JACC: CLINICAL ELECTROPHYSIOLOGY
VOL. 1, NO. 3, 2015

ISSN 2405-500X/$36.00
PUBLISHED BY ELSEVIER INC.
http://dx.doi.org/10.1016/j.jacep.2015.03.011

# Thrombogenic Risk in Patients With Atrial Fibrillation

## Importance of Comorbid Conditions and Intracardiac Changes

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## ABSTRACT

**OBJECTIVES** This study sought to determine the differences between the prothrombotic properties and chamber characteristics in patients with lone atrial fibrillation (AF) and those with AF and comorbidities.

**BACKGROUND** Thromboembolic risk is increased in patients with AF; however, whether this is due to AF per se or its comorbidities remains unclear.

**METHODS** A total of 87 patients undergoing ablation were prospectively recruited for the study, including 30 patients with lone AF, 30 patients with AF and comorbidities in sinus rhythm, and 27 patients with left-sided accessory pathways as controls. Blood samples were obtained from the left atrium (LA), right atrium (RA), and femoral vein (FV) after transseptal puncture. Platelet activation (P-selectin) was measured by flow cytometry. Thrombin generation (thrombin-antithrombin [TAT] complex), endothelial dysfunction (asymmetric-dimethylarginine [ADMA]), and platelet-derived inflammation (soluble CD40 ligand [sCD40L]) were measured using enzyme-linked immunosorbent assay.

**RESULTS** Platelet activation in the LA was significantly elevated compared to that in the FV in patients with lone AF and those with AF and comorbidities compared with that in the FV ($p < 0.05$ respectively). Thrombin generation was significantly elevated in the LA compared with RA in AF patients ($p < 0.05$). There were no significant differences in P-selectin, TAT, and sCD40L among the 3 groups. However, there was a significant stepwise increase in endothelial dysfunction measured by ADMA from controls to lone AF and then to patients with AF and comorbidities ($p < 0.001$ between the 2 groups).

**CONCLUSIONS** Patients with lone AF and those with AF and comorbidities had a greater propensity for atrial thrombogenesis than controls. Prothrombotic risk is greatest in those with comorbid conditions, in whom enhanced thrombogenesis occurs predominantly through increase in endothelial dysfunction. (J Am Coll Cardiol EP 2015;1:210–7)


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Manuscript received January 27, 2015; revised manuscript received February 17, 2015, accepted March 26, 2015.

The most devastating complication associated with atrial fibrillation (AF) remains thromboembolic stroke, with a 5-fold increased risk in patients with nonvalvular AF (1). Patients with AF are known to exhibit a prothrombotic state, endothelial dysfunction, and abnormal left atrial blood flow, thus fulfilling Virchow's triad for thrombus formation (2–7). Abnormal platelet activation has been demonstrated peripherally in patients with nonvalvular AF (8,9), and peripheral blood samples have reflected endothelial dysfunction in different subsets of AF patients compared with controls (8,9).

SEE PAGE 218

However, there is ongoing debate as to whether these changes are due primarily to AF per se or to its associated risk factors. Several studies have suggested that in patients with AF, the prothrombotic state may be the result of concurrent comorbidities such as hypertension, diabetes, and coronary artery disease (10,11). On the other hand, other reports suggest a heightened risk of thrombosis even in patients with lone AF (8,9). Moreover, recent studies have shown that peripheral sampling may not adequately reflect intracardiac changes within the heart (4,5,12). These previous, conflicting data may be partly due to sampling from heterogeneous cohorts or sampling from peripheral versus intracardiac blood (8–11).

In this study, we examined these issues by studying both patients with lone AF and patients with AF and comorbidities, using sampling from both atrial and peripheral blood. We investigated prothrombotic properties (platelet activation, thrombin generation, endothelial dysfunction, and platelet-derived inflammation) within the left atrium (LA), right atrium (RA), and femoral vein (FV) in consecutive patients with lone AF, with AF in the setting of comorbidities, and in controls to determine the relative contribution of these factors to the thrombogenic process.

## METHODS

A detailed description of methods is found in the Online Appendix.

**STUDY POPULATION.** Consecutive patients undergoing catheter ablation for AF at the Centre for Heart Rhythm Disorders, Royal Adelaide Hospital, were screened. Patients were studied provided they had no history of symptomatic AF in the week prior to the procedure and, by continuous monitoring for 48 h immediately prior to ablation, demonstrated no arrhythmia lasting >30 s. This rigorous screening was undertaken to minimize the impact of a recent episode of AF on the patient's prothrombotic state. Patients were also excluded if they had an acute coronary syndrome, surgery or ablation within the preceding 3 months, chronic inflammation or infection, chronic renal or liver disease, or were taking antiplatelet therapy. All patients provided written informed consent to the study protocol, which was approved by the Human Research Ethics Committee of the Royal Adelaide Hospital, Australia.

Sixty patients undergoing catheter ablation for AF who presented in sinus rhythm were prospectively recruited (30 with lone nonvalvular AF and 30 with AF and comorbidities). The control group consisted of 27 prospectively recruited patients with left-sided accessory pathways who underwent ablation during the study period. In addition, a separate reference group of 30 age-matched subjects was recruited to control for the effect of age.

Lone AF was defined, according to previous criteria, as AF in the absence of structural heart disease, hypertension, diabetes mellitus, coronary artery disease, or stroke based on history, physical examination, chest radiography, routine biochemistry, and echocardiography (13).

**STUDY PROTOCOL.** The technique used for ablation of AF in our laboratory has been previously described and is further described in the Online Appendix (7,13,14). For the clinical procedure, a conventional single transseptal puncture was performed using an SLO sheath (St. Jude Medical, St. Paul, Minnesota) and a BRK-1 needle (Daig Corp., Minnetonka, Minnesota). Following transseptal puncture and before intravenous administration of unfractionated heparin, blood samples were simultaneously collected from the peripheral femoral venous sheath (FV, peripheral sample), right atrial sheath (RA) and left atrial sheath (LA). Samples from the RA and LA were collected with care using a slow withdrawal technique with the sheath positioned in the mid chamber, as previously described (4). No ablation was performed prior to completion of the study protocol. In control patients undergoing electrophysiological study and ablation of a left-sided accessory pathway, LA, RA, and FV samples were obtained immediately after transseptal puncture and before administration of heparin. Only peripheral samples were obtained for the age-matched reference group.

**WHOLE-BLOOD FLOW CYTOMETRY.** The surface expression of the platelet activation receptor CD62P (P-selectin) was determined by flow cytometry, using specific monoclonal antibodies (4,7). All

**ABBREVIATIONS AND ACRONYMS**

**ADMA** = asymmetric dimethylarginine
**AF** = atrial fibrillation
**FV** = femoral vein
**LA** = left atrium
**NO** = nitric oxide
**RA** = right atrium
**TAT** = thrombin-antithrombin complex