved in atrial fibrosis—the renin–angiotensin system, TGF-β1, and the oxidative stress pathways. These are activated from a variety of cardiac insults and often work together to promote fibrosis in clinical syndromes with AF. (29)

*The renin-angiotensin-aldosterone system*

The renin-angiotensin-aldosterone system is involved in myocardial fibrosis in hypertensive heart disease, CHF, myocardial infarction, and cardiomyopathy. (30) Patients with primary hyperaldosteronism have an increased incidence of AF. (31) Increased angiotensin II production in transgenic mice with cardiac-restricted angiotensin-converting enzyme (ACE) overexpression causes marked atrial dilation with fibrosis and AF. (32) Atrial angiotensin II levels increase in experimental CHF. (33) Mitogen-activated protein kinases are important downstream mediators of angiotensin II effects on tissue structure, (34) and alters gap-junctional coupling in a way that may promote AF. (35)
Transforming growth factor-β1

Transforming growth factor-β1 is the primary downstream mediator of angiotensin II effects. (36) Angiotensin II induces TGF-β1 expression. (37) Blockade of the angiotensin II type 1 (AT1) receptor suppresses TGF-β1 upregulation. (38) TGF-β1 acts through SMAD signaling pathway to stimulate collagen production. (39) There is an increase in atrial expression of activated TGF-β1 in experimental CHF. (40) Targeted cardiac overexpression of TGF-β1 causes selective atrial fibrosis, conduction heterogeneity, and AF propensity. (41,42) There is normal ventricular structure and function in this model despite equal overexpression in the atria and ventricles, implying that there are differences in atrial versus ventricular vulnerability to fibrosis. The mechanism of atrial vulnerability is unclear but may be related to platelet derived growth factor signalling. (43)

Differences between atrial and ventricular fibrosis

In a variety of cardiac disease models, atrial fibrillation is much more prominent than fibrosis in the ventricles, even when the pro-fibrotic stimulus operates comparably at both sites. There may be several explanations for this. Firstly, atrial tissue is composed of a larger proportion of non-myocytes (predominantly fibroblasts) by volume than the ventricle, (44) a difference which is further exaggerated by congestive heart failure. (40) Secondly, atrial fibroblasts have a greater tendency to activated myofibroblast differentiation both in vitro and in vivo. (43) Thirdly, atrial fibroblast proliferation and gene expression of fibroblast-selective markers were greater than ventricular fibroblast
proliferation in response to a variety of growth factors. (43) Gene microarrays identified platelet-derived growth factor as a potential contributor to atrial-ventricular fibroblast differences. (43) Platelet-derived growth factor inhibition eliminated atrial-ventricular fibroblast proliferative response differences. (43)

Platelet-derived growth factor is a member of the PDGF/vascular endothelial growth factor family and may provide the crucial link between inflammation and atrial vulnerability to fibrosis. It is highly expressed in the myocardium throughout development and adulthood and stimulates proliferation, migration, differentiation, and physiological function of mesenchymal cells. (45) Pressure overloaded mice developed atrial fibrosis and increased susceptibility to atrial fibrillation due to PDGF mediated mast cell activation. (45) This effect was attenuated by mast cell stabilizers and neutralizing PDGF alfa receptor antibodies and suppressed by BM reconstitution from mast cell deficient mice. (45) The data shows that atrial fibrosis and AF susceptibility is promoted by inflammation via PDGF related mechanisms.

**Cellular mediators of fibrosis**

Fibrosis results when circulating and locally synthesized pro-fibrotic factors act on resident cardiac cells to increase collagen production without offsetting increases in collagen degradation. Cardiomyocytes account for approximately 45% of the atrial myocardium by volume. (44) Non-myocytes consist of fibroblasts predominantly, but also vascular endothelial cells and migrating leucocytes. (44) The fibroblast was traditionally thought to be a passive bystander in the myocardium, but now recognized to be play an active role in normal cardiac structure and function. (46) In response to
autocrine-paracrine mediators, mechanical stretch and inflammation, fibroblasts proliferate and undergo phenotypic change into myofibroblasts. Myofibroblasts produce growth factors, cytokines, chemokines, ECM proteins and proteases and are thus important in the pro-fibrotic process. (46) It has been demonstrated that fibroblasts establish functional electrical connections via Cx45 gap junctions with myocytes within the rabbit sinus node, indicating that cardiac connective tissue may be involved in impulse conduction in vivo. (47) It is conceivable that similar electrical connections may occur between atrial myocytes and fibroblasts, thus contributing to atrial electrical heterogeneity and AF substrate.

Role of inflammation in AF and atrial fibrosis.

Clinical investigations have reported associations between circulating levels of cytokines, C-reactive protein, complement and the activation state of leukocytes and AF. (48,49) Whether inflammation is an epiphenomenon in AF or causally related to AF initiation and maintenance remains unclear. A key mechanistic link between AF and inflammation is atrial fibrosis. Increased deposition and turnover of matrix proteins within the atria is accelerated in atrial myocytes after challenge with cytokines, C-reactive protein and complement. A key mediator in extracellular matrix turnover is matrix metalloproteinases. (50) The activity of MMPs is regulated by myeloperoxidase, a leukocyte-derived heme enzyme which generates reactive oxygen species which in turn activates MMPs. When MPO-deficient mice were treated with angiotensin II to promote leucocyte activation, there was reduced activity of MMPs, blunted atrial fibrosis and protection from AF. (51) These findings were further corroborated in humans with AF.
who were found to have higher plasma concentrations of MPO and a larger MPO burden in right atrial tissue compared to individuals without AF. (51)

**Therapeutic Implications**

Conventional anti-arrhythmic drug therapies have limited effectiveness and are associated with pro-arrhythmia and side effects. (52) Accordingly, attenuation of structural remodeling, so called upstream therapy, have increasingly become the focus of therapeutic developments. These include angiotensin-converting enzyme and angiotensin-receptor inhibitors, statins, or omega-3 fatty acids and fish oil that target atrial remodelling. Several retrospective clinical studies demonstrated a decreased incidence of AF in patients treated with ACE inhibitors or angiotensin-receptor blockers, mostly in the setting of LV dysfunction. (53-58) However, the large placebo-controlled GISSI-AF trial (59) showed that valsartan did not reduce recurrence rates of AF in patients with pre-existing cardiovascular diseases. *The study was performed on patients with hypertension and AF who were already receiving established therapies for AF.* While this raises questions about the value of angiotensin receptor blockers for secondary prevention of AF, the negative result may be attributable to the fact that intervention was started after atrial remodelling was established and the short 1 year follow up was insufficient time for the beneficial effects on atrial remodeling to manifest. In addition, a significant proportion of patients in both groups were taking ACE inhibitors. Further prospective studies are therefore needed to establish the therapeutic value of ACE inhibitors and angiotensin receptor blockers in prevention of AF. *In particular, future studies should focus on patients with frequent paroxysmal AF prior to the development of*
structural remodeling and persistent AF and patients with left ventricular dysfunction who are at risk of AF.

Aldosterone is produced by the heart and exerts many cardiac effects. Plasma aldosterone concentrations are elevated in patients with AF. (60) Patients with primary hyperaldosteronism have a greater risk of AF than blood pressure matched controls. (31) In small animals, aldosterone causes atrial fibrosis and spironolactone prevents fibrosis, (61) and suppresses AF in experimental heart failure. (62) However, data from clinical trials are not available. The effect on AF of aldosterone-receptor blockers alone or in combination with ACE inhibitor or angiotensin-receptor blockers is an area that deserves further investigation in a clinical trial setting. Pirfenidone, 5-methyl-1-phenyl-2(1H)-pyridone, an anti-fibrotic agent, reduces TGF-β1 levels and prevents development of the AF substrate in experimental congestive heart failure. (63)

Atrial tissue inflammation contributes to atrial arrhythmogenic remodelling and AF. Corticosteroids prevents AF in animal and clinical studies, (64,65) although their potential toxicity prevents their widespread clinical use. Both statins and omega-3 fatty acids have anti-inflammatory and anti-oxidant effects. Statins are effective against several substrates that maintain AF, (66,67) and may be effective against postoperative AF. (68) The effects of omega-3 fatty-acids on AF remains unclear. (69) Animal studies (70) show model-dependent AF-preventing effects, and may be effective especially in patients at risk of fibrotic structural remodelling.
Conclusion

There is a strong association between atrial fibrosis and AF in the both clinical and experimental settings. Atrial fibrosis causes abnormal conduction through the atria, creating a substrate for AF. The mechanism of AF produced by atrial fibrosis is controversial, as both macroreentry and focal sources originating from atria and pulmonary veins have been demonstrated. Molecular pathways involved in atrial fibrosis are beginning to emerge. The renin–angiotensin system and TGF-β1 play an important role in atrial fibrosis, with the atrium being more susceptible to fibrosis than the ventricle. Conventional upstream therapies with ACE inhibitors or angiotensin receptor blockers may be useful adjunctive therapies for AF, but their benefit in primary versus secondary prevention needs to be further defined. Identifying downstream mediators of fibrosis, specifically in the atrium, will be important in developing new anti-fibrotic strategies.
Disclosures

None

Reference List


5. S. Willems, H. Klemm and T. Rostock et al., Substrate modification combined with pulmonary vein isolation improves outcome of catheter ablation in patients with


33. D. Li, K. Shinagawa and L. Pang et al., Effects of angiotensin-converting enzyme inhibition on the development of the atrial fibrillation substrate in dogs with


46. Powell DW, Mifflin RC, Valentich JD, Crowe SE, Saada JI, West AB.


