Combining wet lab and computational simulations to predict optimal antibiotic drug regimens for *Mycobacterium tuberculosis*

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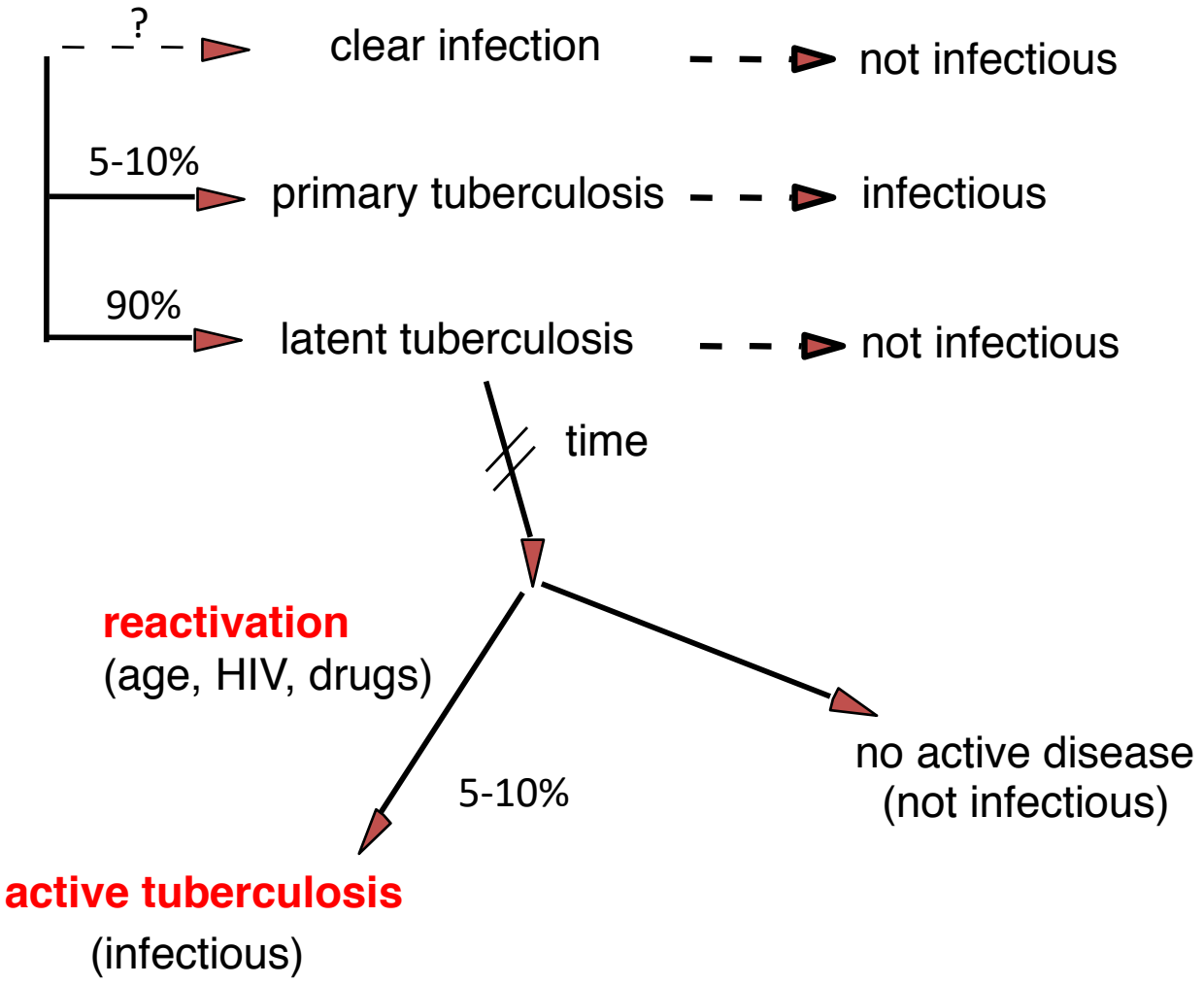
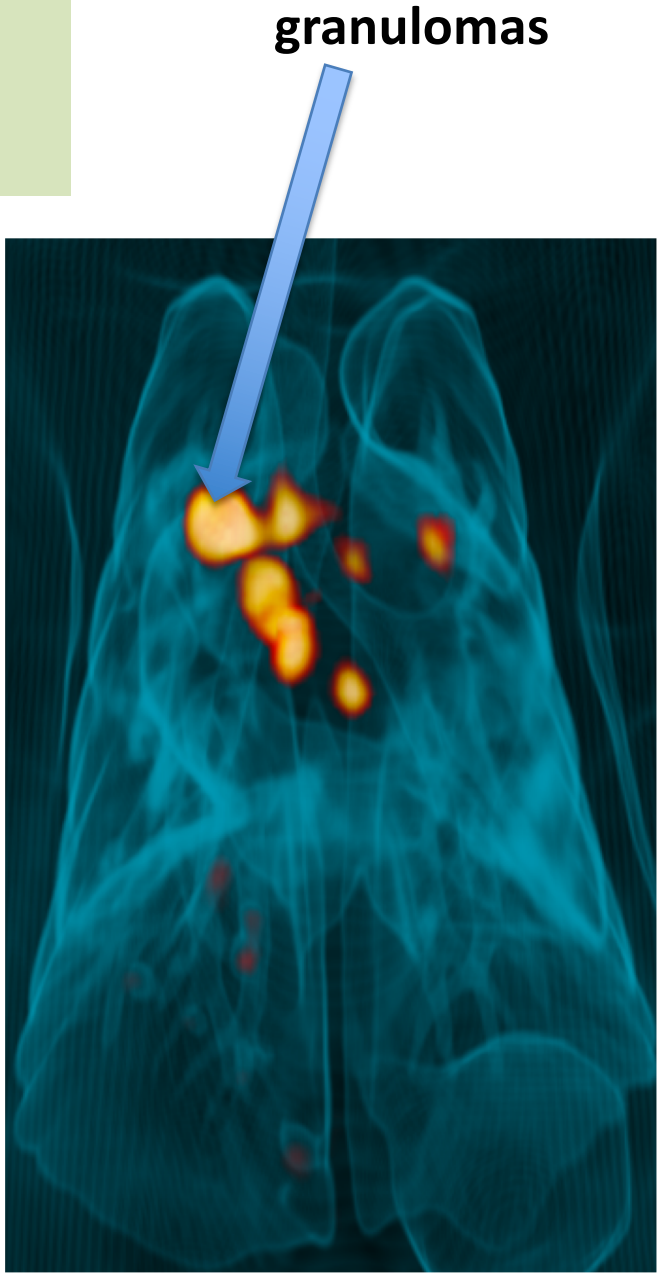
MSM : 1U01HL131072

END TB

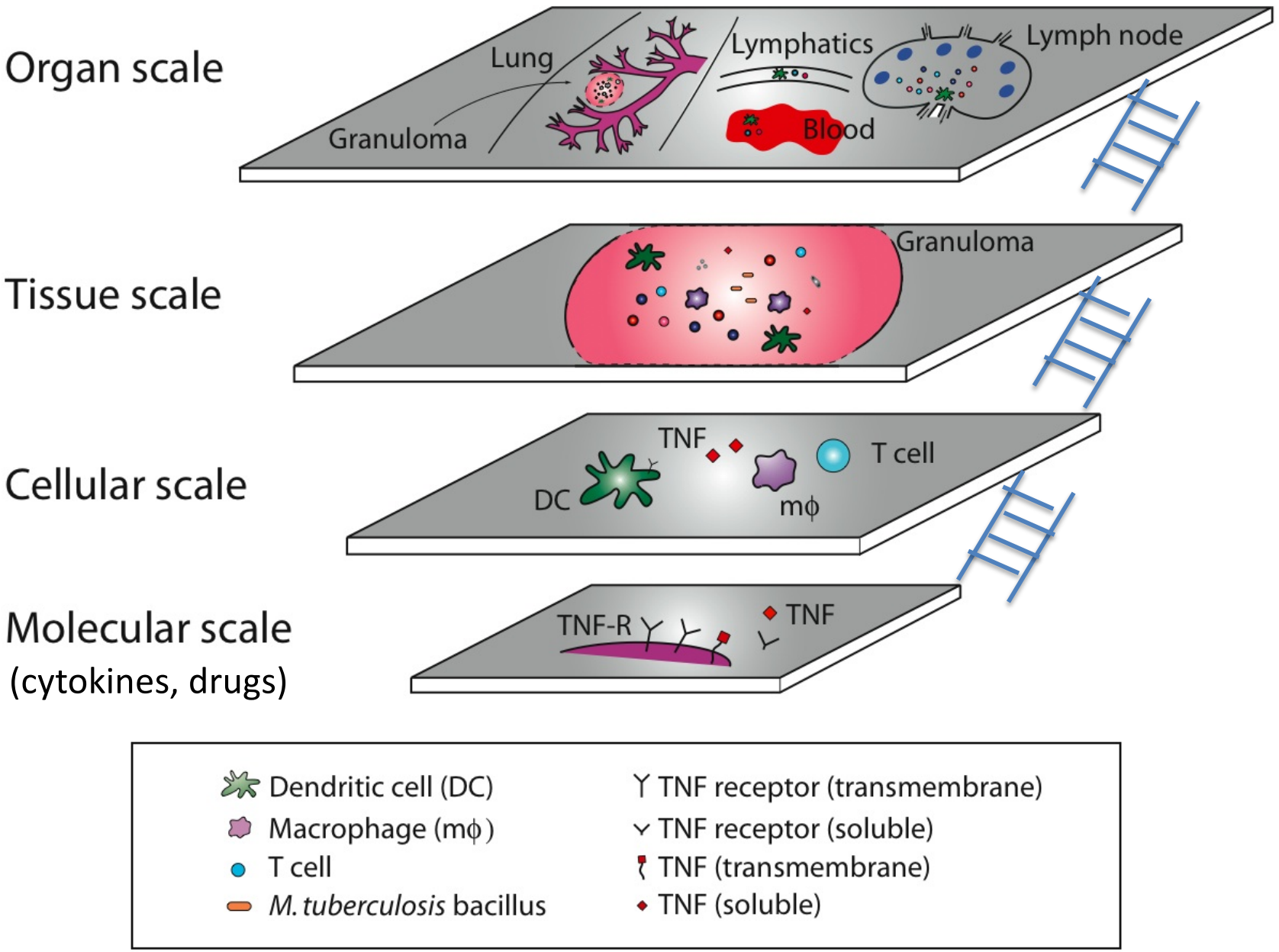
Tuberculosis (TB):

Infectious disease caused by *Mycobacterium tuberculosis* (Mtb).
One–third of the world's population is infected with Mtb, and new infections occur at a rate of one per second.
3 people die every minute, i.e. 2 million deaths/year.





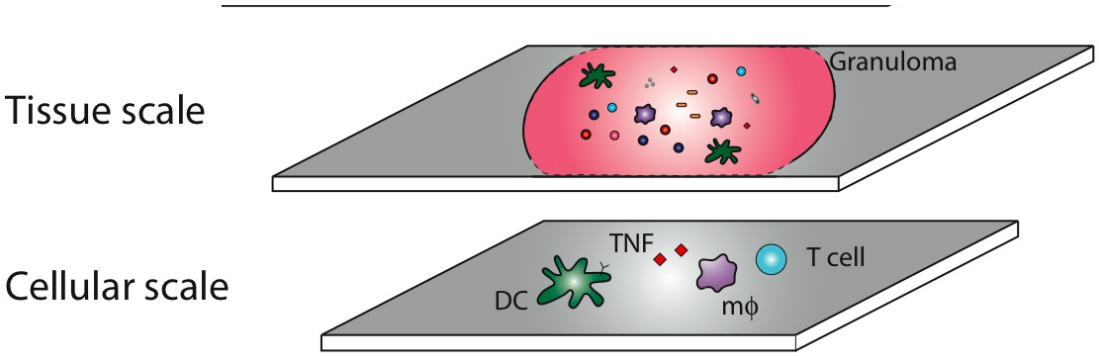
Granulomas are a result of multi-scale dynamics in both space & time



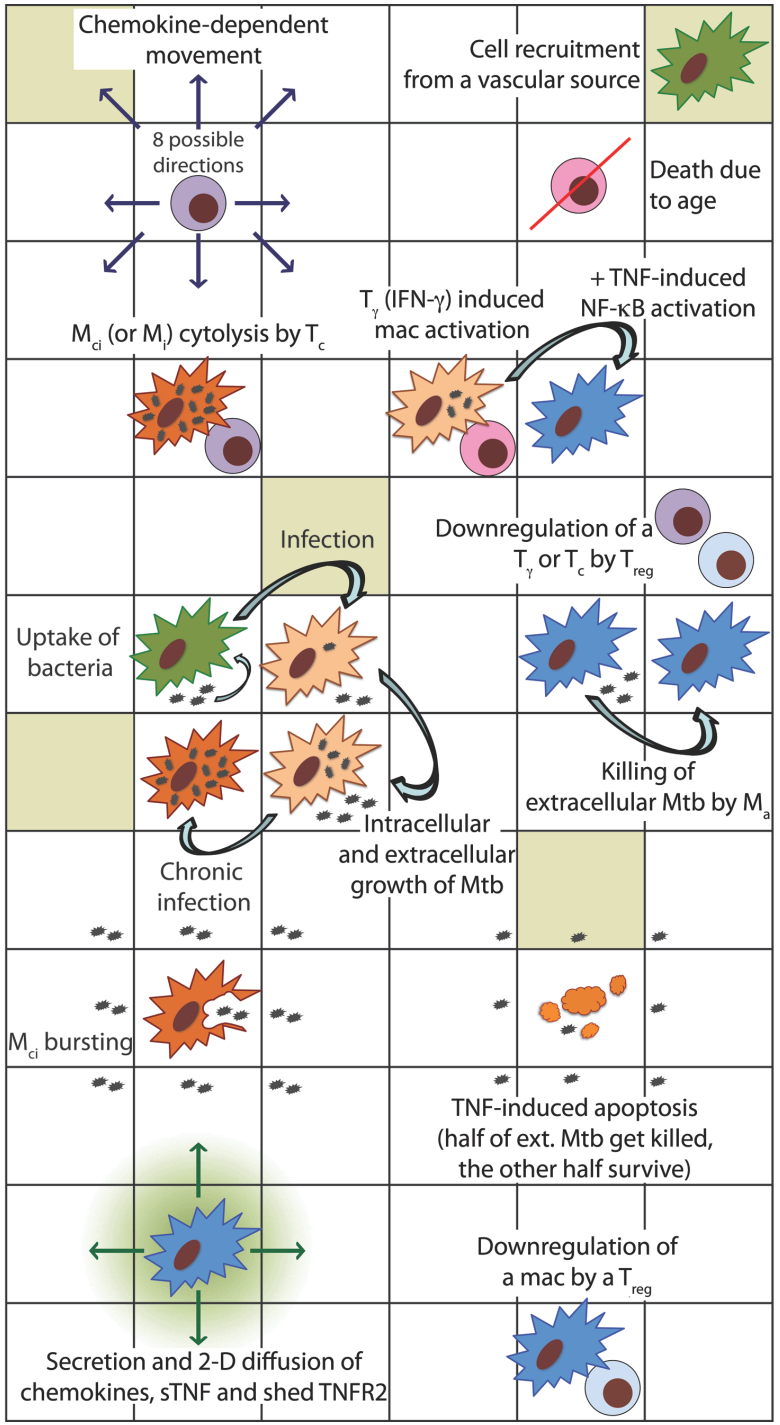
GranSim (2D/3D)

Cellular/tissue scale Model--

A stochastic **model** that captures discrete cellular dynamics via a set of well-described interactions between immune cells and Mtb leading to tissue scale outcomes



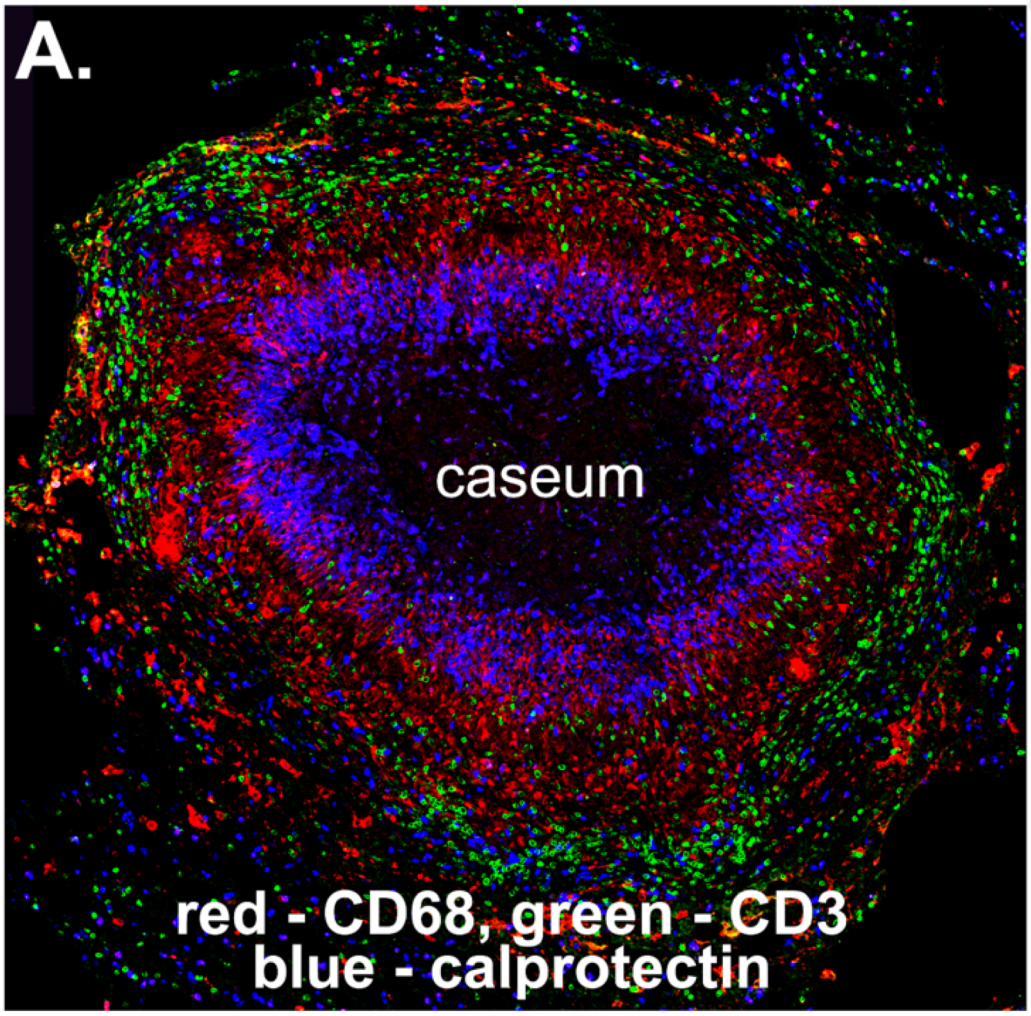
****Leads to "emergent behavior"**

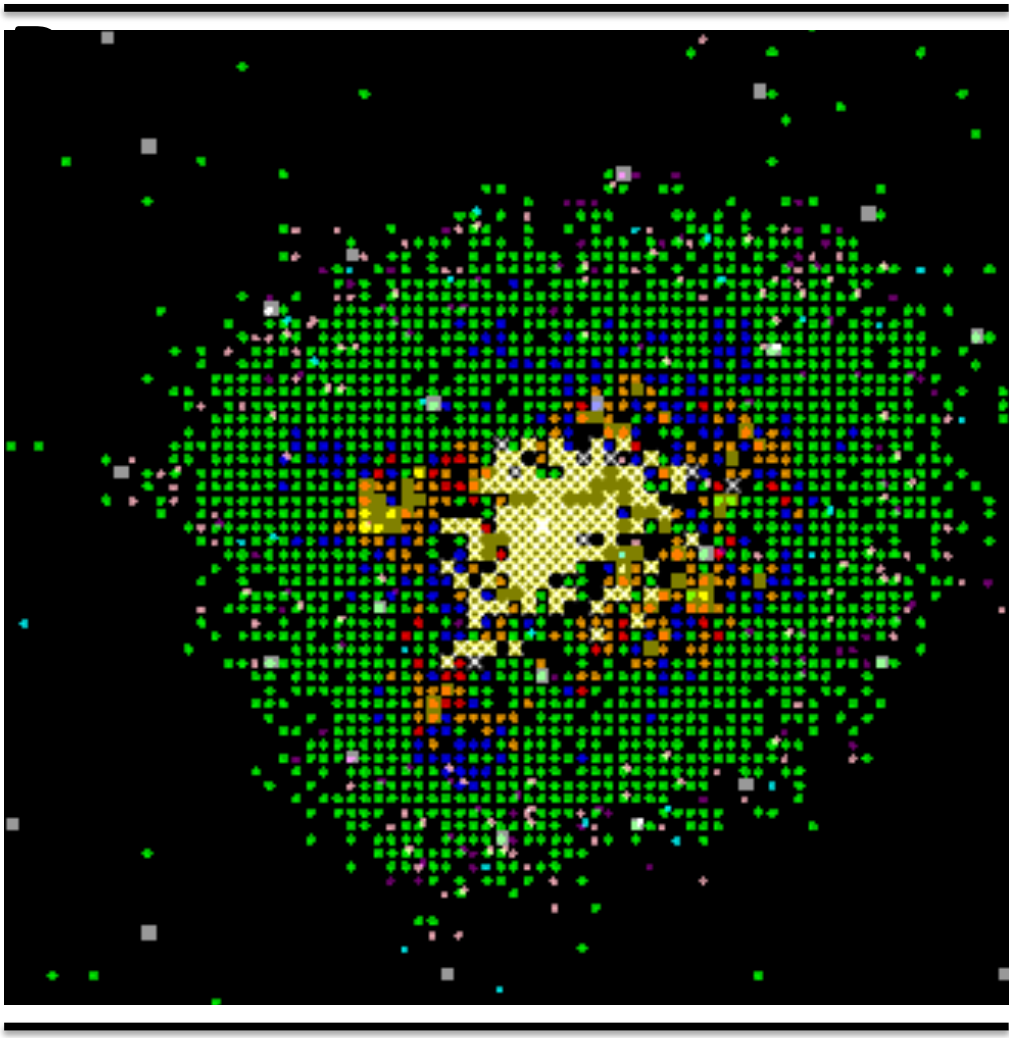


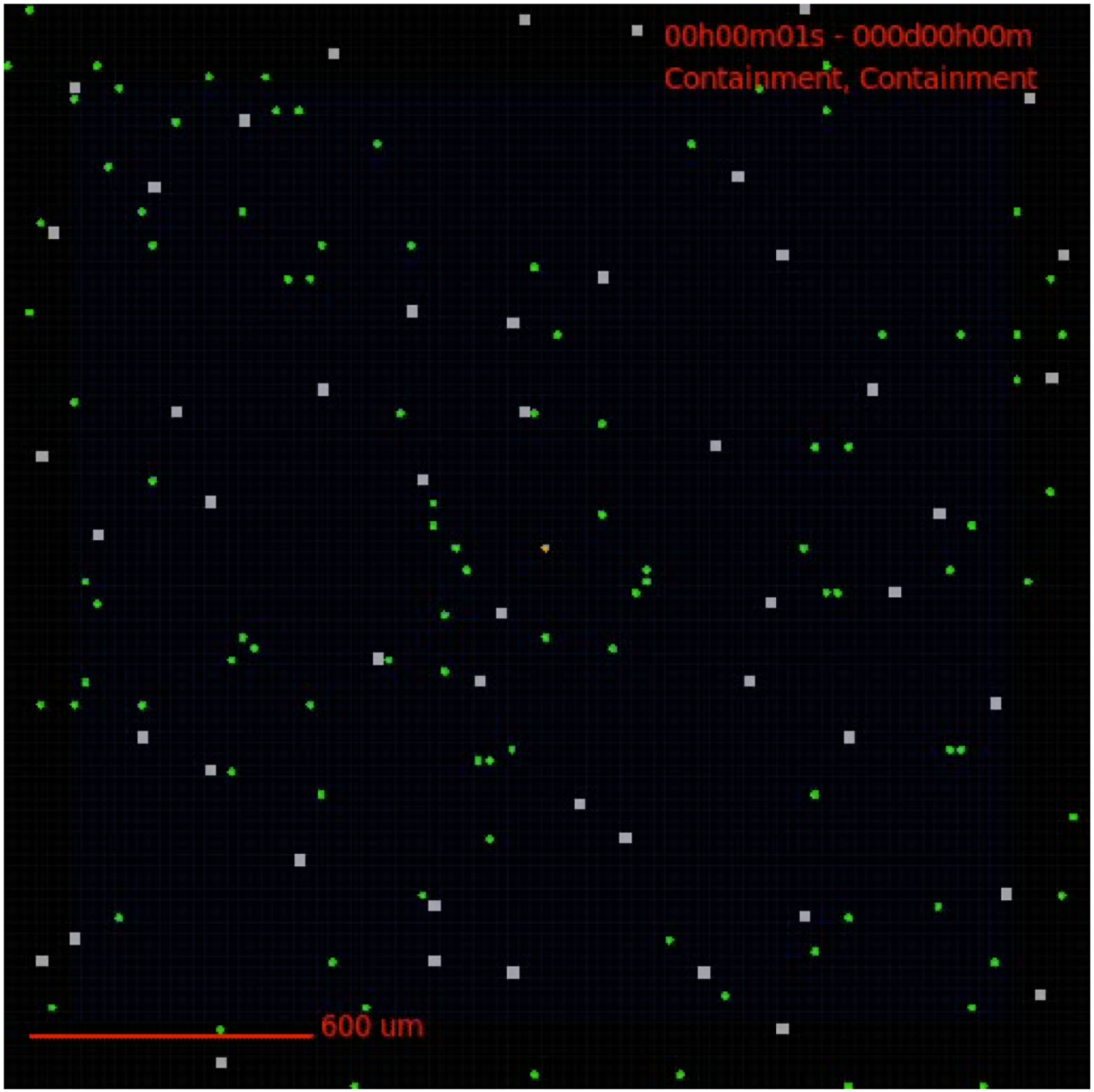
*Segovia-Juarez et al J. Theor Biol. 2004

* Ray *et al*, J. Immunol. 2009

Experimental & computer generated granuloma







Granuloma formation

Macrophages	Extrac. Mtb
Resting	Caseated
Active	T cells
Infected	Regulatory
Chronically Infected	Gamma
	Cytotoxic

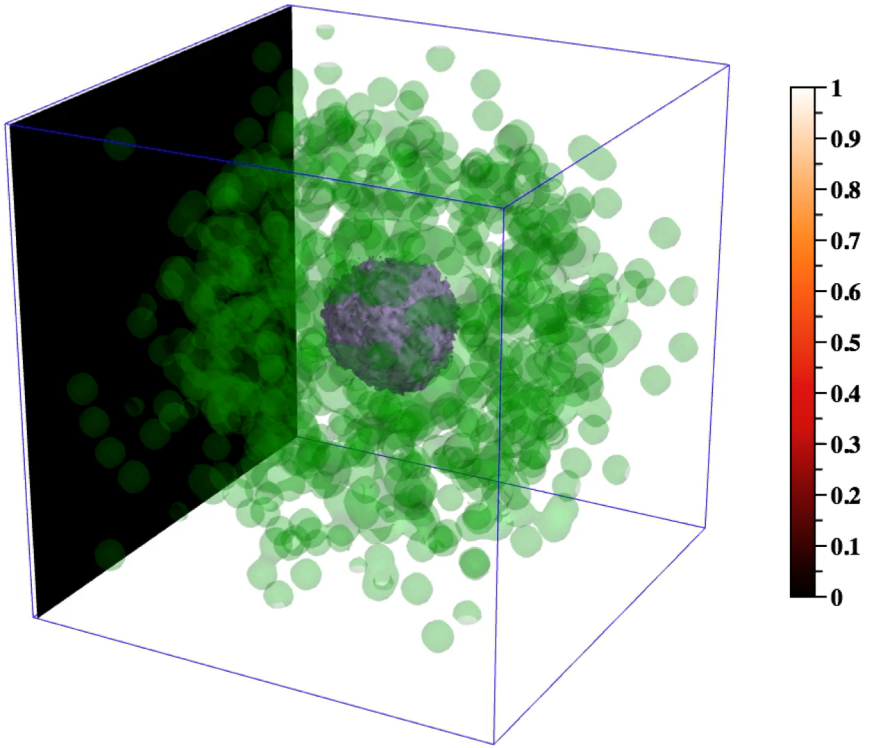
*Calibrated to NHP data
*Simulation begins with a single infected macrophage with one intracellular bacteria

*Video simulates 200 days of infection

*2mmx2mm lung tissue

Visualizing simulated molecule chemical gradients at day 50 post-infection for one simulation (using DataTank).

Viewing chemical gradients in 3D space 50 days post infection



BLACK:a 2D, yz-slice at x=50,

GREEN: we use 3D level sets to depict the 3D spatial distributions of two biologically-relevant TNF concentrations for macrophage recruitment.

***TNF concentration is 0.05 or greater (i.e. level set = 0.05); this is the minimum concentration that macrophages can sense, a concentration below this value is not detectable by macrophages.**

PURPLE:
TNF concentration is 50 or greater (i.e. level set = 50); this is the maximum concentration that macrophages can sense

Why is TB so hard to treat?

1.Mycobacterium

- Slow growing bacterium
- Acquires antibiotic resistance

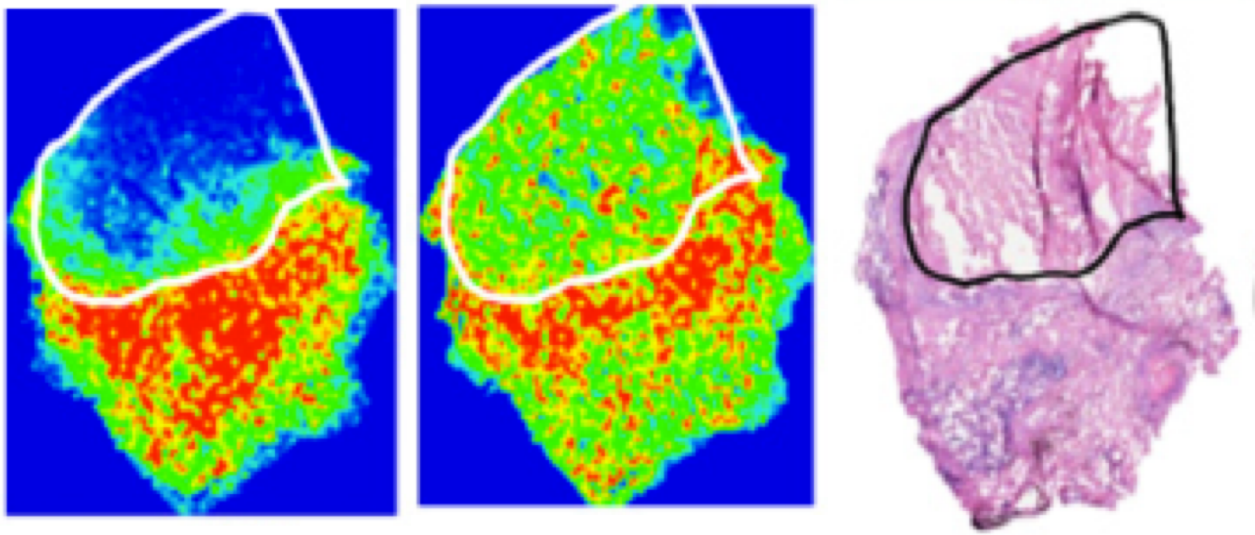
2. Granulomas

- Granulomas present physiological barrier to antibiotic diffusion

3. Patient-unfriendly treatment

- Long -9 months
- multiple drugs given
- many side-effects

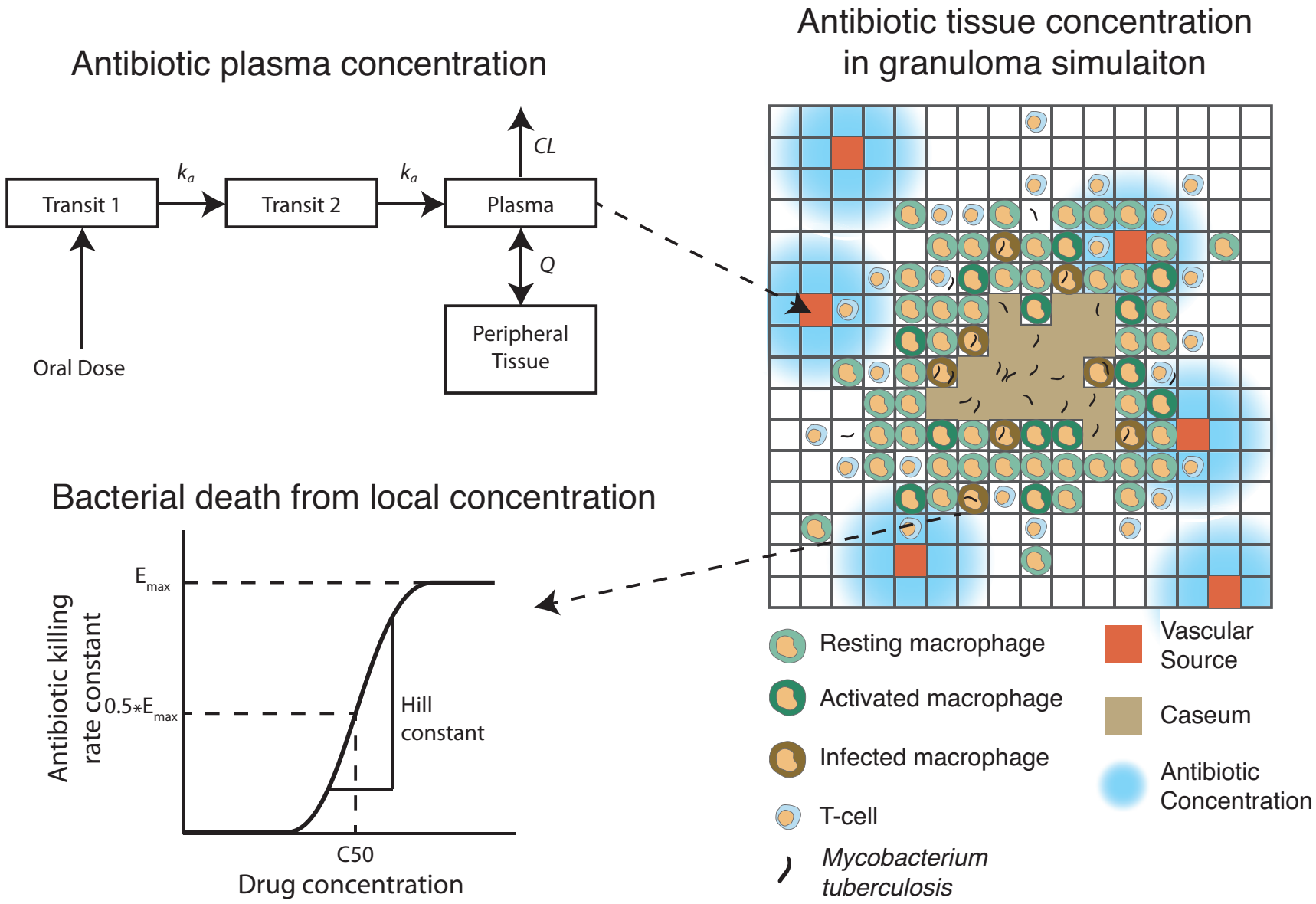
- 4-drug standard regimen: **INH, RIF, PZA, EMB**



Prideaux *et al*. *Nat. Med.* (2015)

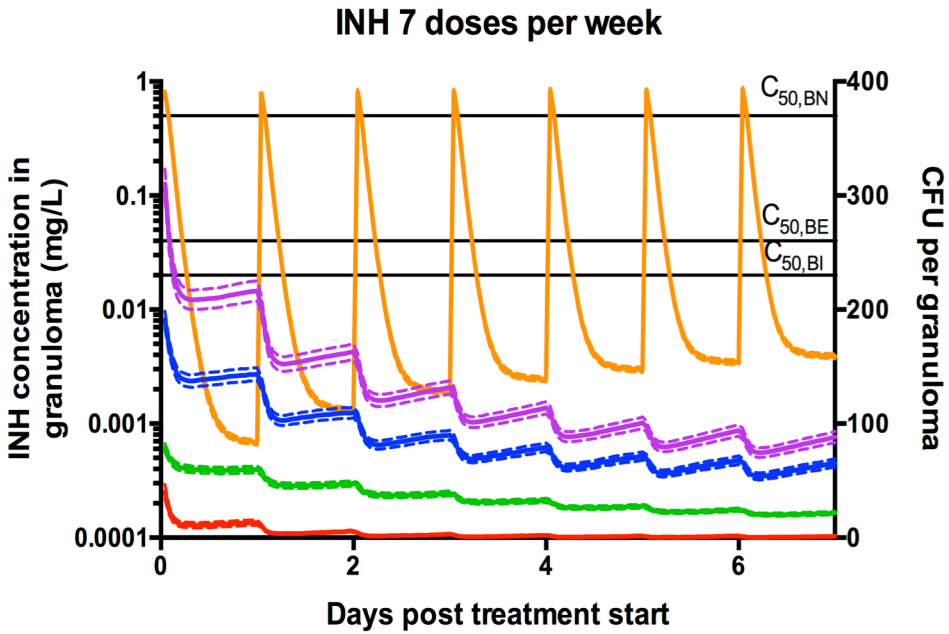
Pharmacokinetics/Pharmacodynamics Modeling

(PK/PD)

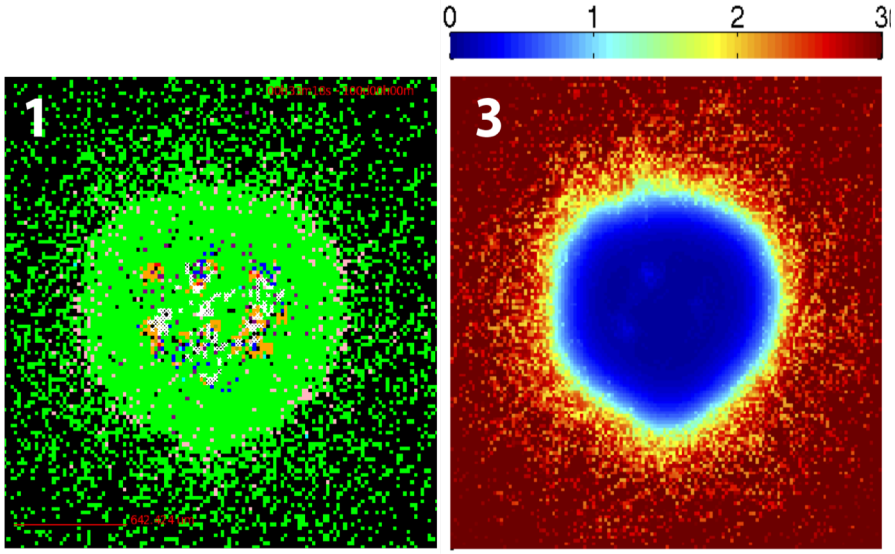


Elsje Pienaar et al. *A computational tool integrating host immunity with antibiotic dynamics to study tuberculosis treatment J. Theo. Biol, 2015*

2. Predicting granuloma antibiotic exposure



Orange trace – INH concentration over time
Purple trace – Total bacteria over time



Antibiotic exposure inside granuloma much lower than outside

Concentration oscillates between above and below effective concentrations

Pienaar *et al*. *BMC Sys. Bio.* (2015)

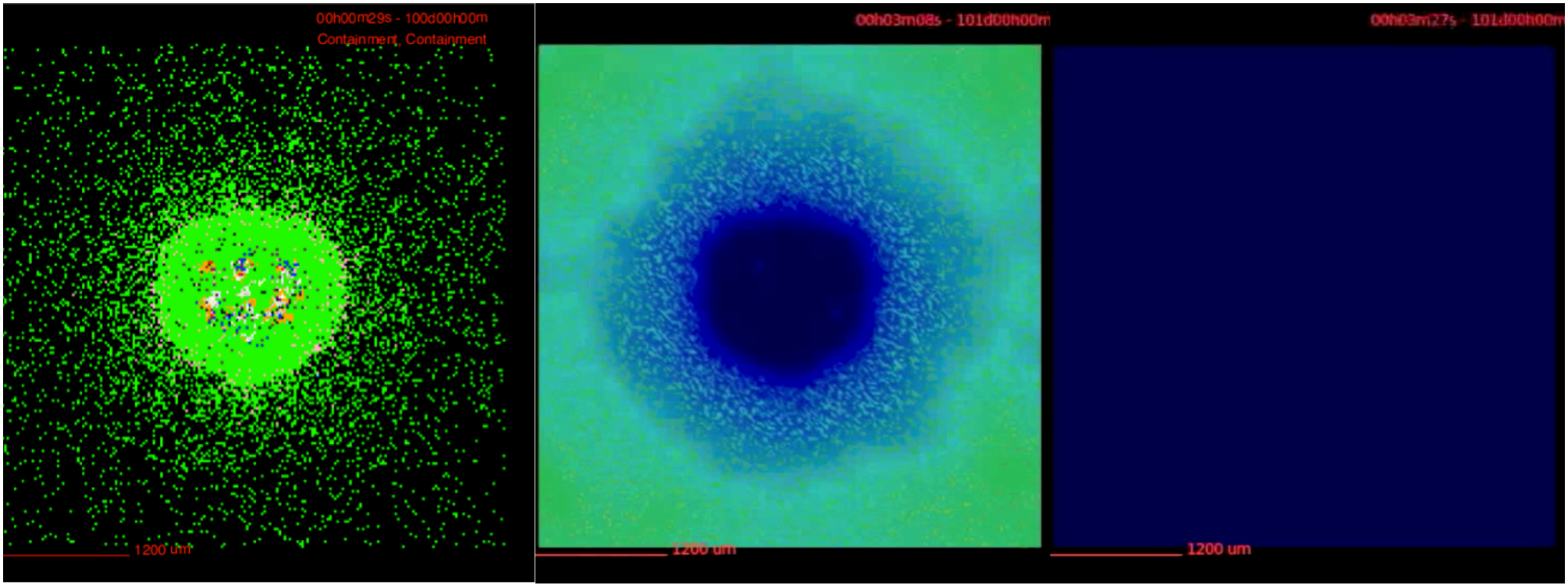
Antibiotic spatial dynamics during daily dosing

RIF

Color scale: 0 – 10 mg/L

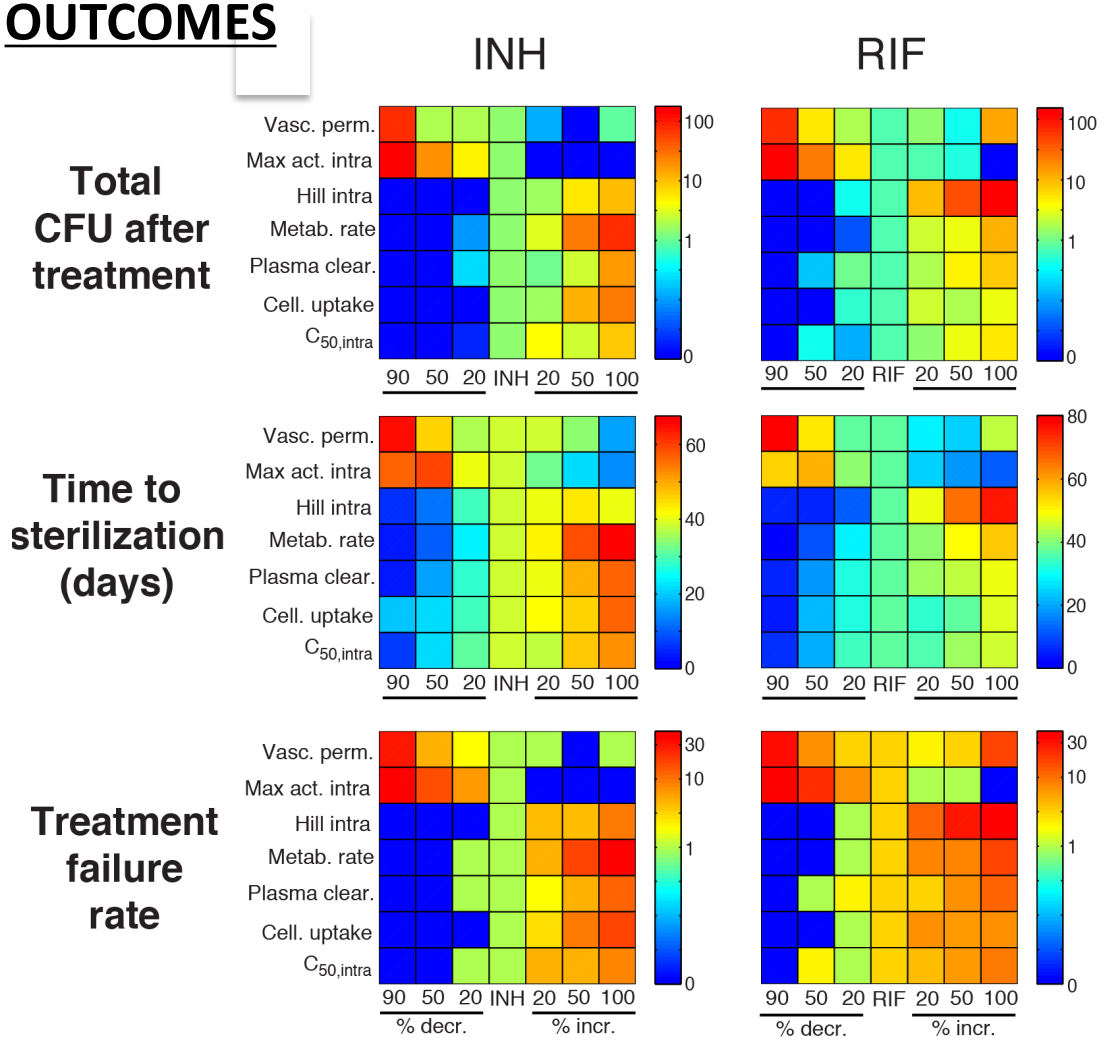
INH

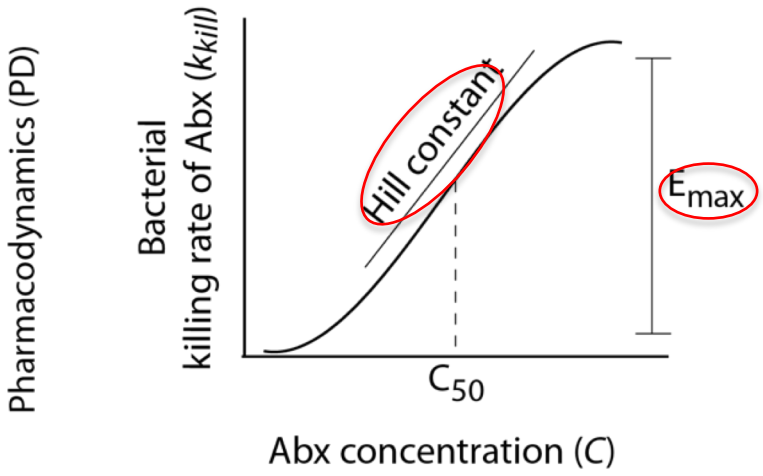
Color scale: (0 – 0.5 mg/L)



Can we predict what can improve drugs?
i.e. which PK and PD properties are good targets for modification?

OUTCOMES





Use sensitivity and uncertainty analysis to determine this

3. What's the best antibiotic regimen?

- Current regimen is 4 drugs 6-9 months
- Too many options to test
  - Clinically or computationally
- Optimization problem

Regimen design space (RDS)

Treatment segments (M)	2
Number of drugs (c)	10
Drugs per segment (n)	4
Dose (D, mg/kg)	5
Frequency (F, week^{-1})	7

Number of possible regimens:

$$RDS = \left(\binom{c}{n} (D \times F)^n \right)^M = 9.9 \times 10^{16}$$

Cicchese *et al*. CMBE (2017)

Defining the optimization problem

Regimen of *n* antibiotics (2*n*-dimensional)

$$\boldsymbol{x} = \{D_1, F_1, D_2, F_2 \ldots D_n, F_n\}$$

Antibiotic 1 Dose

Antibiotic 1 Dose Frequency

INH and RIF regimen (4-dimensional)

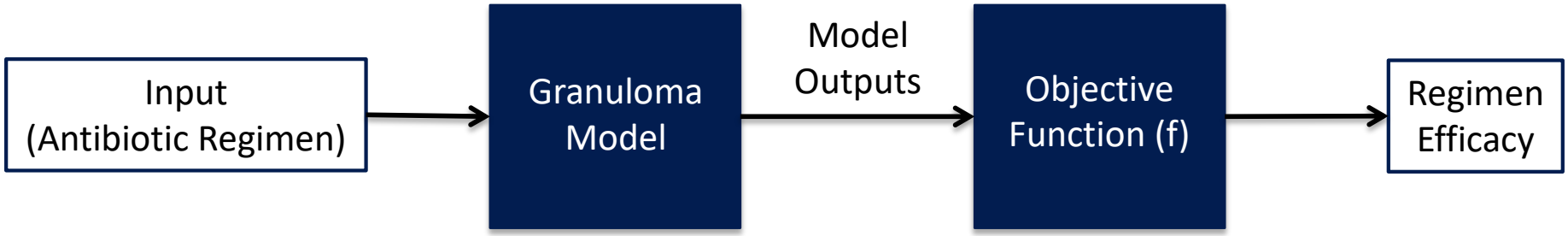
$$\boldsymbol{x} = \{10\ \text{mg/kg}, 7\ \text{wk}^{-1}, 15\ \text{mg/kg}, 2\ \text{wk}^{-1}\}$$

INH
10 mg/kg given
7 times per week

RIF
15 mg/kg given
2 times per week

Objective function is a function of model outputs

- Time to sterilize granuloma of bacteria
- Antibiotic dose
- Measures of resistance
- Potentially more



Cicchese *et al*. CMBE (2017)

Optimization algorithm options

Genetic Algorithm

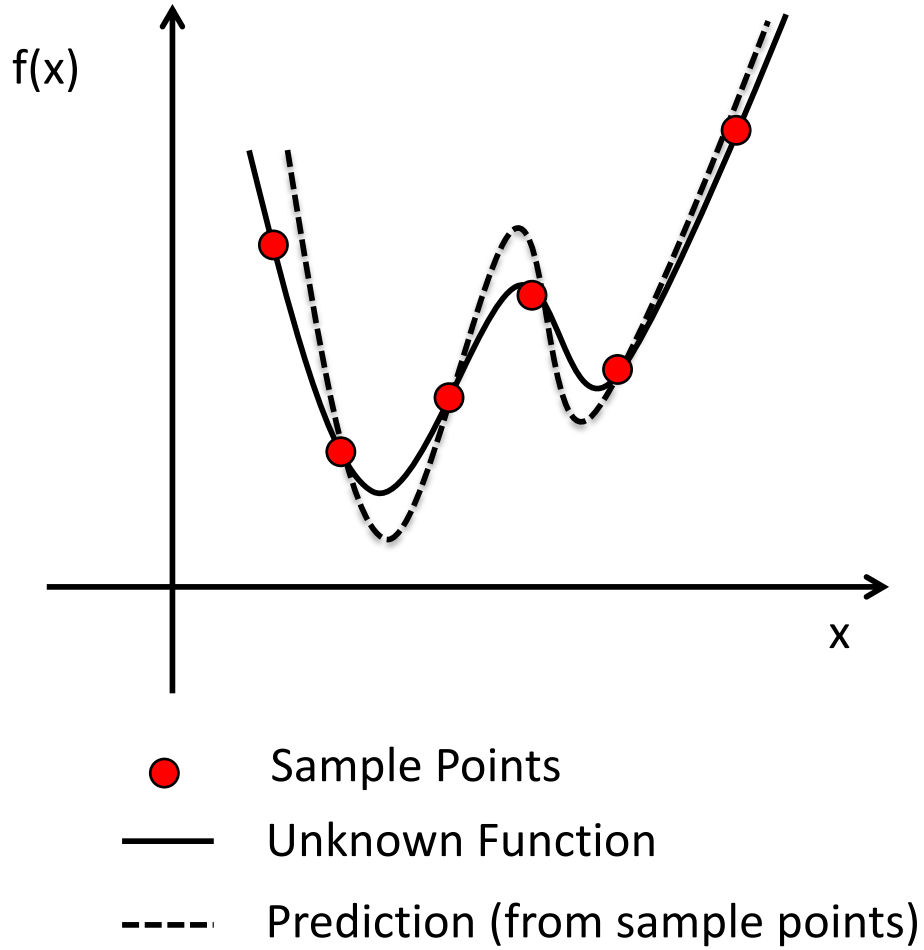
1. Initialize population

2. Population evolution
   i. Evaluation fitness
   ii. Select parents
   iii. Generate new population
   iv. Check stopping criteria

3. Solution found

Man *et al*. *IEEE Trans.* (1996)
Jin and Branke, *IEEE Trans.* (2005)

Surrogate-Assisted Optimization



Forrester *et al*. (2008)
Akhtar *et al. J Global Opt* (2016)

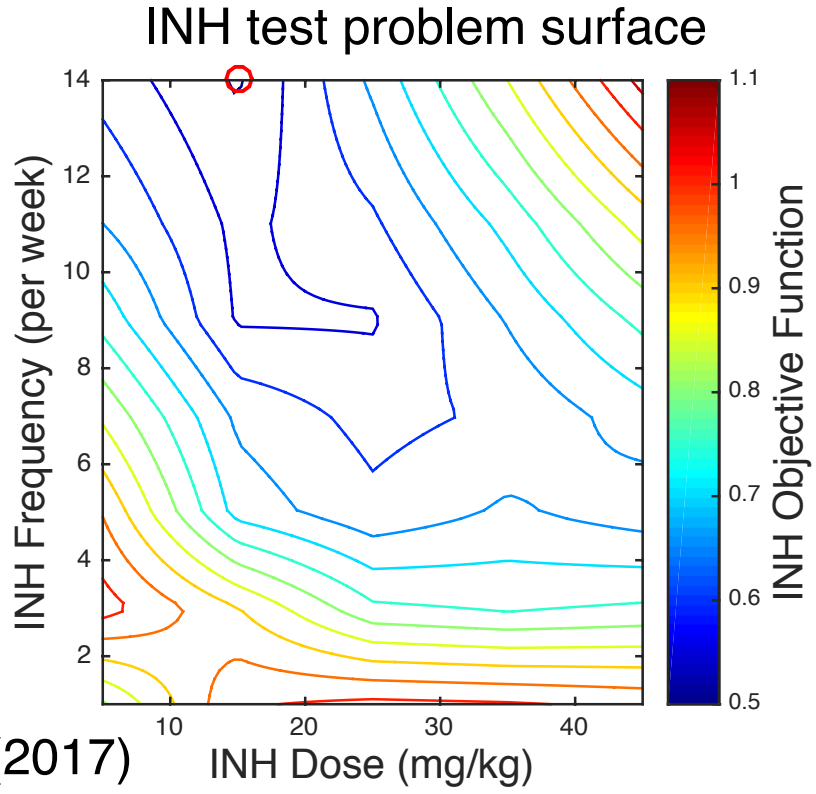
Test problems for comparing algorithms

- Single-antibiotic test problems, optimize dose and frequency
- Based on published simulations (Pienaar *et al. BMC Syst. Biol.* (2015))
- Generate objective function surface based on simulation output
- Use to test performance of optimization algorithms
  - Accuracy: distance from the known solution
  - Efficiency: how much of the design space sampled

Objective function

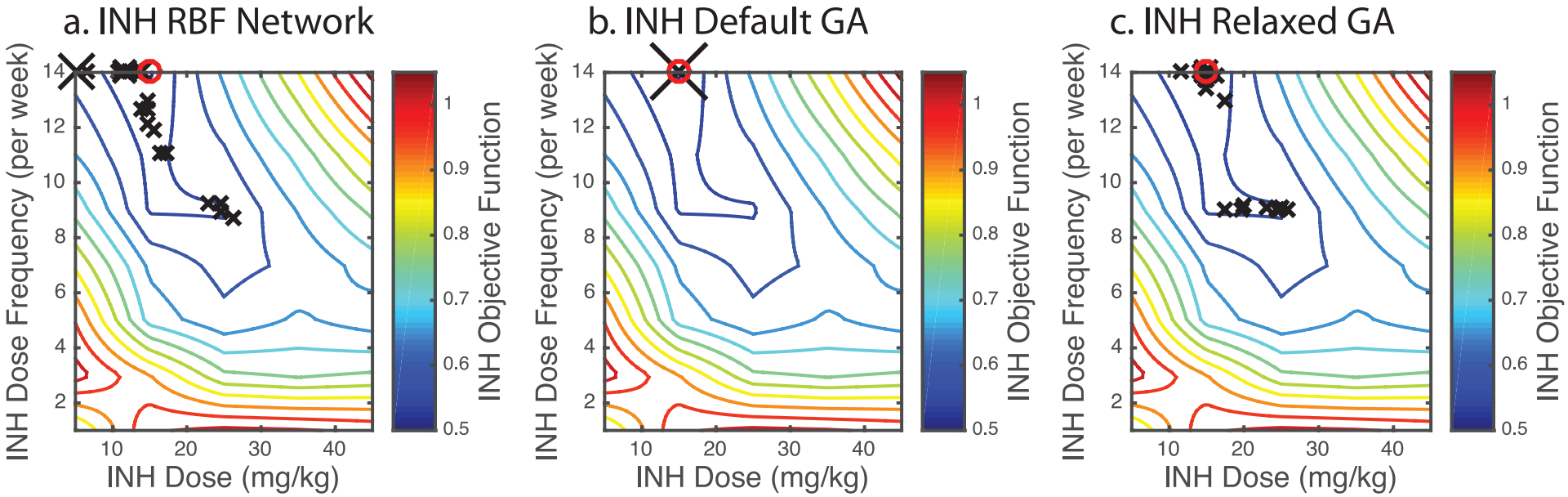
$$f(\mathbf{x}) = \frac{t_c(\mathbf{x})}{t_{c,\max}} + \frac{D(\mathbf{x})}{D_{\max}}$$

$$f = (\text{time to clear}) + (\text{dose})(\text{freq.})$$



Cicchese *et al.* Cellular and Molecular Bioengineering (2017)

GA most accurate in predicting solutions



Surrogate-Assisted Optimization

Genetic Algorithms

- Each algorithm solved the test problem with 30 independent optimizations
- Each “x” corresponds to the estimated optimal solution

Cicchese *et al*. CMBE (2017)

Surrogate-assisted optimization requires fewer function evaluations

Optimization method	**Average number of function evaluations**
Surrogate-assisted optimization	21
Default genetic algorithm	3,855
Relaxed genetic algorithm	347

With modifications, surrogate assisted models can efficiently predict optima with sufficient accuracy

Cicchese *et al*. CMBE (2017)

Predicted optimal regimens for 2 drugs

Dose-weight Parameter (ω)	INH Dose (mg/kg)	INH Dose Freq. (week^{-1})	RIF Dose (mg/kg)	RIF Dose Freq. (week^{-1})	Weekly INH Dose (mg/kg)	Weekly RIF Dose (mg/kg)	Total Weekly Dose (mg/kg)	Predicted Time to Sterilize (days)	Simulated Time to Sterilize (days)
0.75	28.9	7.3	8.6	1.9	210.7	16.1	226.8	10.2	16.3
1	22.5	1.1	25.7	4.9	25.9	125.8	151.8	17.3	21.9
	25.7	7.3	8.6	1.9	187.4	16.1	203.4	11.8	16.8
2	22.5	1.1	17.1	3.7	24.1	63.3	87.4	25.9	31.0
	22.5	7.3	4.3	1.9	171.2	8.0	179.2	15.5	21.9
3	19.3	1.1	12.9	3.7	20.7	47.8	68.4	30.2	42.4
	19.3	4.9	4.3	1.9	94.5	8.0	102.5	29.9	31.4

*Note: Doses for non-human primate that yield similar exposure levels in humans

Identifies two major regions:

1. 20-25 mg/kg INH daily, low dose RIF twice a week
2. 20-25 mg/kg INH weekly, ~15 mg/kg RIF daily

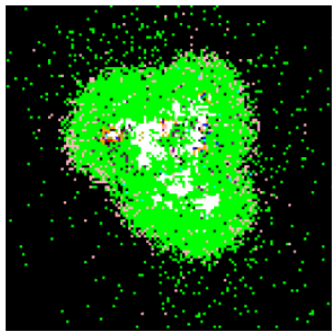
***Major regions similar to human equivalent doses for CDC regimens**

Surrogate model predictions are fast, accurate and relevant

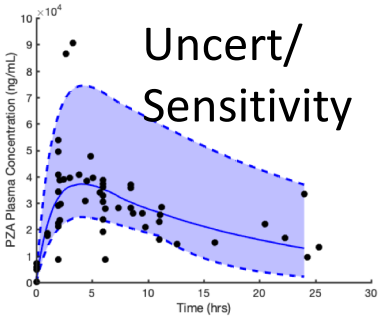
Cicchese *et al*. CMBE (2017)

OPTIMIZATION PIPELINE

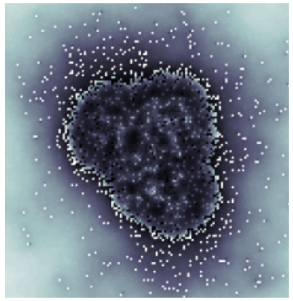
1. GranSim

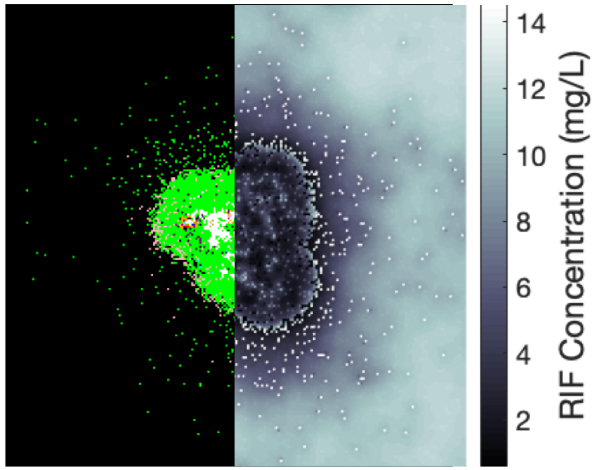


2. /PD of Antibiotics



Uncert/ Sensitivity





Multi-scale antibiotic and granuloma model

3. Surrogate-assisted optimization

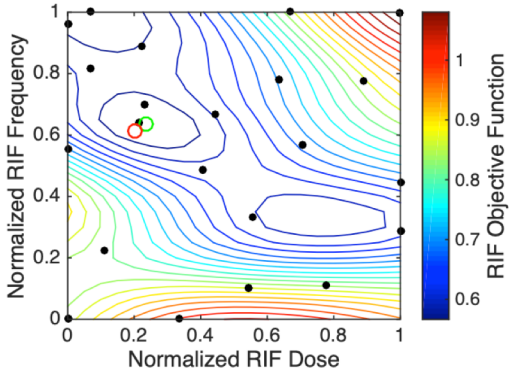
Test different regimens

Objective function

Build surrogate model

Resample regimen space

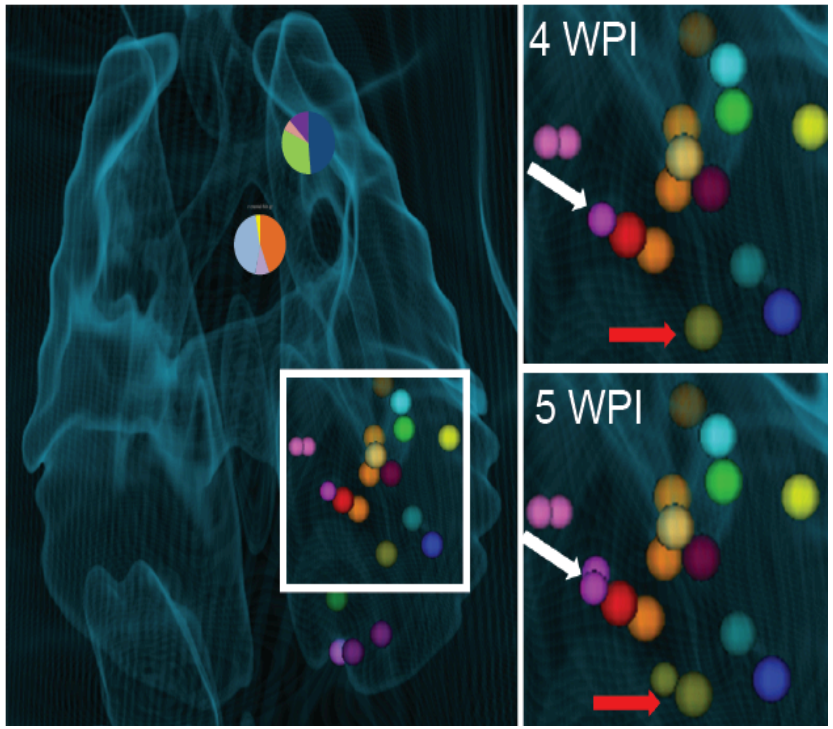
4. Predicted optimal regimen(s)

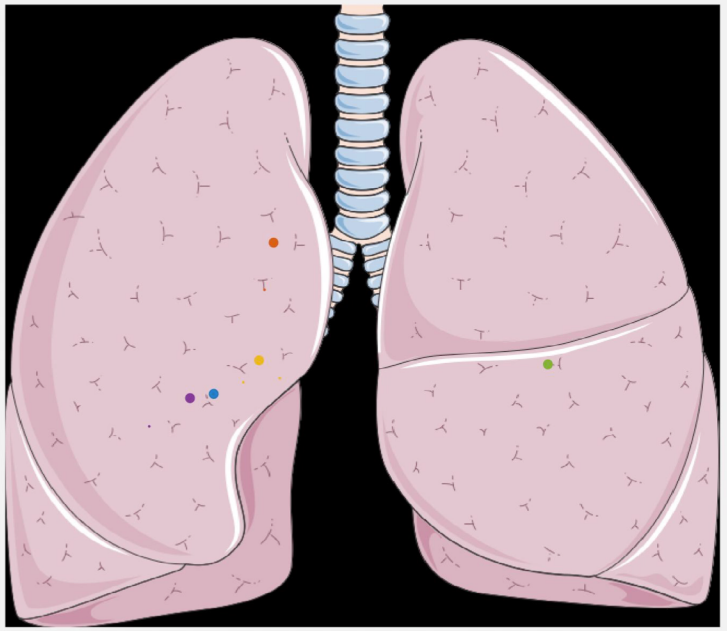


5. Test in virtual treatment trial

6. Animal model testing

Capturing Multiple granulomas-





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Martin CJ, Cadena AM, Digitally Barcoding *Mycobacterium tuberculosis* Reveals *In Vivo* Infection Dynamics in the Macaque Model of TuberculosisLeung VW, Lin PL, Maiello P, Hicks N, Chase MR, Flynn JL, Fortune SM. MBio. 2017 May 9;8(3). pii: e00312-17. doi: 10.1128/mBio.00312-17.

A multi-scale systems pharmacology approach to tuberculosis therapy

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Grant #: U01 HL131072

Introduction

Tuberculosis (TB) is a bacterial infection, primarily affecting lung tissue, that is considered one of the world's deadliest infections. Caused by infection with the pathogen *Mycobacterium tuberculosis* (Mtb), standard treatment of TB often requires six months or longer of multiple antibiotics. Length of treatment and the emergence of drug-resistant TB indicate a need for better antibiotic regimens.

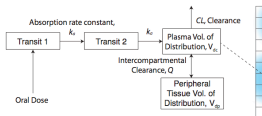
Granuloma formation in lung tissue during infection
- lesion that forms due to immune response
- hallmark of TB, location of Mtb
- physiological barrier to antibiotic diffusion

Design of better regimens requires modeling at multiple scales:
1. **Molecular scale** - pharmacokinetic (PK) and pharmacodynamic (PD) models to predict antibiotic distribution in granulomas, secretion and diffusion of cytokines (TNF, IL10, etc.)
2. **Cellular and tissue scale** - agent-based models to simulate cellular interactions and rules, with emergent granuloma formation representing tissue-level structure
3. **Whole body** - combination of granuloma simulations through linked antibiotic plasma PK to simulate treatment of infected host

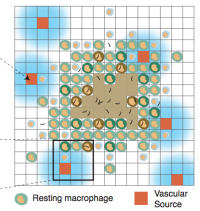
Using computational modeling and surrogate-assisted optimization algorithms, we can screen the **regimen design space** for optimal therapies and test efficacy in a **virtual clinical trial**

Modeling granuloma formation and PK/PD

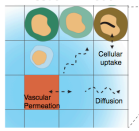
Plasma PK compartmental model



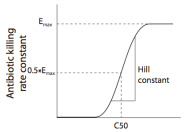
Granuloma agent-based model

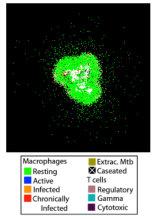


Tissue PK Model



Antibiotic Pharmacodynamics





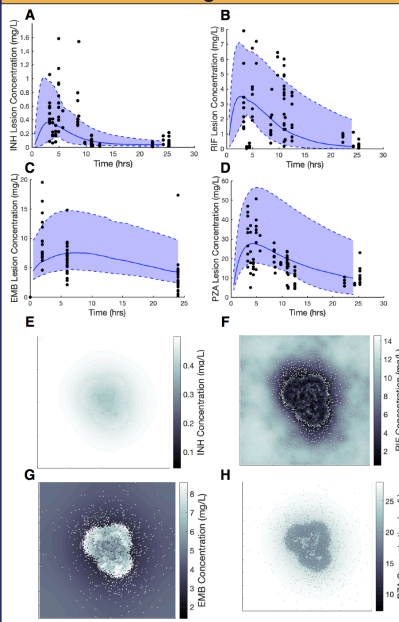
PK/PD model components [2]
Plasma PK - ODE's to model plasma concentration

Tissue PK - PDE's to model antibiotic diffusion, binding and degradation

PD model - killing rate constant based on Hill curve

Granuloma model [1] - agent-based model to capture immune cell interaction and responses, bacterial growth and states, and structural environment for antibiotic diffusion

Predicting *in vivo* antibiotic distributions



Parameters of PK models are fit to capture the range of antibiotic concentrations observed *in vivo*.

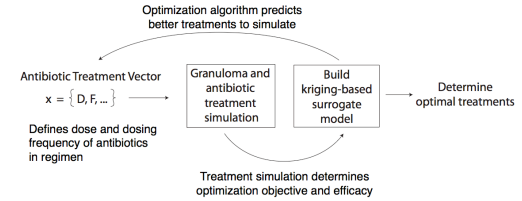
(A-D) Black dots represent experimentally measured antibiotic concentrations in granulomas after oral dosing in human lesions (INH, RIF and PZA) or rabbits (EMB) [3,4]. Blue shaded region is range of simulation, with the solid blue line representing the simulation's median concentration based on varying plasma PK parameters.

(E-H) Spatial distribution of the antibiotics in the agent-based simulation around the time of peak average granuloma concentration (2 to 4 hours depending on the antibiotic). Lighter shades indicate higher concentrations. Many antibiotics have lower concentrations in granulomas, or fail to diffuse completely into the granuloma.

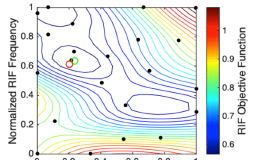
First-line TB Antibiotics:
INH = isoniazid
RIF = rifampin
EMB = ethambutol
PZA = pyrazinamide

Optimizing antibiotic regimens

Surrogate-assisted optimization of antibiotic regimens [5]



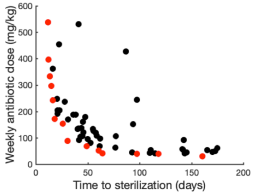
Single-objective optimization



$$f(\mathbf{x}) = \frac{t_c(\mathbf{x})}{t_{c,\max}} + \frac{D(\mathbf{x})}{D_{\max}} \qquad f = (\text{time to clear}) + (\text{dose})(\text{freq.})$$

A single-objective function that simultaneously attempts to minimize sterilization time and weekly antibiotic dose shows how changing dose size and dosing frequency affects regimen optimality. Black dots represent regimens used to build surrogate, the green circle is the surrogate predicted optimum compared to the true global optimum (red circle)

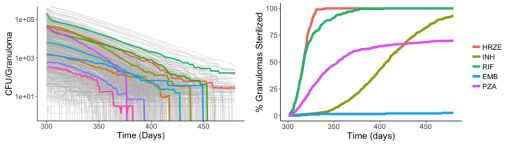
Multi-objective optimization



Multi-objective optimization provides the trade-off between two opposing objectives (such as antibiotic dose and sterilization time). Red points represent regimens that are pareto-optimal, and black dots are non-optimal simulated regimens that were sampled.

Predicting regimen efficacy and virtual clinical trials

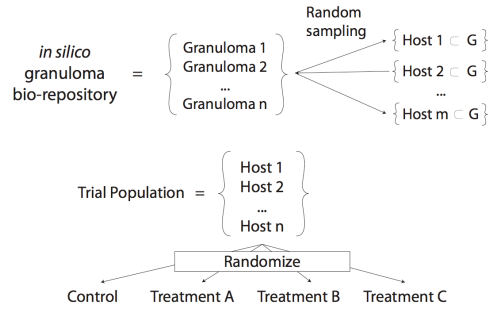
Simulating antibiotic therapies gives sterilization dynamics



Decrease in number of bacteria for different granulomas (gray lines) during treatment with daily doses of INH. Colored lines are selected simulations to aid in visualization.

Comparison of percentage of simulated granulomas sterilized with different antibiotics over duration of treatment gives clinically relevant antibiotic efficacy ranking.

Combining granulomas into hosts for virtual clinical trials



Conclusions

- Multi-scale modeling of granuloma formation and antibiotic treatment combines agent-based models of cellular interactions, pharmacokinetic models involving ODE's and PDE's, and optimization algorithms to search for better regimens.

- The model quantitatively captures range of antibiotic concentration experimentally observed, as well as qualitative spatial distributions.

- Surrogate-assisted optimization provides an efficient way to search for optimal antibiotic regimens.

- Treatment simulations can be used to compare antibiotics in sterilizing granulomas. Grouping granulomas together into hosts provides a computational framework to compare regimens through virtual clinical trials

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