MICRO RNA 181B REGULATES THE DIFFERENTIAL EXPRESSION OF TF ISOFORMS BY BINDING TO TOPOISOMERASE I AND THEREBY LINKS VASCULAR INFLAMMATION TO THROMBOGENICITY.

Poster Contributions
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Background: Endothelial cells (ECs) promote a complex interplay between proinflammatory and prothrombogenic stimuli. Inflammatory cytokines, such as TNFα, have been shown to up-regulate TF, the primary initiator of the extrinsic coagulation. It is known that inflammation regulates the differential TF isoform expression via Topoisomerase I (Topo I) and alternative splicing. Here, we investigate the impact of micro RNAs on TF isoform expression (procoagulatory full length (fl) and angiogenic alternatively spliced (as) TF) and thrombogenicity.

Methods: Human ECs were stimulated with TNFα and assessed for differentially expressed miRNAs. Candidate miRNAs were studied regarding their effect on TF alternative splicing. Data were confirmed by using mimics and antagonists. In addition, myocardial biopsies from 57 patients with suspected inflammatory cardiomyopathy were probed for miRNA and TF isoform expression.

Results: Upon stimulation with TNFα miR-181b and let-7b were differentially expressed. Following TNFα treatment miR-181b caused a decrease in the flTF/asTF ratio on mRNA and protein level (p<0.001), thereby reducing the TF activity in HMEC-1 (p<0.001). Topo I 3’UTR was identified as a target for miR-181b. Upon stimulation with TNFα miR-181b was associated with a down regulation of Topo I mRNA and protein (p<0.001), which altered SR protein phosphorylation. Moreover, myocardial biopsies obtained from 57 patients were divided into a group with high and low TNFα mRNA levels (p<0.001). In the group with high TNFα expression miR-181b expression was significantly increased compared to the group with low TNFα mRNA levels (p<0.05). Increased miR-181b levels were associated with a higher flTF/asTF ratio compared to low miR-181b levels. Conversely, given high TNFα levels miR-181b was correlated with a lower flTF/asTF ratio.

Conclusion: This study shows, for the first time, the role of miRNAs for alternative splicing by interacting with Topoisomerase. MiR-181b was found to alter blood coagulation by regulation of TF isoform expression. In conclusion, miR-181b signalling links vascular inflammation with coagulation and may be involved in pathological vascular biology.