Atrial fibrillation and hypercoagulability

Impact

Atrial fibrillation (AF) is the most common form of sustained cardiac tachyarrhythmia. It often starts asymptomatically, which allows it to progress until it causes adverse cardiovascular events (e.g. thromboembolic stroke), before it is even detected. AF prevalence increases with age. Due to ageing of the general population in the coming decades, the burden on the worldwide health care systems is expected to increase significantly. New insights on mechanisms that affect the initiation and progression of AF are therefore highly needed.

Previously, it has been demonstrated that inhibition of specific coagulation factors may reduce pathological processes that are involved in AF substrate development. In this context, the general aim of our thesis was to evaluate how hypercoagulability is linked to AF progression. Furthermore, we investigated the effect of two main AF risk factors (age and heart failure [HF]) on AF substrate development and coagulation activity.

Our *in vitro* experimental work as described in chapter 3 focused on one of the most important cell types in the heart, cardiac fibroblasts (CF). We proved that the coagulation factors thrombin and FXa activate CF and promote pro-fibrotic processes. Moreover, we have shown that FXa has a pronounced pro-inflammatory effect on CF, which appears to be mediated by PAR-1 activation and inhibited by the FXa-direct inhibitor rivaroxaban. These findings again demonstrate that coagulation factors elicit direct non-hemostatic functions. This observation could also support other researchers to extend the study of the pleiotropic effects of these coagulation proteins not only in the heart but also in other organs of the body.

A major part of this thesis was based on *in vivo* investigations making use of the goat model. This model is a well-established and extensively researched model for the pathogenesis of AF. Because AF can be maintained in the goat for several months, this animal model also represents an interesting tool to investigate the long-term effects of AF on the coagulation system. However, suitable coagulation assays for goat plasma are not available from commercial sources. In chapter 4 we described how we customized the Calibrated Automated Thrombography (CAT) assay to provide a global view of the goat coagulation profile by assessing thrombin generation (TG) in goat plasma. To the best of our knowledge, this is the first study that reports the customization of the CAT assay for goats. In future applications of the AF goat model, in fact for any (experimental) goat model, the main changes in the coagulation system can now be included for evaluation. For instance, other interesting research questions, which can be now answered, may focus on the effect that the presence of other AF-related comorbidity, such as hypertension, in combination with AF, would have on the coagulation system, or on how coagulation activity

would be affected by paroxysms of AF and/or restoration of sinus rhythm after a prolonged period of AF. Moreover, additional modifications of the CAT assay can give insights on the contribution of both pro- and anti-coagulant pathways.

The possible link between pathological activation of the coagulation system and cardiac remodeling was further supported by chapter 5. Using the customized CAT assay, we showed that anticoagulation treatment via the DOAC rivaroxaban, prevented atrial cardiomyocyte hypertrophy induced by four months of AF. These findings are potentially important for clinical considerations. In fact, they provide insights on the pleiotropic effects of activated coagulation factors in the atrial myocardium and on the fact that their direct inhibition via (clinically approved) DOACs (in this case rivaroxaban), may not only prevent the risk of stroke in AF patients but also prevent cardiac remodeling processes (e.g. inflammation, fibrosis and myocyte hypertrophy). Clinical trials on the effect of DOACs on outcome in patients with AF (e.g. stroke, cognitive decline, cardiovascular outcome) offer opportunities to test this hypothesis. For example, the ongoing NOAH-AFNET 6 trial investigates the effect of DOACS (edoxaban) on stroke and cardiovascular complication in low risk patients with atrial high rate episodes but without documented AF. The control group does not receive anticoagulation and using the implanted devices an effect of the DOAC on progression to AF can be quantified.

The findings of chapter 6 highlighted the importance of the synergistic effect of AF and its risk factors (in this case age) on coagulation activity and AF substrate development, while in chapter 7 we showed that the lone presence of the AF risk factor HF strongly increased the AF substrate, which was associated with a substantial decrease of clotting potential. Although a causal relation between the clotting potential and atrial remodeling could not be proven in the studies, we believe that the knowledge acquired can be the basis for future investigations, nonetheless for important translational considerations.

Taken together, our findings support the relation between pathological activation of the coagulation system and atrial remodeling. The methods and findings described in this thesis are of potential value to all researchers involved in AF, its risk factors or coagulation and can be of importance to clinical considerations. In fact, based on the effect that activated coagulation factors elicit on the atrial myocardium, our data support the thought that early anti-coagulation therapy in AF may not only mitigate the risk of stroke but also slow down AF progression.