Collagen type VI is a ubiquitous extracellular matrix component that forms complex and extensive microfibrillar networks in all connective tissues, often associated with basement membranes. Structurally, it consists of three α-chains (α1, α2 and α3), where each α-chain contains a short triple helix and N- and C-terminal globular regions. More recently, three additional chains (α4, α5 and α6) were discovered, which may substitute for α3 in some tissues. The globular regions are homologous to the type A domains of von Willebrand factor (vWF-A). We have recently described the antimicrobial properties of this collagen against a number of Gram-positive and Gram-negative human pathogens. However, the molecular mechanisms of bacterial killing are still elusive. Therefore, in this study, we applied an in silico approach to unravel the antimicrobial activity of collagen type VI in further detail. Sequence and structural analysis showed that the vWF-A domains of all three α-chains contain numerous amphipathic amino acid motifs of putative antimicrobial nature. In addition, we also could show that recombinantly expressed vWF-A domains bind to negatively charged surfaces such as heparin and bacterial membranes. Five such motifs were finally chosen from N8, N9 and C1 domains of the α3- chain for further characterization in bacterial killing assays. The data suggest that amphipathic, heparin-binding amino acid motifs in the globular vWF A-like domains harbour the antimicrobial properties of collagen VI.

Acknowledgement: The authors gratefully acknowledge the skilful work of Maria Baumgarten. We wish to thank the Core Facility for Integrated Microscopy (CFIM), Panum Institute, University of Copenhagen, for providing an excellent electron microscopy platform/environment.
Fig. 1: *S. pyogenes* (AP1) were treated with α1, α2 or α3 chain of collagen VI and permeabilization was visualized by using scanning electron microscopy. Extensive membrane disruption and leakage of intracellular contents are observed in the presence of these proteins and are indicated with arrowheads. Bar represents 5 µm.