University of Basel

DBM Basel
Department of Biomedicine

TRANSCRIPTOMIC PROFILING OF HUMAN-*STAPHYLOCOCCUS AUREUS* INTERACTIONS:

Deciphering the survival of intracellular *S. aureus* in human macrophages under clinically relevant antibiotic exposure using dual RNA-sequencing, microscopy and proteomics

Project Background

Deep-seated *Staphylococcus aureus (S. aureus)* infections cause major morbidity and mortality. Despite appropriate antibiotic therapy guided by susceptibility testing, treatment failure rates remain high, ranging from 20% to 50%. Novel and more effective therapies are thus urgently needed. While *S. aureus* is usually not considered an intracellular pathogen, recent studies have demonstrated its ability to survive within macrophages, potentially contributing to persistent infections and treatment failures.
In our laboratory, we performed dual RNA-Seq analysis on fresh biopsies obtained directly from patients undergoing surgery for *S. aureus* deep-seated infections. By comparing healthy and infected tissues from the same patient, we identified up-regulation of genes involved in response to bacteria, humoral immune response and phagocytosis specifically at the site of infection. Moreover, analysis of bacterial transcripts within the infected site indicated downregulation of metabolic and virulence genes supporting the hypothesis of dormant intracellular S. aureus. In parallel, we optimized an *in vitro* model to reproduce the patient setting in which we can evaluate the effect of clinically relevant antibiotics including Levofloxacin, Flucloxacillin, Linezolid and Rifampicin. Microscopy at different time points demonstrated a high level of intracellular *S. aureus* and CFUs counting demonstrated survival under antibiotic exposure.

Master Project

For this project, the student will perform infections of monocytes derived macrophages from healthy donors at different time points using three new and yet, not commercially available drugs. In parallel, microscopy and quantification of intracellular survival will be performed. The student will then learn how to analyse such data sets and will compare it to data from other antibiotics already available in the lab. Based on the results, the student will generate specific mutants for genes that seem relevant to survive intracellularly and under antibiotic pressure. Those mutants will then be used for infection to confirm their potential roles in host-pathogen interactions.

Methods That the Student Will Acquire

Cell culture, PBMCs extraction, monocytes isolation, differentiation of monocytes into macrophages, cell infection, live microscopy, RNA extraction, RNA-sequencing, RNA-Seq data analysis, cloning, flow cytometry, scientific writing…

Illustrative Figure

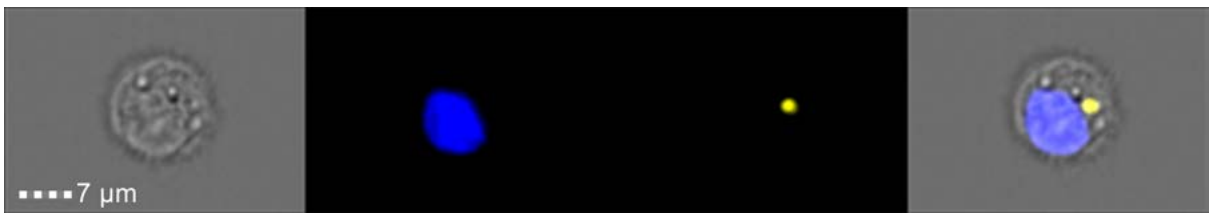

Image of a monocyte infected by *S. aureus* after 1h of infection. *S. aureus* is marked in yellow (JAN-VAN669) the macrophage nucleus in marked in blue (DAPI).

Contact

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