Diagnosis and understanding of chronic infections. Remember the biofilm

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Title: Diagnosis and understanding of chronic infections. Remember the biofilm

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Aim: Recent evidence suggests that the microbial community, its spatial distribution and activity play an important role in the prolongation of treatment and healing of chronic infections. The purpose of this study was to improve sampling, diagnosis and treatment of prosthetic joint infections (PJI) and chronic wounds, especially considering the biofilm issue.

Method: Systematic and optimized sampling of various specimen types, sonication of prosthesis and extended culture were applied on patients with chronic wounds and PJI patients. Optimized DNA extraction, quantitative PCR, PNA FISH, next generation sequencing and bioinformatics tools were applied on different types of specimens for optimized diagnosis. For further investigation of the microbial pathogenesis, *in situ* transcriptomics and metabolomics were applied.

Results / Discussion: In both chronic wounds and PJIs, molecular techniques detected a larger diversity of microorganisms than culture methods in several patients. A heterogeneous distribution of bacteria in various specimens from the same patient was evident for both patient groups. In chronic wounds, multiple biopsies from the same ulcer showed large differences in the abundance of fx *P. aeruginosa* at different locations. Transcriptomic and metabolomic analyses indicated the important virulence genes and nutrient acquisition mechanisms of *S. aureus in situ*.

Conclusion: Our studies show that diagnosis of chronic biofilm related infections required multiple specimen types, standardized sampling, extended culture and molecular analysis. Using a well-designed diagnostic algorithm has the potential for making personalized diagnosis and treatment of biofilm related infection possible. It is our ambition to translate studies on bacterial activity into clinical practice.

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