

Determining the drivers of antibiotic resistance epidemiology

Gwen Knight

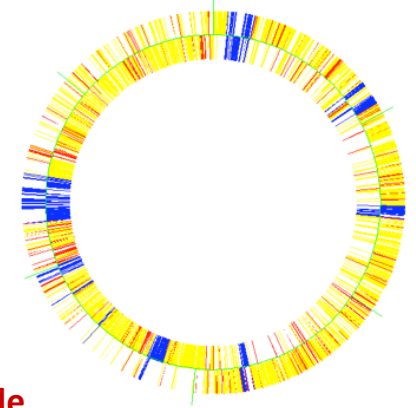
LONDON
SCHOOL *of*
HYGIENE
& TROPICAL
MEDICINE



What drives antibiotic resistance
changes in hospital-associated MRSA?

MRSA overview

- *S. aureus* is a commensal (~25% population^{1,2})
- UK: MRSA causes 16% of all hospital-acquired infections³
- Many “lineages”, but small number dominate in each country
e.g. UK MRSA: **CC22** and **CC30**



Core

Core variable

Mobile Genetic Elements (MGEs)⁴

1. van Belkum (2009)

2. Klutymans (1997)

3. Smyth (2008)

4. Lindsay (2006)

Aims

- Quantify *naturally occurring* fitness differences between *clinical* MRSA isolates associated with
 1. Antibiotic resistance
 2. Lineage background
- Use these to parameterise models of antibiotic resistance evolution in MRSA

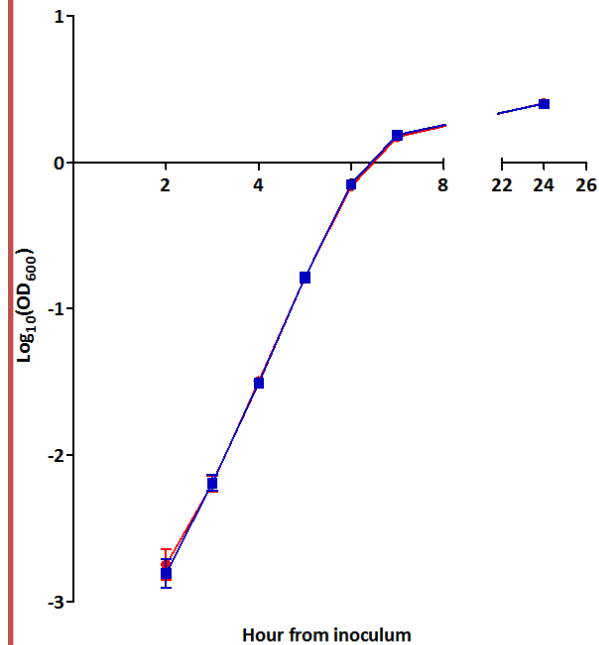
Isolates

Antibiotic	202	215	201	211	228	221	226	205	224	206
Ampicillin	Red	Red	Red	Red	Red	Orange	Red	Red	Red	Red
Penicillin	Red	Red	Red	Red	Red	Red	Red	Red	Red	Red
Oxacillin	Red	Red	Red	Red	Red	Orange	Red	Red	Red	Red
Teicoplanin	White	White	White	White	White	White	White	White	White	White
Vancomycin	White	White	White	White	White	White	White	White	White	White
Ciprofloxacin	Red	Red	Red	Red	Red	Red	Red	Red	Red	Red
Rifampicin	White	White	White	White	White	Red	White	Red	White	Red
Chloramphenicol	White	White	White	White	White	White	White	White	White	White
Clindamycin	Red	Red	Red	Red	Yellow	Yellow	Red	Red	Red	Red
Erythromycin	Red	Red	Red	Red	Red	Red	Red	Red	Red	Red
Gentamicin	Red	White	White	White	Red	Red	Red	White	White	Red
Kanamycin	Red	Red	Red	Red	Red	Red	Red	White	White	Red
Tobramycin	Red	Red	Red	Red	Red	Red	Red	White	White	Red
Tetracycline	White	White	White	White	White	White	White	White	White	Red
Fusidic Acid	White	White	White	White	Red	Red	White	White	White	Red
Trimethoprim	Red	Red	White	White	Red	Red	Red	White	White	Red
Mupirocin	White	White	White	Red	White	White	Red	White	White	White
# of isolates	1	2	1	1	1	2	1	1	1	1
Plasmid presence	×				×	×	×			

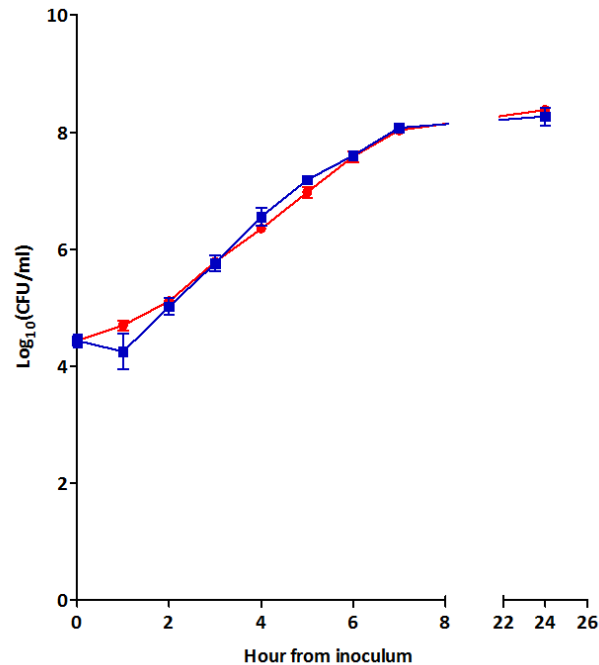
1. Effect of antibiotic resistance on fitness

> Pairwise comparison: **With plasmid (202)** vs **Without plasmid (215)**

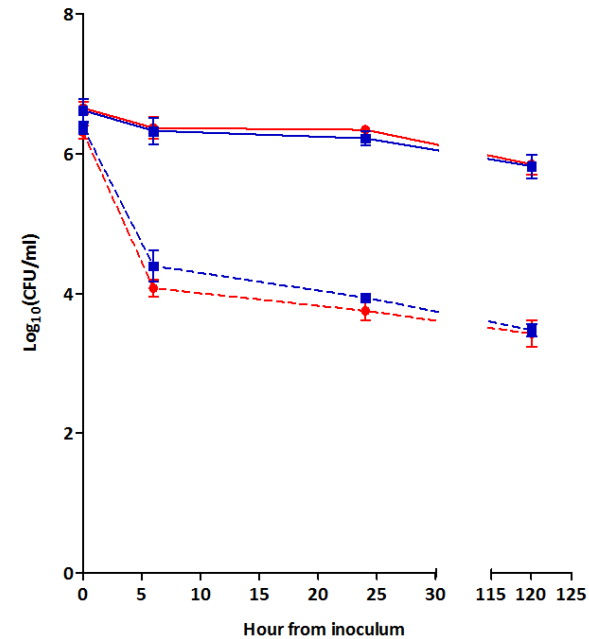
Independent growth



Co-culture/competition



Survival



(Knight, 2013)

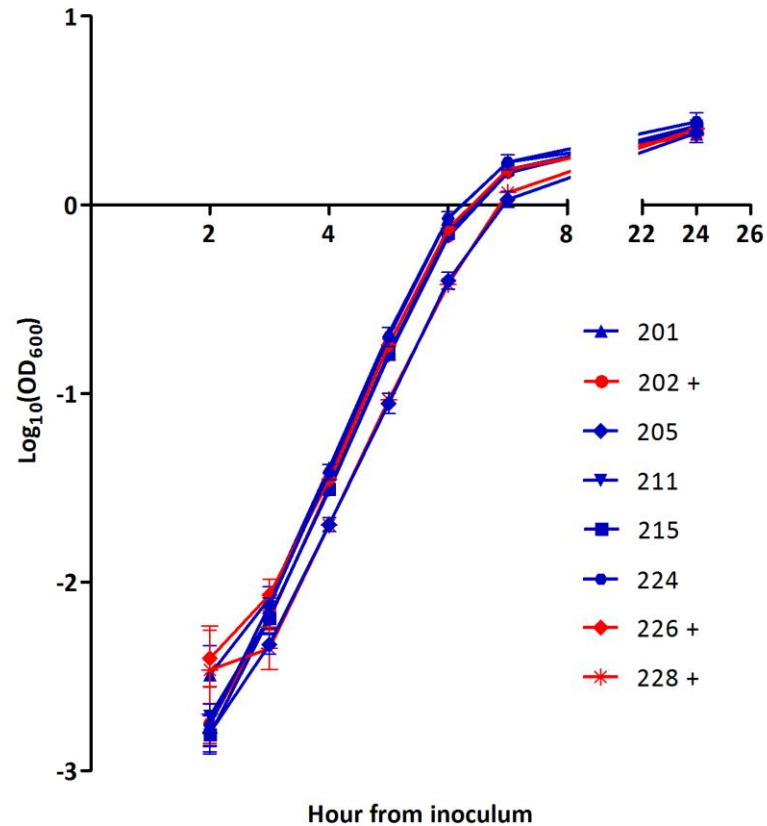
> Under a range of mathematical measures: no significant difference ($\alpha=0.05$)

1. Effect of antibiotic resistance on fitness

> Across antibiogram comparison

With plasmid vs Without plasmid

Independent growth



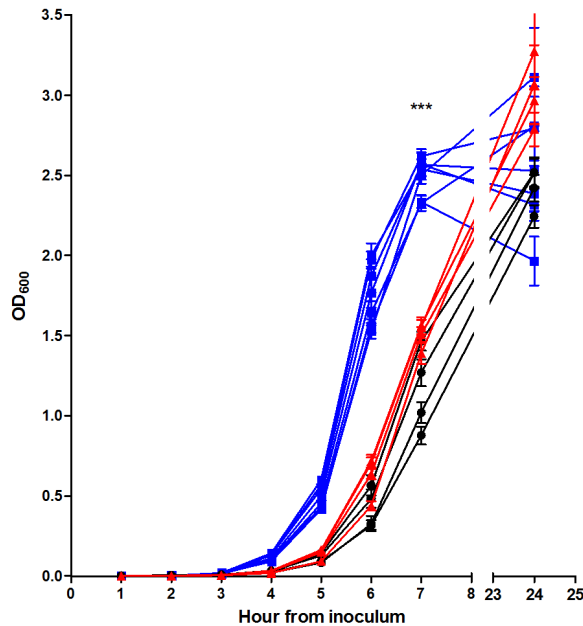
(Knight, 2013)

> Under a range of mathematical measures: no significant difference ($\alpha=0.05$)

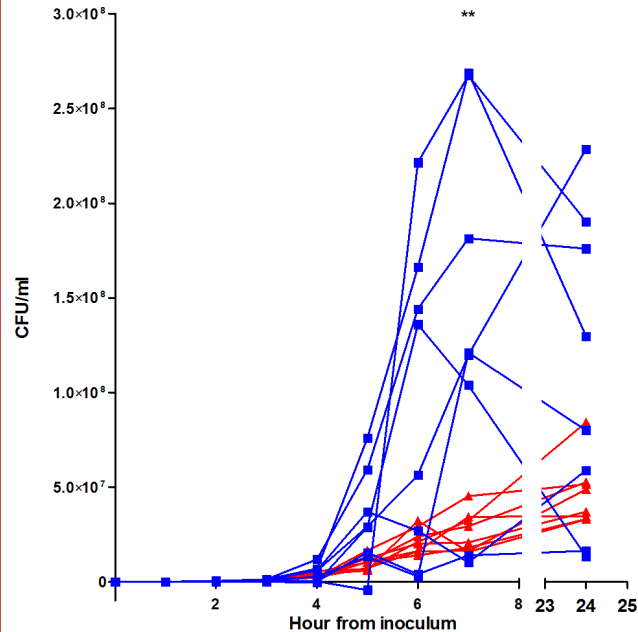
2. Effect of lineage background on fitness

> Across lineage comparison **CC22** **CC30** **CC239**

Independent growth

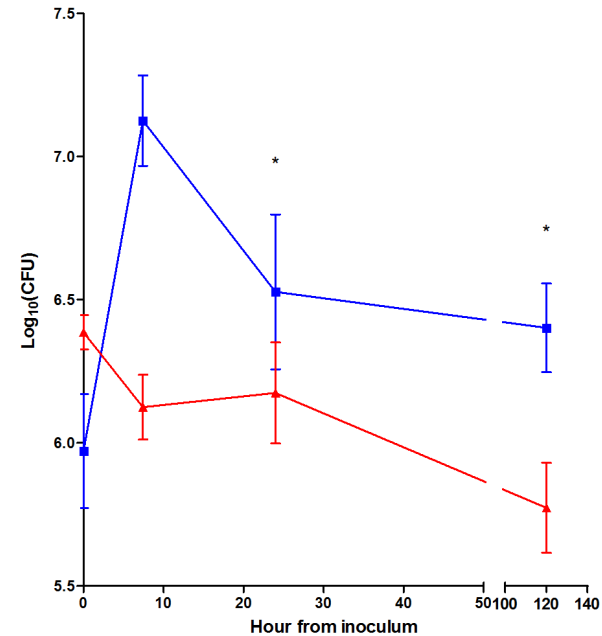


Co-culture/competition



8 replicates (each plotted), 2 isolates

Survival



(Knight, 2013)

> Under a range of mathematical measures: there was significant difference ($\alpha=0.05$)

Fitness differences within MRSA populations

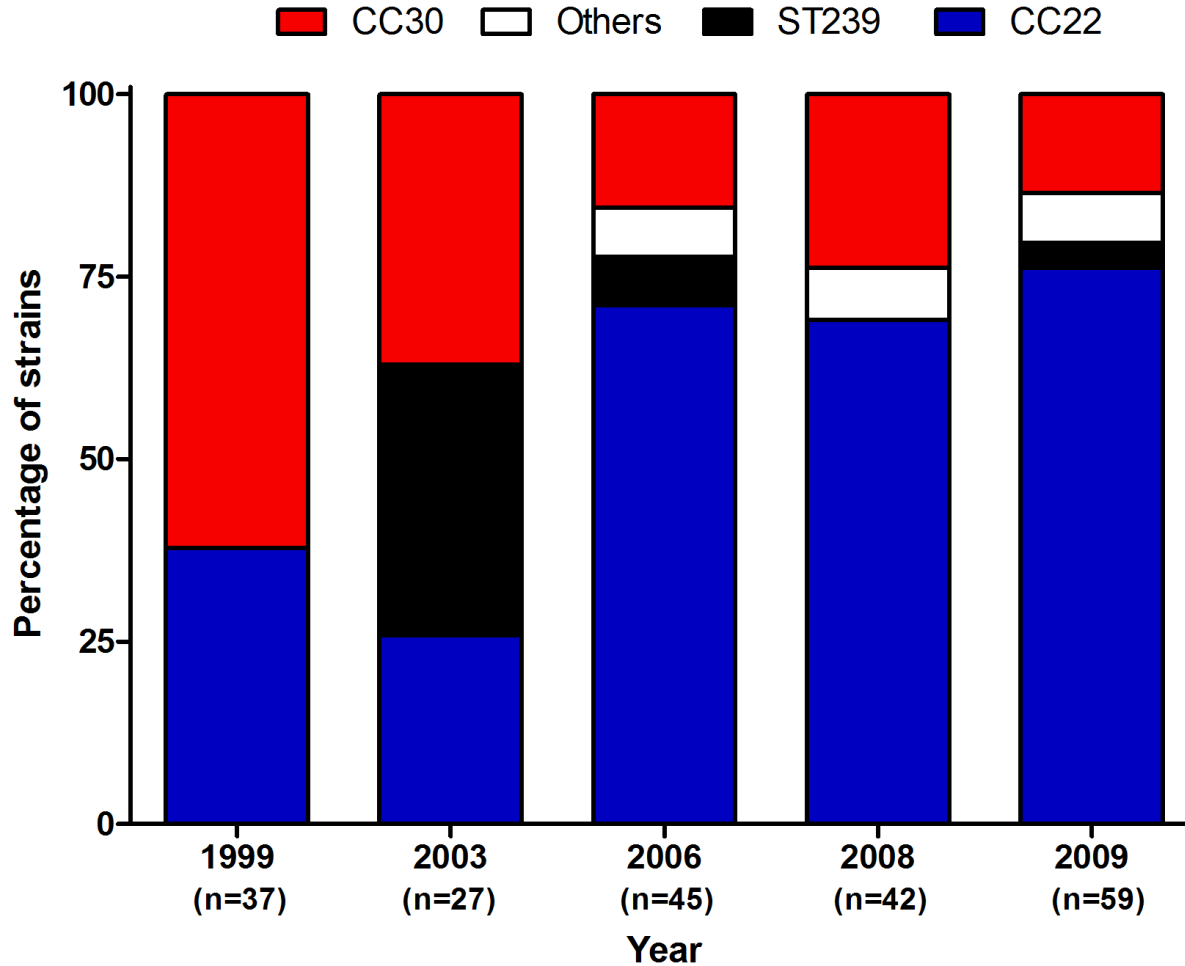
1. Antibiotic resistance had little effect on fitness
 - even when due to large plasmid carriage
 - across a range of different resistances

2. Lineage background had a large impact
 - due to *SCCmec* size differences

What implications does this have for the clinical MRSA population?

Lineage changes in UK

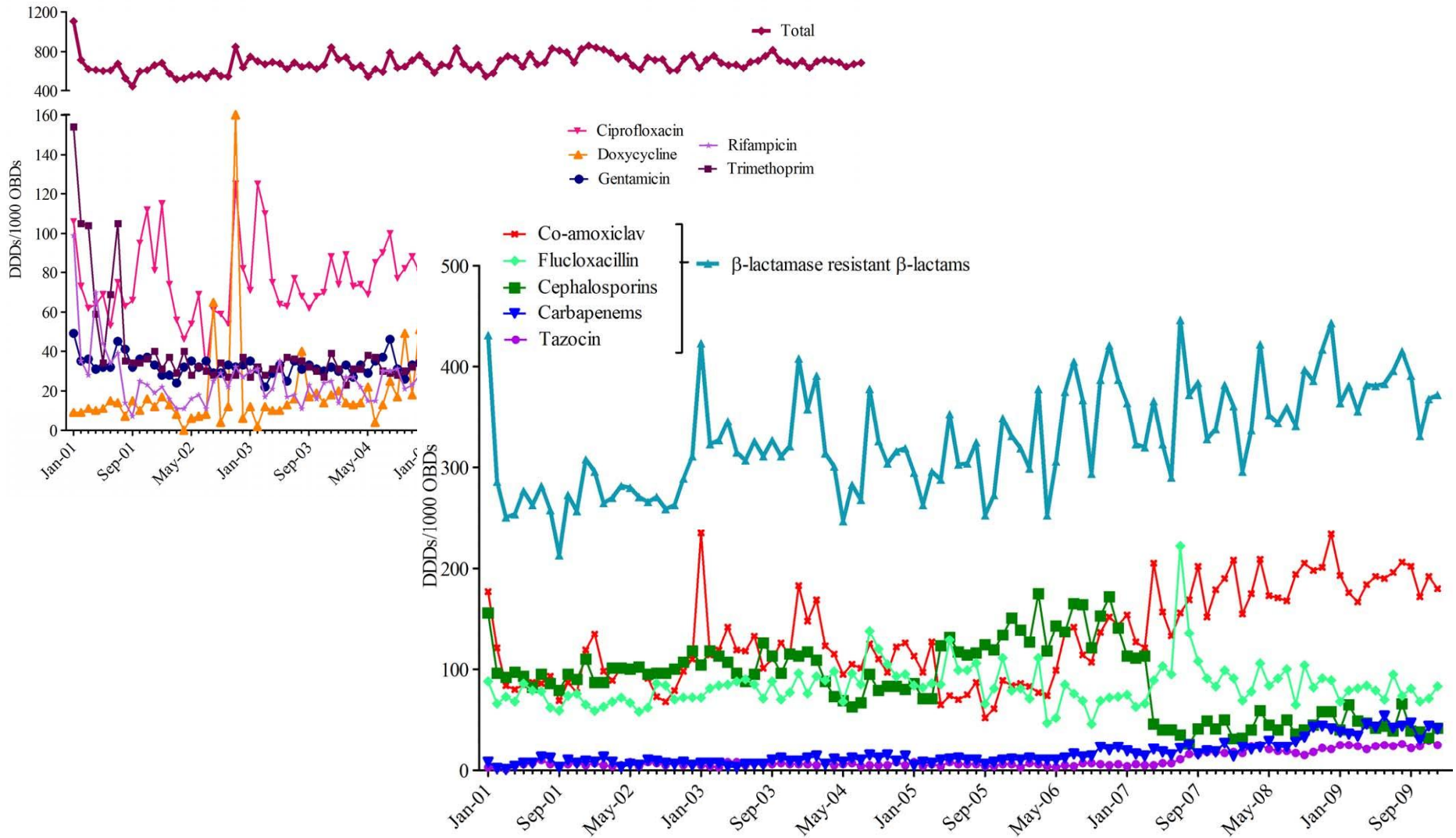
- UK hospital clones: CC22 *SCCmecIV* and CC30 *SCCmecII*¹



1. Ellington (2010)

2. Knight (2012)

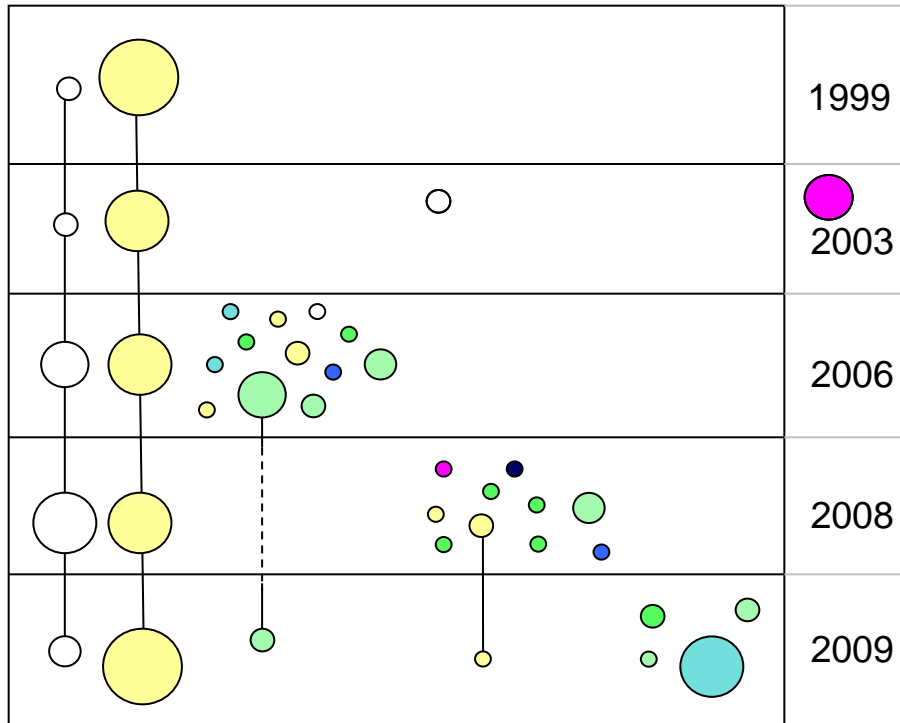
Antibiotic usage roughly constant



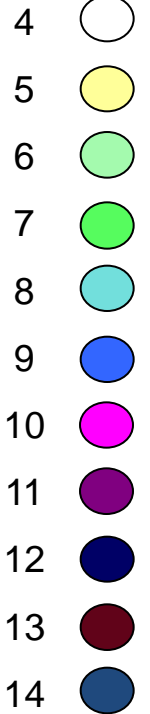
Resistance changes

- Phenotypic resistances (n = 18)

CC22



of Res:

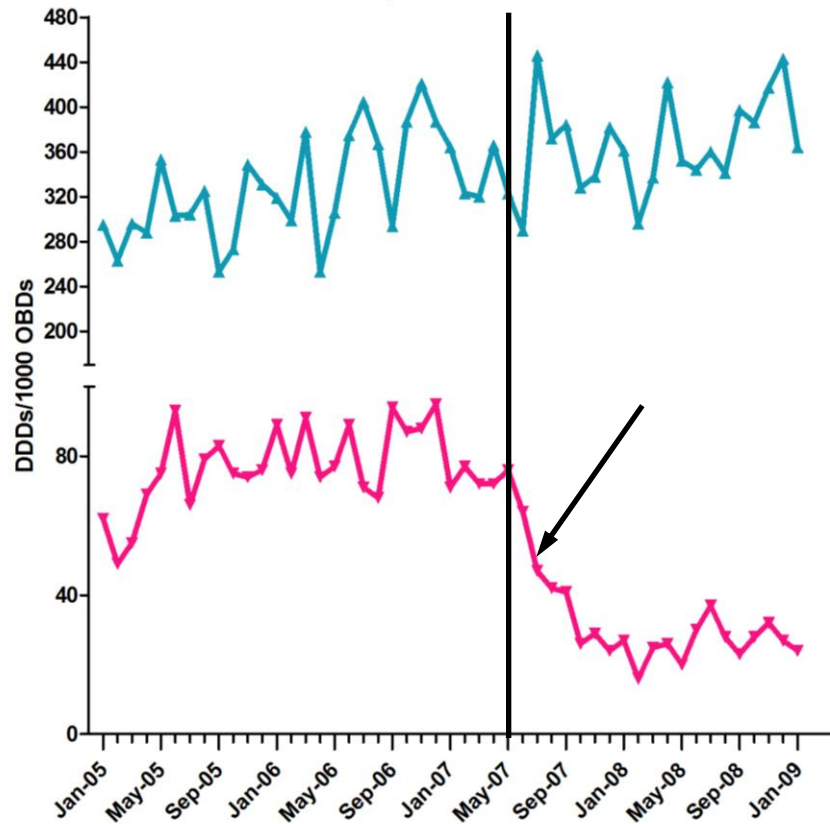


1. Shuffling coincides with success
2. > 99% MRSA resistant to fluoroquinolones

Does hospital wide prescribing of ciprofloxacin (a fluoroquinolone) contribute to the selection of colonizing MRSA?

Controlling MRSA infection rates

“New antibiotic guidelines for the whole Trust with restriction of cephalosporins and quinolones” (Spring 2007)



— Beta-lactamase resistant beta-lactams
— Ciprofloxacin

(Knight, 2012)

Modelling

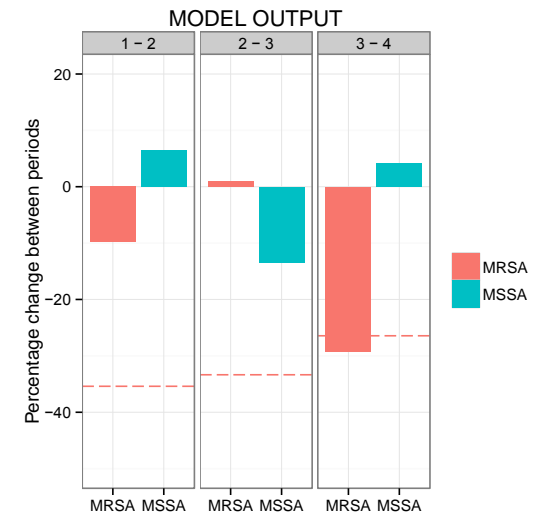
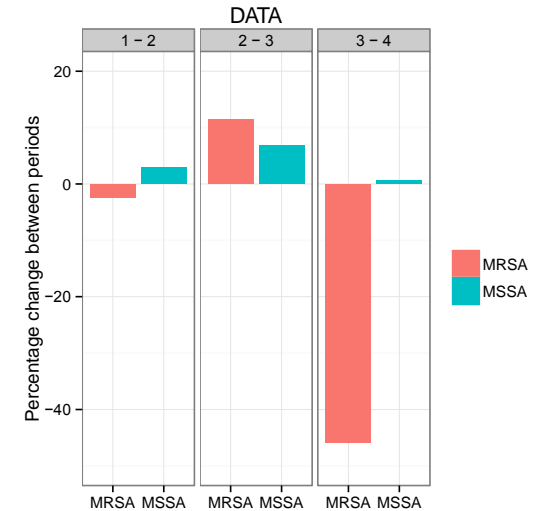
- Test above hypothesis
 - Stochastic differential equation model

$$\frac{dF}{dt} = \lambda_F - \beta FS - (1 + \bar{c})\beta FR + \nu R - \mu F + (a - \bar{c}\beta R)S$$

$$\frac{dS}{dt} = \lambda_S + \beta FS - aS - \mu S - b\beta SR$$

$$\frac{dR}{dt} = \lambda_R + (1 + \bar{c})\beta FR - \nu R - \mu R + (b + \bar{c})\beta SR$$

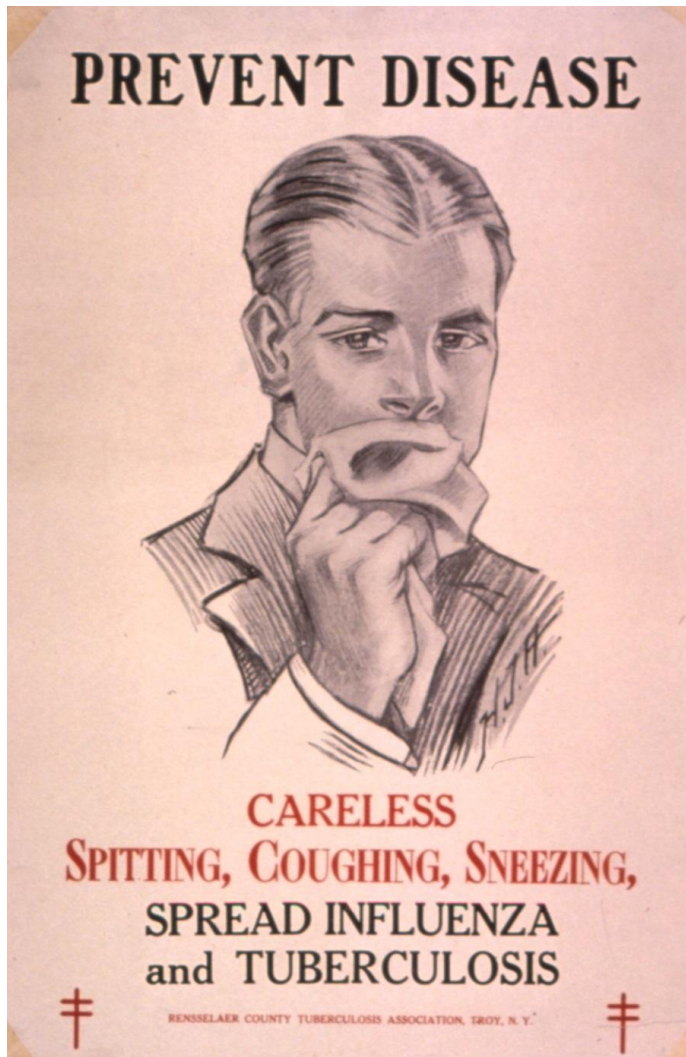
- Parameters from St George's



For, MRSA (UK), the key drivers are
fitness / shuffling / ciprofloxacin resistance

What drives antibiotic resistance changes in TB?

Tuberculosis



EVERY YEAR 9 MILLION
PEOPLE GET SICK WITH TB.

3 MILLION DON'T GET THE
CARE THEY NEED.

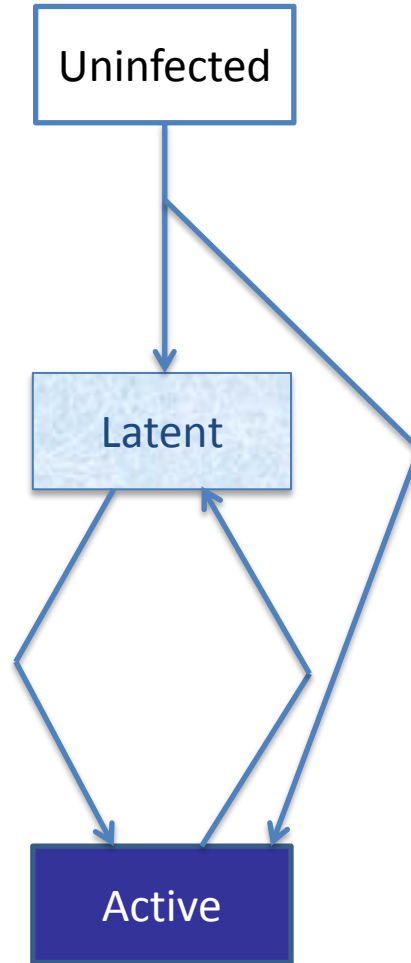
HELP US REACH THEM.

**REACH THE
3 MILLION.**

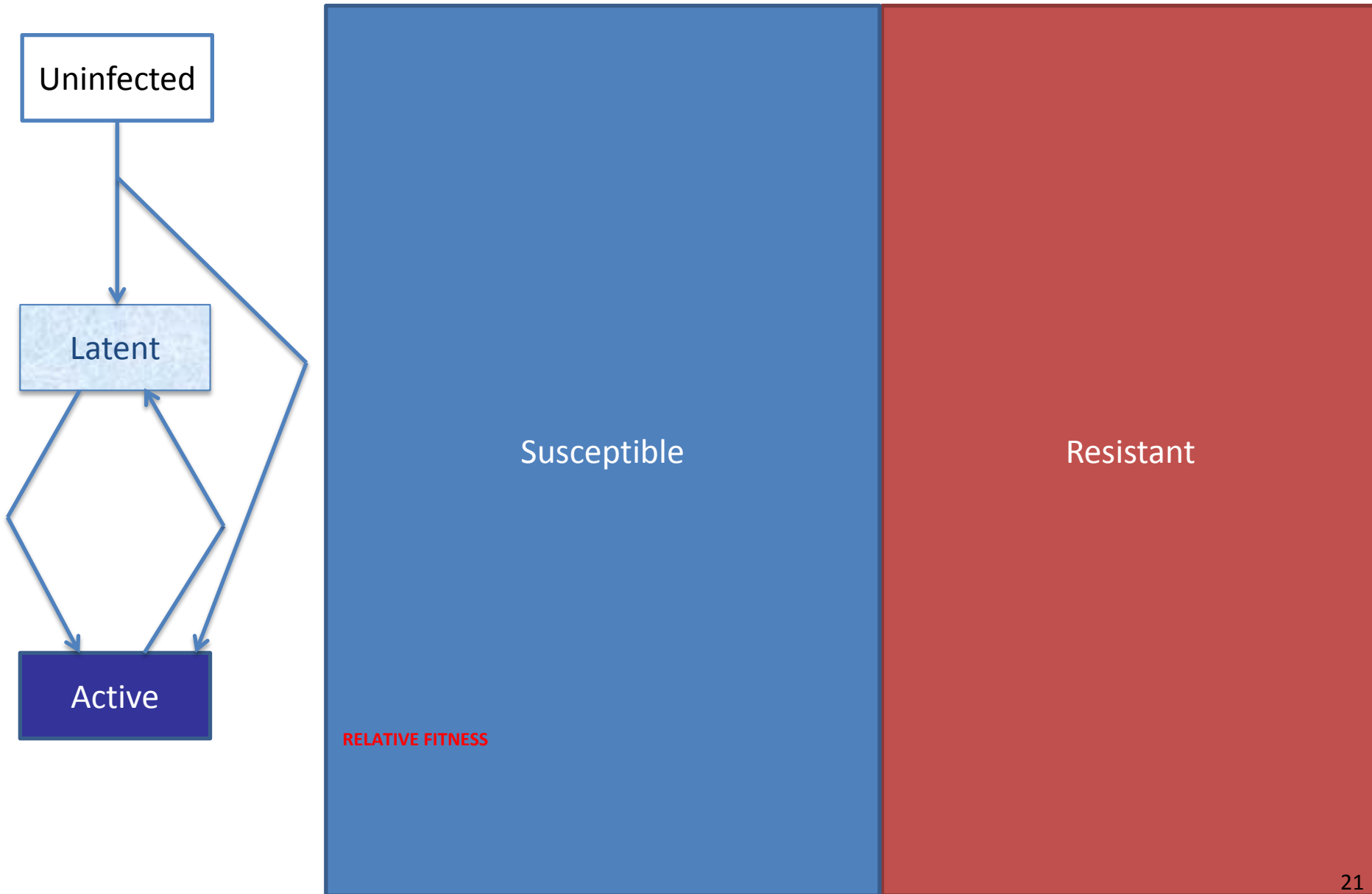
FIND. TREAT. CURE TB.

A modern poster with a solid red background. The text is in white, sans-serif fonts. At the top, it says 'EVERY YEAR 9 MILLION PEOPLE GET SICK WITH TB.' followed by '3 MILLION DON'T GET THE CARE THEY NEED.' and 'HELP US REACH THEM.' In the center is a large, stylized white logo that combines the letters 'T' and 'B'. Below the logo, the text 'REACH THE 3 MILLION.' is written in a bold font, followed by 'FIND. TREAT. CURE TB.' in a smaller font.

Natural history of TB

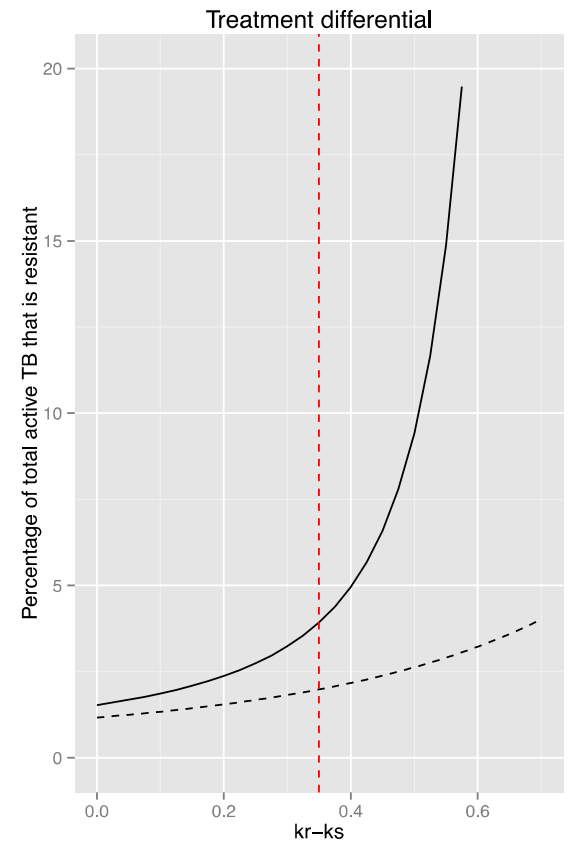
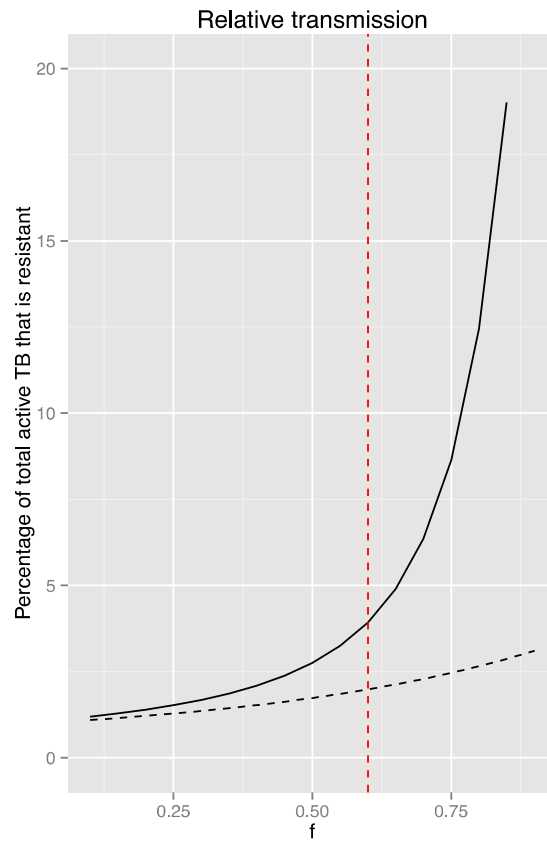
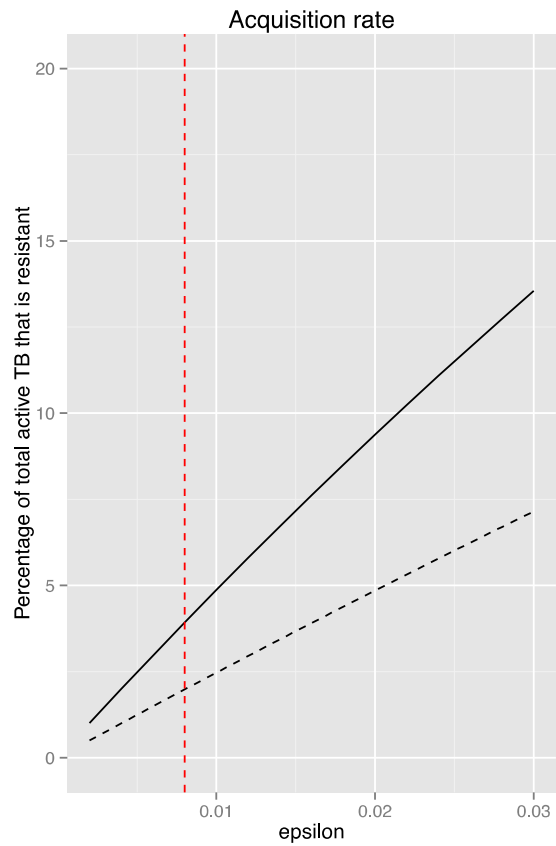


Typical TB model structure



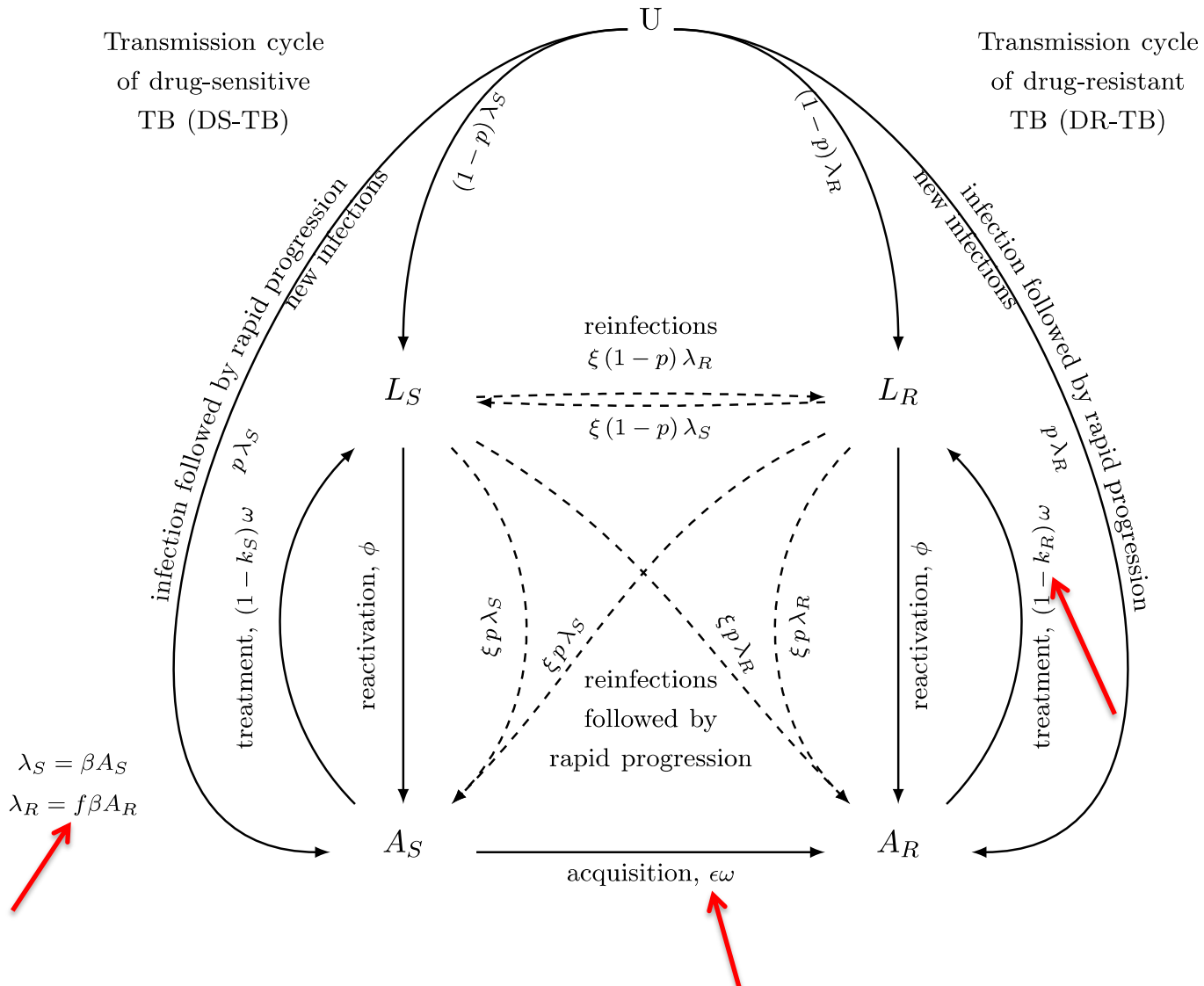
What are the key drivers of TB resistance evolution?

The key drivers from modelling



--- 5 years
— 50 years

The key drivers



What do we know about resistance in TB?

Fitness experiments

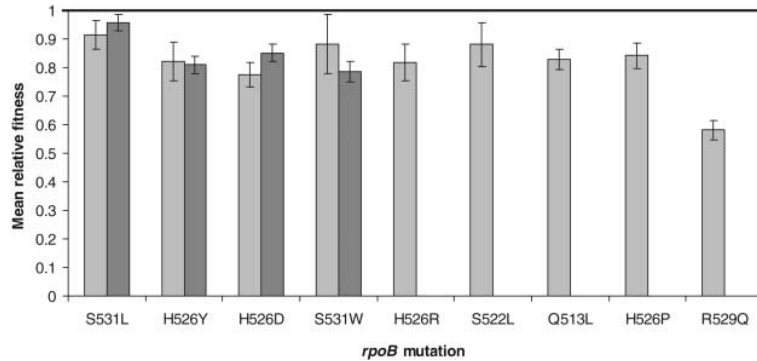


Fig. 1. Relative competitive fitness of laboratory-derived rifampin-resistant mutants of *M. tuberculosis*. All mutants had a statistically significant fitness cost (error bars indicate 95% confidence intervals). This cost was less in *rpoB* S531L mutants than in other *rpoB* mutants, irrespective of the strain background. Light gray bars, CDC1551 mutants; dark gray bars, T85 mutants. Y, Tyr; W, Trp; P, Pro.

(Gagneux, 2006)

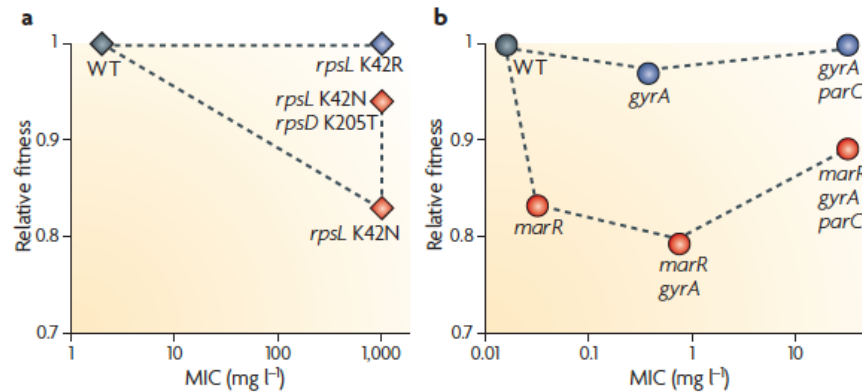
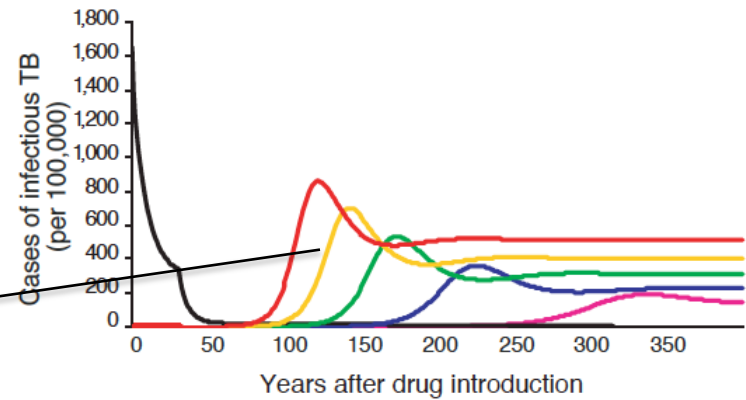
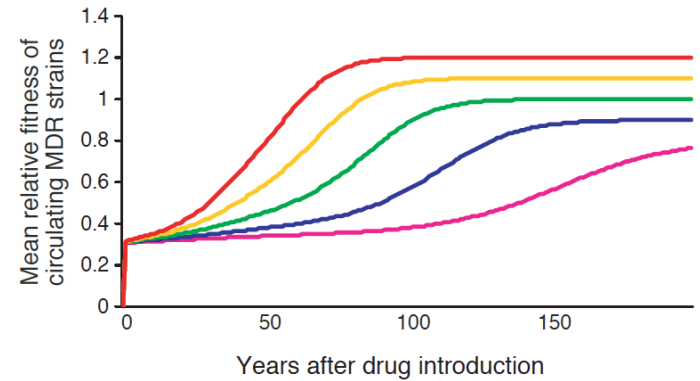
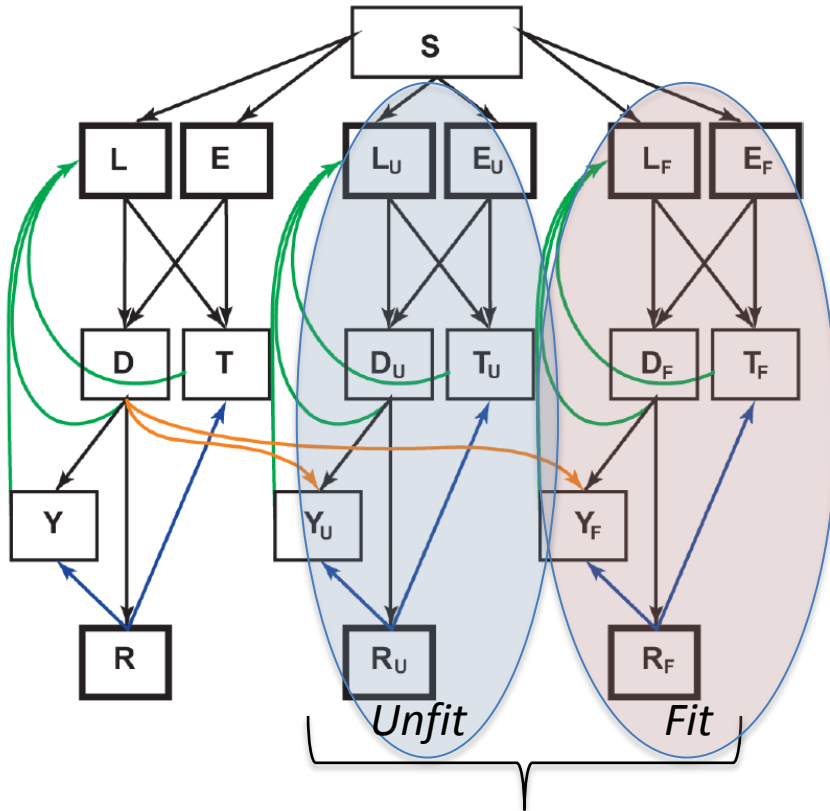


Figure 3 | Relationship between antibiotic resistance and bacterial fitness. Numbers shown on the y axes are from empirical data^{28,61,88}. **a** | High-level resistance to

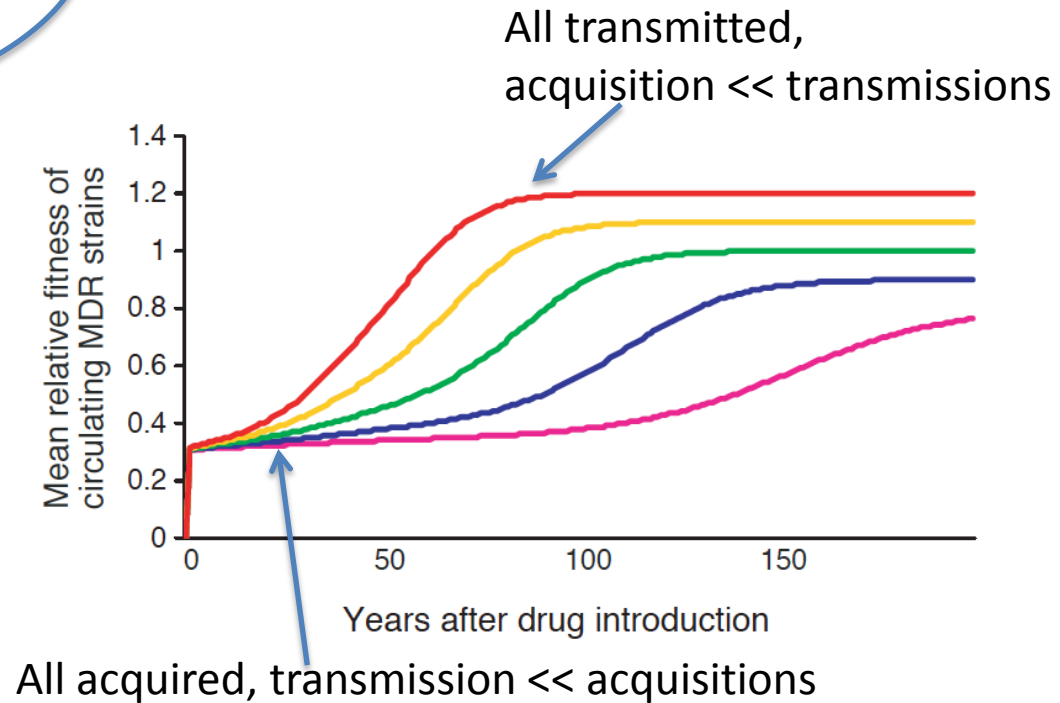
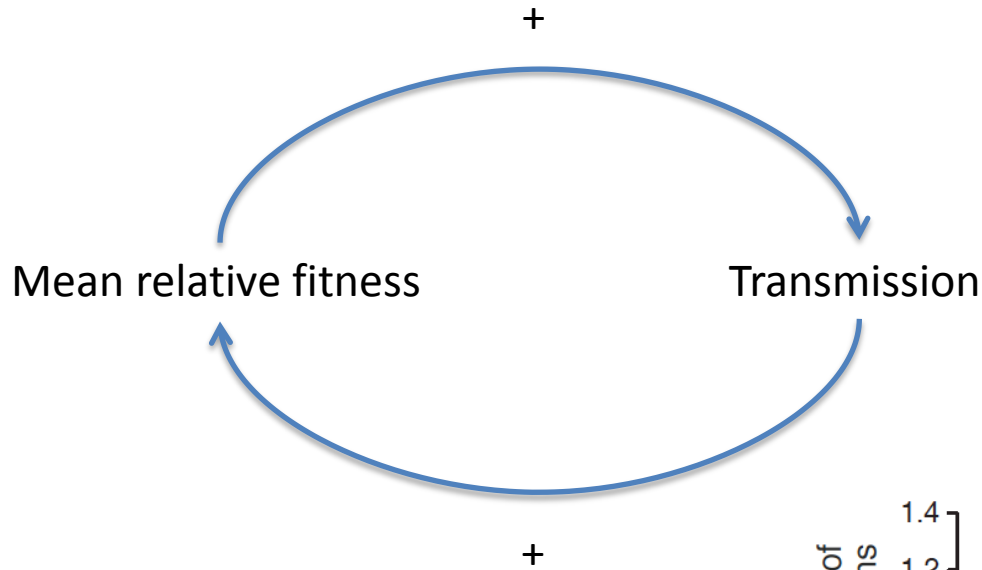
(Andersson, 2010 & Comas, 2012)

Modelling heterogeneous fitness



(Cohen & Murray, 2004)

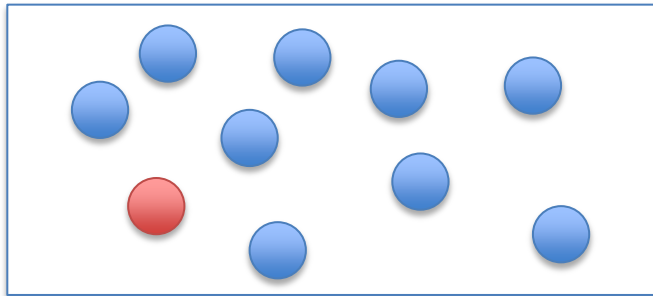
How does mean relative fitness change over time?



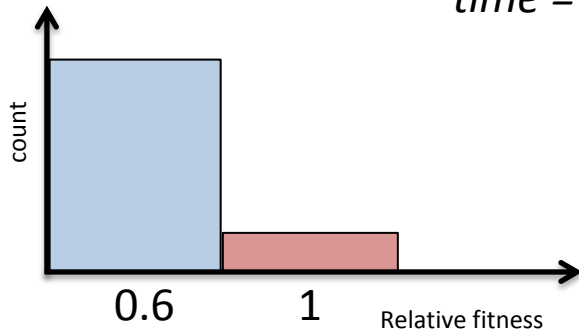
Are “generalized” functions of fitness over time achievable?

Heterogeneous fitness

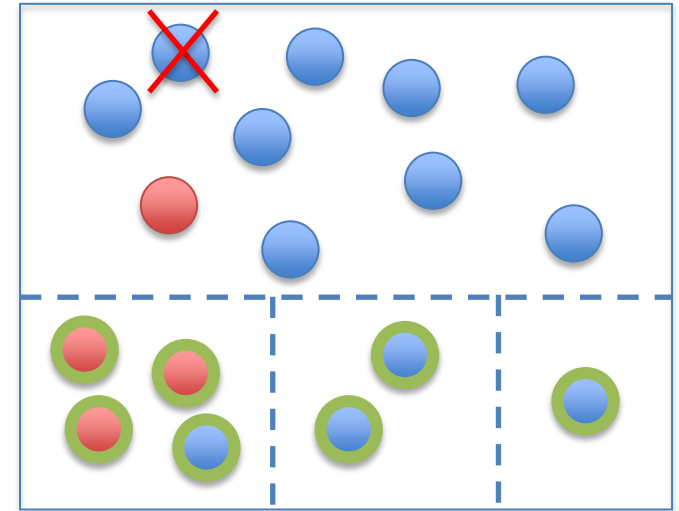
Number of individuals with active TB due to resistant strain



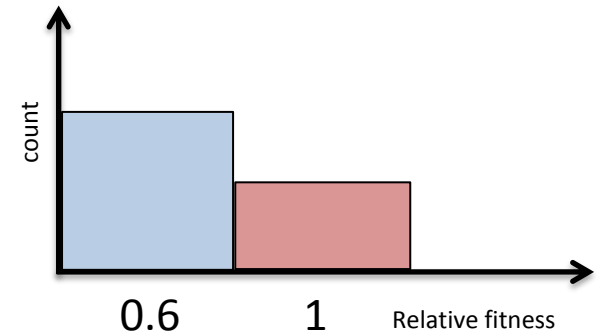
time = t



Mean relative fitness = 0.64

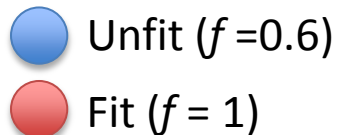


time = t+1



Mean relative fitness

$$\begin{aligned} &= 12/16 \times 0.6 + 4/16 \times 1 \\ &= 0.7 \end{aligned}$$

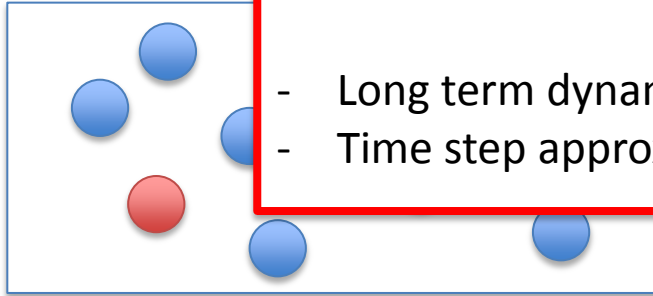


Heterogeneous fitness

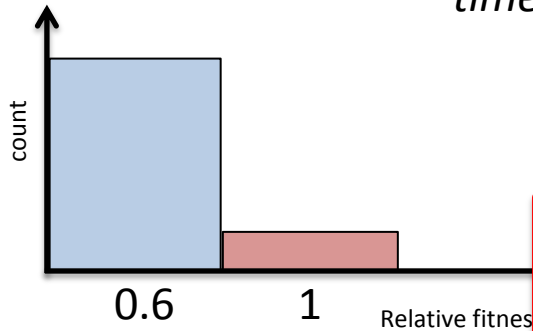
Number of individuals with active TB due to resistant strain

Challenges

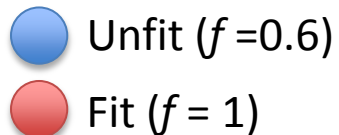
- Long term dynamics of TB
- Time step approximation



time = t

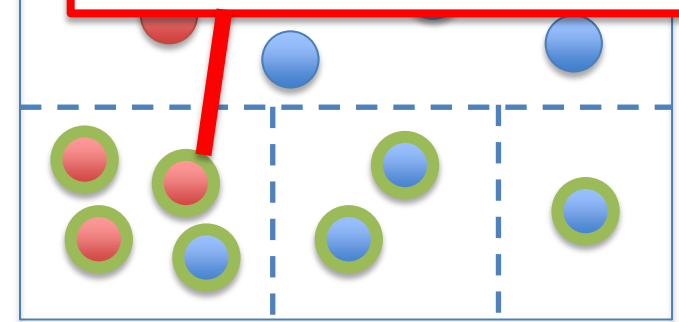
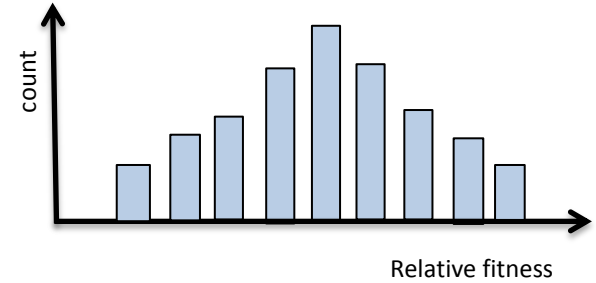


Mean relative fitness = 0.64



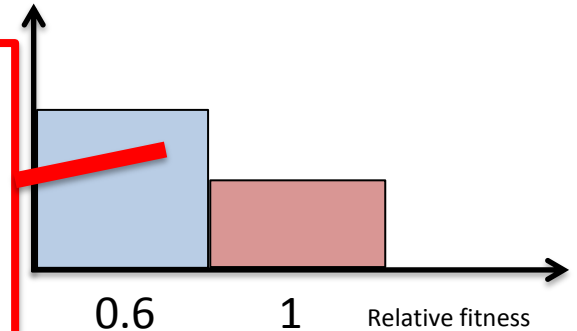
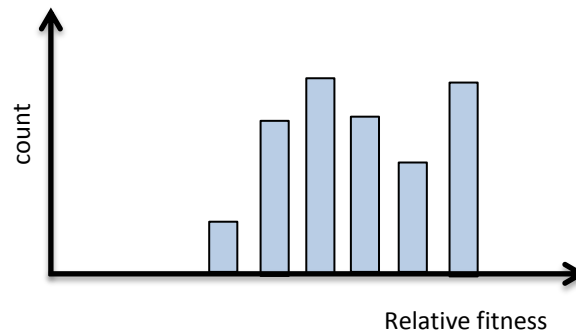
Acquisition
Reactivation

Select acquisitions from different distributions



time = t+1

Multiple fitness levels

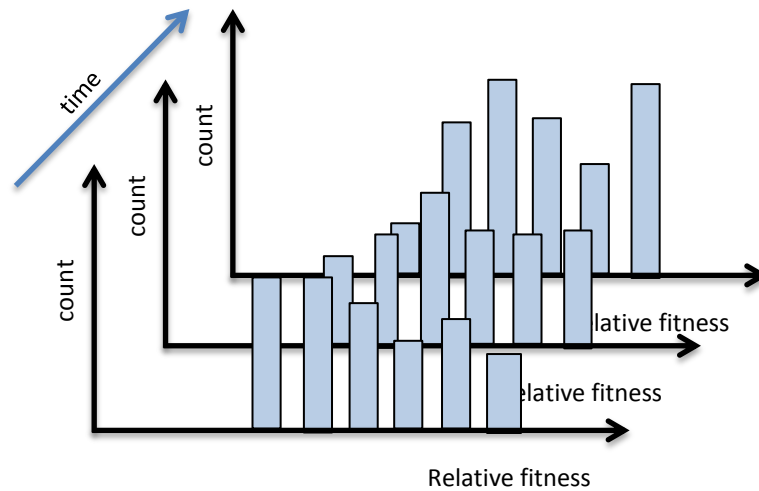


Mean relative fitness
 $= 12/16 \times 0.6 + 4/16 \times 1$
 $= 0.7$

Generalised fitness function

- Proportion at fitness level “x” (just transmission)

$$\begin{aligned}x(t+1) &= \frac{Z(t)x(t)r_x}{Z(t)(x(t)r_x + y(t)r_y)} \\ &= \frac{x(t)r_x}{x(t)r_x + y(t)r_y} \\ &= \frac{x(t)r_x}{f(t)}\end{aligned}$$



$$\begin{aligned}\lambda_S &= \beta A_S \\ \lambda_R &= f\beta A_R\end{aligned}$$

Generalised fitness function

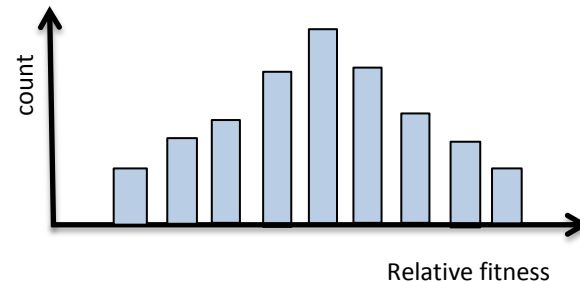
- $M(n,t)$ = matrix of distribution over n fitness levels over time for ACTIVE cases
- v = vector of n relative fitness levels

- Mean relative fitness:

$$f(t+1) = M(n,t+1)v$$
$$= (\text{Active remain}) * M(t) + (\text{Active$$

Dependent on the relative fitness in the last time step

Select acquisitions from different distributions

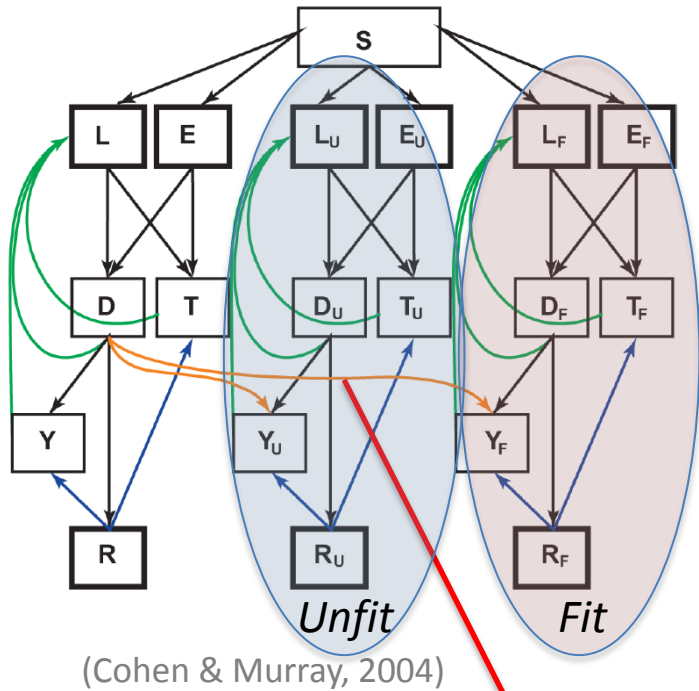


new rel. fit = transmissions x rel. fit + acquisitions x rel. fit

+ reactivations x rel. fit

Requires keeping track of latent population distribution of fitness

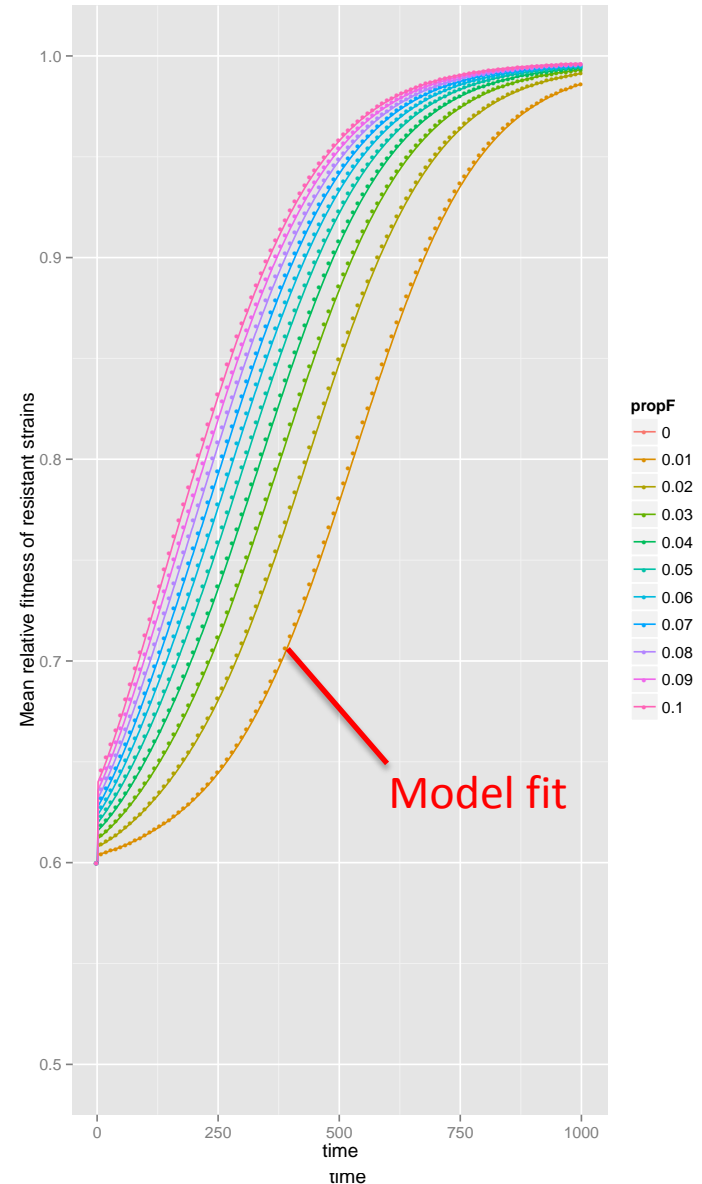
Model fit



Proportion of acquisitions with fit strain = propF

Next steps: Investigate impact of

- Different distributions of fitness costs to acquisitions
- Stochastic effects (small population sizes and extinction)
- Compensatory mutations



lel fit

Conclusions

- MRSA
 - Resistance levels give information on the selective pressures seen in the environment e.g. on commensals
 - Three key drives to being a successful resistant MRSA isolate in a UK hospital:
 - Fitness / Shuffle / ciprofloxacin resistance
 - The most successful were not the most resistant
- TB
 - Levels of resistance to new regimens will be governed by
 - Fitness costs to resistance / treatment success / acquisition rates
 - To be continued...
- Fitness is a dynamic process
 - More data is needed on how this changes in populations over time

Acknowledgements

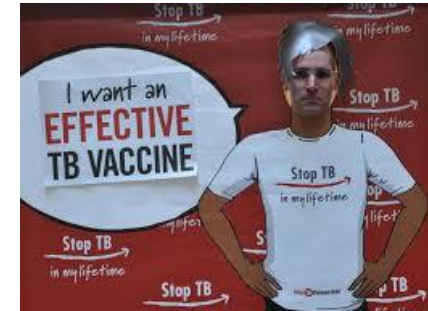
- MRSA
 - Jodi Lindsay
 - Emma Budd
 - St George's Healthcare NHS Trust



UCL CoMPLEX



- Tuberculosis
 - Ted Cohen
 - Richard White
 - David Dowdy
 - Sourya Srestha
 - Mariam Fofana
 - Frank Cobelens

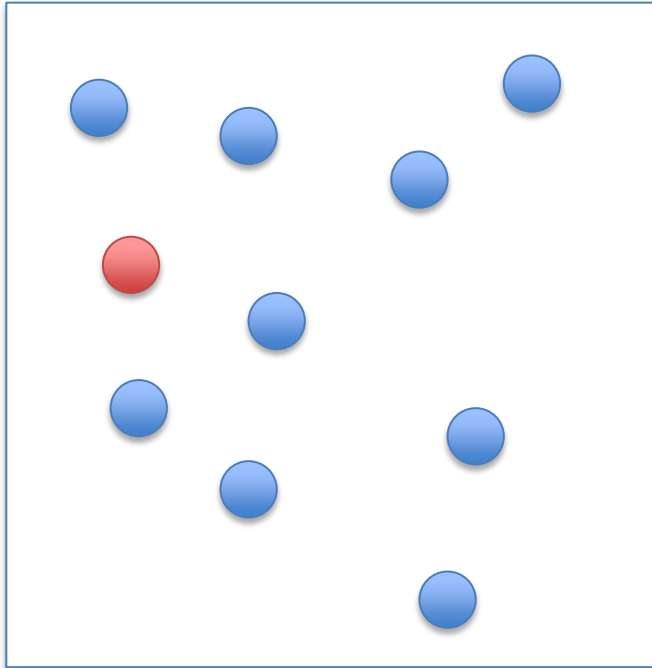


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BILL & MELINDA
GATES foundation

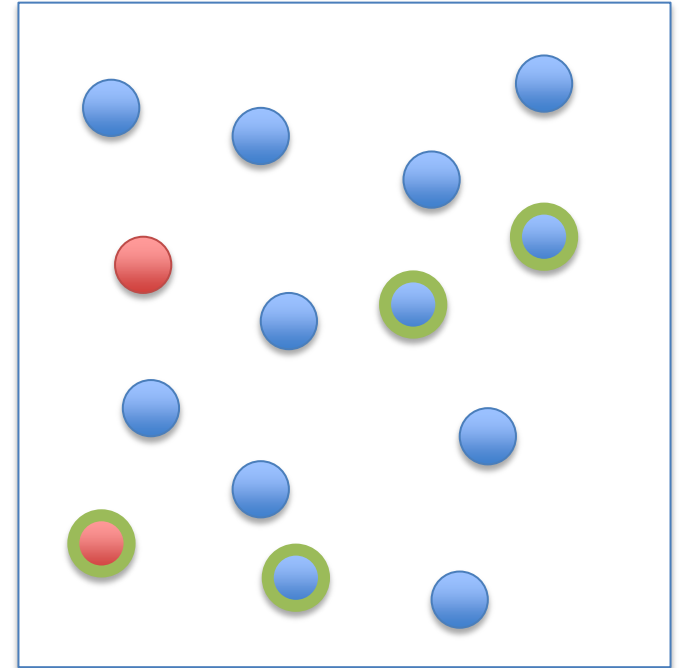
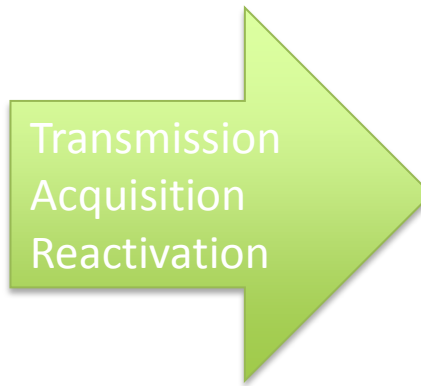
Heterogeneous fitness

Number of individuals with active TB due to resistant strain

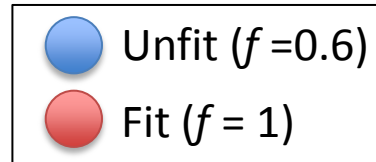


Mean relative fitness
= $0.9 \times 0.6 + 0.1 \times 1$
= 0.64

$$\lambda_S = \beta A_S$$
$$\lambda_R = f \beta A_R$$



Mean relative fitness
= $12/14 \times 0.6 + 2/14 \times 1$
= 0.66

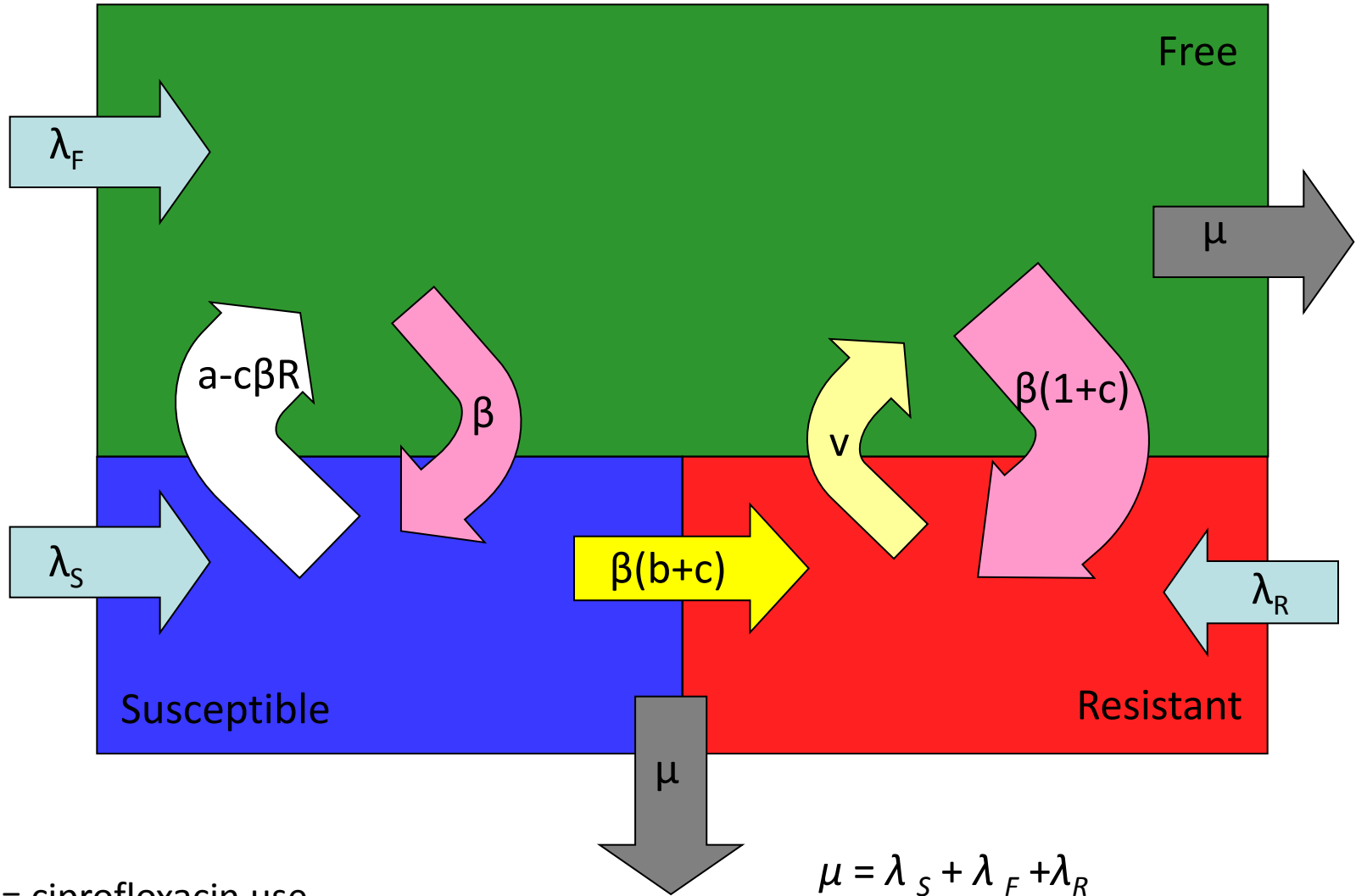


Treatment and antimicrobial resistance in TB

- First line: 6 months combination therapy
- Resistance:
 - MDR-TB = resistance to two of first line drugs
 - XDR-TB = MDR + resistance to any of second line
- 4% of new and 20% of previously treated MDR-TB¹
- >70% undetected¹
- MDR-TB treatment 24months...
- New regimens under investigation
 - Shorter
 - New drugs



Modelling explanation

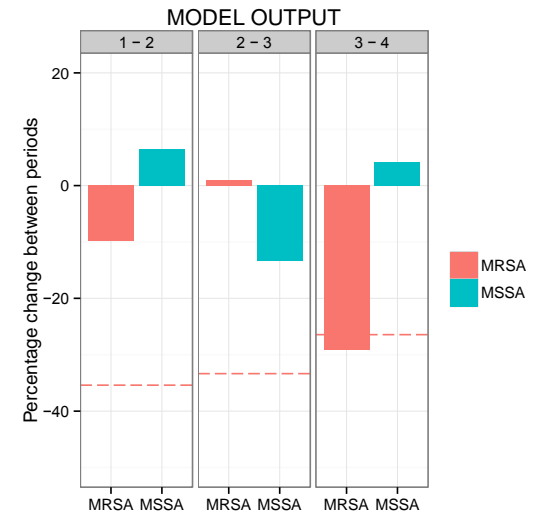
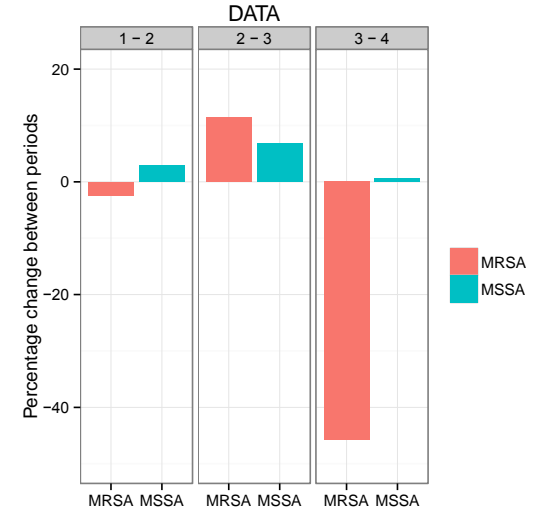


c = ciprofloxacin use

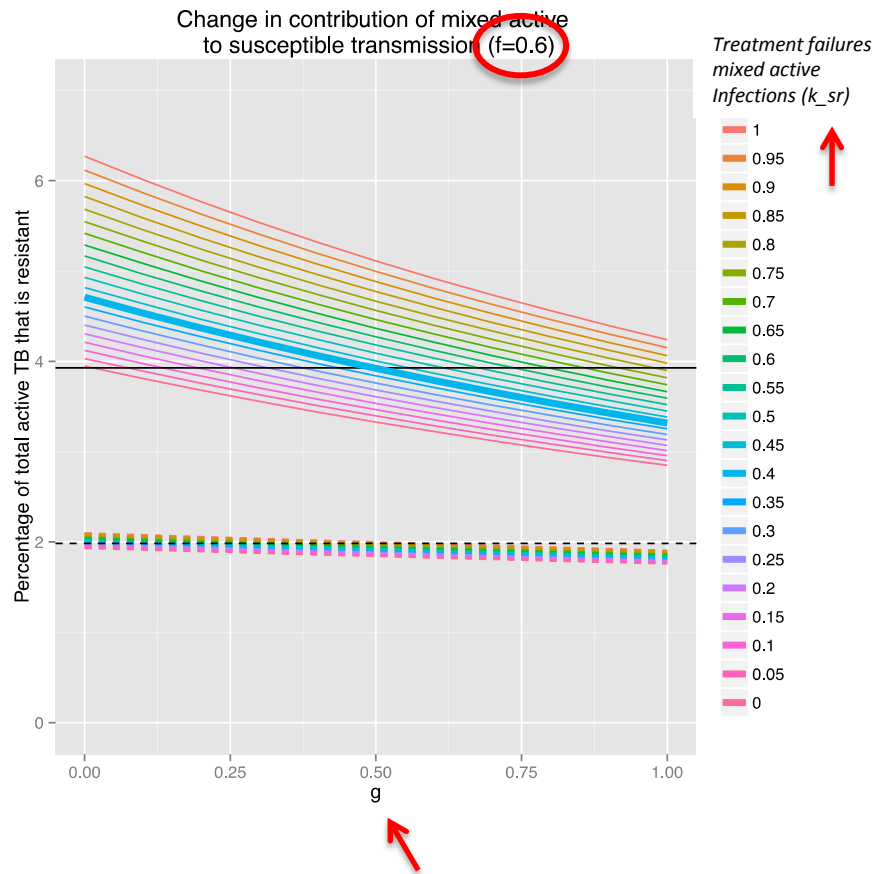
$$\mu = \lambda_S + \lambda_F + \lambda_R$$

Modelling explanation

- Using hospital parameters the effect of the decline in fluoroquinolones can be captured
- Using to make predictions about other control mechanisms



Dependence on relative fitness



SS model (no MI)

----- 5 years

————— 50 years

$$\lambda_S = \beta(A_S + gA_{SR})$$

$$\lambda_R = f\beta(A_R + (1 - g)A_{SR})$$

fitness for the levels of (r_x, r_y) . If $Z(t)$ is the pool of individuals that can be infected via transmission of resistant strains multiplied by the *per capita* transmission rate (β) and proportion that progress to active TB p , then the number of transmissions T at time $t + 1$ is:

$$T(t + 1) = Z(t)(x(t)r_x + y(t)r_y) \quad (3)$$

and, as a reworking of (2), the mean relative fitness of the resistant strain population at time t is

$$f(t) = (x(t)r_x + y(t)r_y) \quad (4)$$

This is the fitness value used to determine the number of transmissions of the resistant strains in time step $t + 1$.

$$\begin{aligned} x(t + 1) &= \frac{Z(t)x(t)r_x}{Z(t)(x(t)r_x + y(t)r_y)} \\ &= \frac{x(t)r_x}{x(t)r_x + y(t)r_y} \\ &= \frac{x(t)r_x}{f(t)} \end{aligned}$$