Studies on virtual whole-blood infection assays

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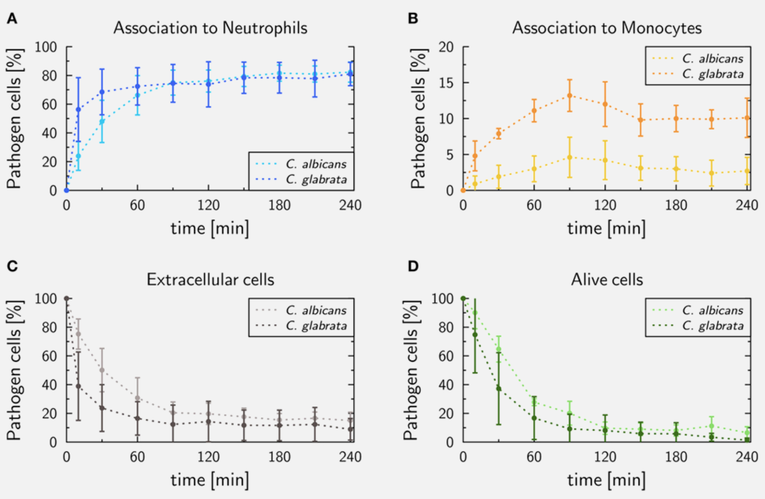
Infection dynamics in healthy individuals and patients

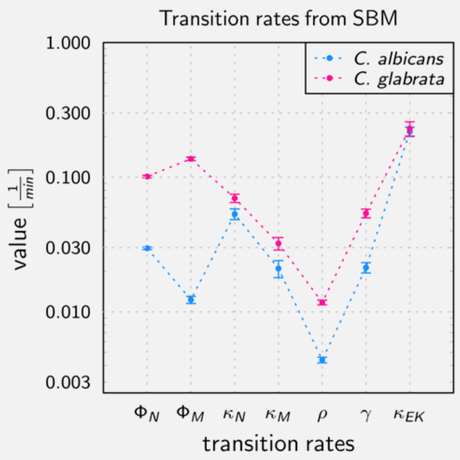
Healthy Individuals:

Experiments: We performed whole-blood infection assays, where blood from healthy donors is infected with *C. albicans* or *C. glabrata* cells. With phagocytosis assays and survival plates we determined pathogen populations of alive, killed, extracellular and phagocytosed cells by monocytes and neutrophils.



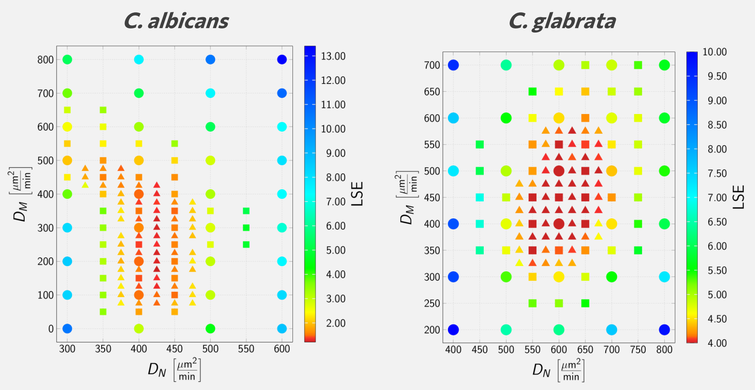
[1,2]

State-based model: The SBM consists of states and transitions between these states that resemble the biological system. Fitting the SBM to the experimental data allowed quantification of immune reaction rates, such as phagocytosis and killing rates.



Rate		Description			
Φ_N	...	phagocytosis rate of neutrophils	ρ	...	pathogen immune evasion
Φ_M	...	phagocytosis rate of monocytes	κ_{EK}	...	secretion of anti-microbial peptides by neutrophils upon first phagocytosis
κ_N	...	killing rate of neutrophils	γ	...	half-life time of anti-microbial peptides
κ_M	...	killing rate of monocytes			

Agent-based model: To investigate also spatial aspects we build an ABM, where single cells are simulated in a continuous three-dimensional environment. Based on the experimental data and the previously estimated rates we could determine diffusion coefficients of neutrophils (D_N) and monocytes (D_M).



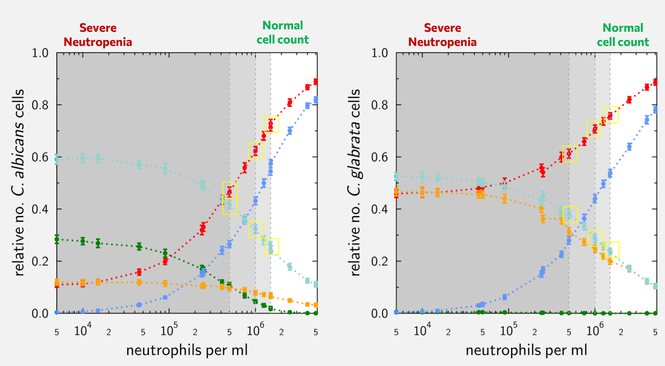
- Higher diffusion coefficients for *C. glabrata*
- Neutrophils play major role in the immune response
- Monoctes are less important for *C. albicans* infections
- *C. albicans*: sensitive to variations in D_N

Virtual Neutropenic Patients (VNP):

[3]

We simulated infection dynamics in VNP with different severity degrees of neutropenia by reducing the number of neutrophils in the ABM. We selected five VNP (see Table) and identified patterns to characterize the infection outcome that were based on the fraction of killed (P_K) and immune evaded pathogens (P_{AIE}).

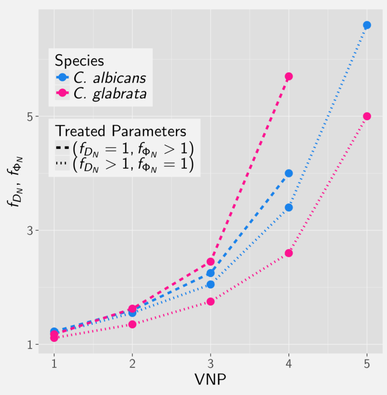
VNP	Severity Degree	Neutrophils/µl
1	mild	1250
2		1000
3	moderate	750
4		500
5	severe	250



In silico Treatment of VNP:

To simulate the *in silico* treatment of VNP we increased the phagocytosis rate Φ_N and diffusion coefficient D_N of neutrophils in a stepwise fashion and compared the infection outcome to the previously determined patterns

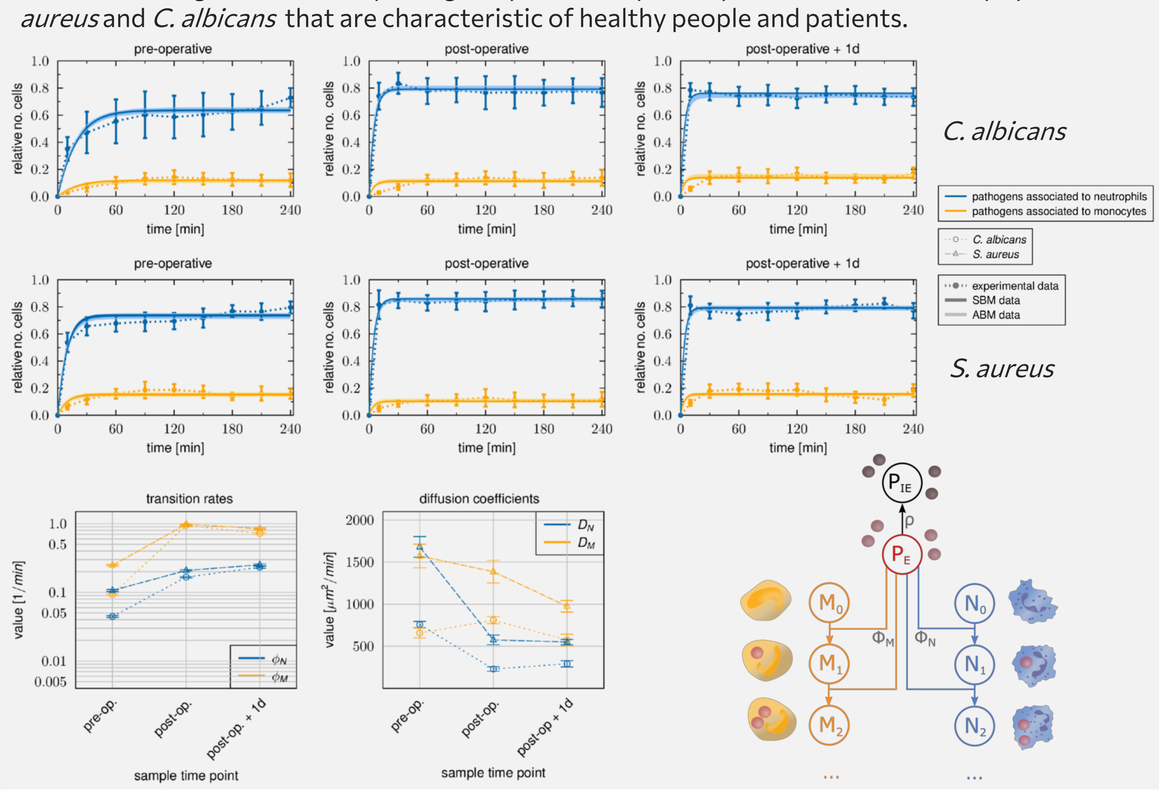
- $f_{\Phi_N} = \frac{\Phi_N^T}{\Phi_N^{min}}$ and $f_{D_N} = \frac{D_N^T}{D_N^{min}}$
- Φ_N^T, D_N^T – value after treatment
- Φ_N^{min}, D_N^{min} – value before treatment



Hospital Patients:

[4]

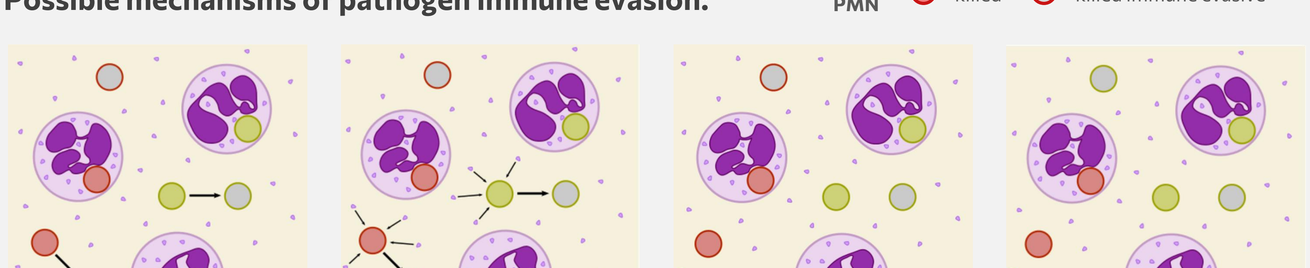
We quantified functional parameters of innate immune cells in blood from patients undergoing cardiac surgery. These patients experience a well-characterized inflammatory insult, which results in mitigation of the pathogen-specific response patterns towards *Staphylococcus aureus* and *C. albicans* that are characteristic of healthy people and patients.

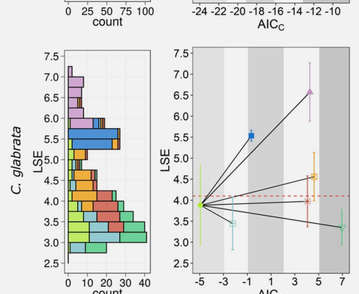


Pathogen immune evasion

Whole-blood experiments with blood from healthy donors and patients revealed that a certain fraction of pathogens remain extracellular. In combination with mathematical modelling we could show that this can only be explained by pathogen immune evasion (IE). However, as of now the underlying mechanism is not understood. Therefore, we compared simulations with various possible immune evasion mechanisms in order to generate hypotheses that allow for targeted experimental investigations.

Possible mechanisms of pathogen immune evasion:





- Model for pre-existing IE could be falsified by automated image-analysis
- Three base models and their combinations
- Least-square error (LSE) to measure agreement with experimental data
- Corrected Akaike Information criterion (AIC_C) as measure for model quality with penalty for number of parameters
- More complex models could not compensate for their penalized complexity by a better agreement with the experimental data
- Model-specific patterns could be derived
- Proposal of new experiments that will contribute to the identification of IE mechanism in the future

[5, 6]

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