Functional interactions between factor V and TFPIα during onset of blood coagulation

ADDENDUM

Valorisation

VALORISATION

This chapter is intended to show how the knowledge obtained in this thesis is relevant outside a scientific context.

Valorisation - "*The process of value-creation out of knowledge, by making this knowledge suitable and available for economic or societal utilisation and to translate this into high-potential products, services, processes and industrial activity.*" [1]

The societal impact of a research project is often obvious when it produces a product or solution that can readily be utilised for an individual or group. This type of applied research is however not possible without prior knowledge about the basic mechanisms, which is obtained from fundamental research. Fundamental research does not have a commercial objective, as it is driven by a desire to test hypotheses and obtain new knowledge. As a result the societal impact may not be obvious at first, when only a small piece of the puzzle is found. The research can however end up being a center piece that connects other pieces of the puzzle.

The fundamental research on the underlying mechanisms of coagulation is pivotal for the development of novel drugs and treatments of venous thrombosis (VT). There is still much to be achieved in this area as VT is the third most common cardiovascular disease, right after coronary artery disease and stroke [2]. Patients suffering from VT carry an increased morbidity and mortality. A venous thrombotic event may lead to pulmonary embolism with death as a major outcome. These patients are highly dependent on long-term treatment, which currently still carries strong side effects such as an increased bleeding risk. The prevalence of VT rises sharply as a person's age rises [3]. With increasing life expectancy it can be predicted that VT will have an even stronger impact on society in the near future. Therefore much is to be gained with a better understanding of the mechanisms of blood coagulation, which in turn can lead to improved and targeted treatments.

The work in this thesis focusses on gaining a better understanding about the interaction between coagulation factor V (FV) and tissue factor pathway inhibitor-α (TFPIα). While this curiosity-driven research started off as fundamental in design, its results have already lead to the development of an assay for the detection of possible risk factors in VT, as explained below.

In **Chapter 2** we found that FV has anticoagulant properties in the presence of TFPIα. Increasing FV from 0 to 10% of normal FV levels increased thrombin generation, while a further increase up to 100% FV gradually decreased thrombin generation. These results imply that partial FV deficiency increases the risk on thrombosis. In fact, low FV levels have been associated with increased risk of thrombosis and a patient with recurrent thrombotic episodes due to low FV

levels has recently been described [4]. *Vice* versa, besides FV affecting TFPIα, TFPIα also affects FV. **Chapter 3** describes how the interaction between the C-terminus of TFPIα and FV prevents FV from reaching its full procoagulant potential. Insights in these mechanisms will benefit future research as the inhibition of TFPIα is currently of high interest as a possible bypassing agent for hemophilic patients with factor VIII inhibitors [5].

The interaction between FV and TFPIα was found to differ between various forms of FV. Certain FV mutations decreased the affinity for TFPIα, while other FV variants had an increased affinity. We therefore set out to develop a functional assay that could measure the susceptibility of different FV variants to inhibition by TFPIα. This assay, described in **Chapter 4**, provides a valuable tool to measure this newly discovered property of FV. The TFPIα susceptibility assay can be used in population studies to determine if the susceptibility of FV to TFPIα correlates with a risk on thrombosis or bleeding.

The interactions between FV and TFPIα as described in Chapters 3 and 4 are likely caused by the splicing isoform FV-short, which has a much higher affinity to TFPIα than full-length FV. FV-short levels are thought to vary greatly between individuals and are likely to be important determinants of the TFPIα levels. Increased levels of FV-short have already been associated with a bleeding phenotype [6]. It is however unknown if lower FV-short levels are also a risk factor for thrombosis. So far there are no assays for the quantification of FV-short. We therefore set out to develop the first functional assay for the detection of FV-short, as described in **Chapter 5**. This assay will in the future provide a valuable tool for the identification of the determinants of FV-short and for the correlation of FV-short levels with clinical end-points.

We are confident that the information obtained and described in this thesis provides a valuable contribution to the existing knowledge and will benefit future (applied) thrombosis research.

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