Evolution of antibiotic resistance in *Pseudomonas aeruginosa*

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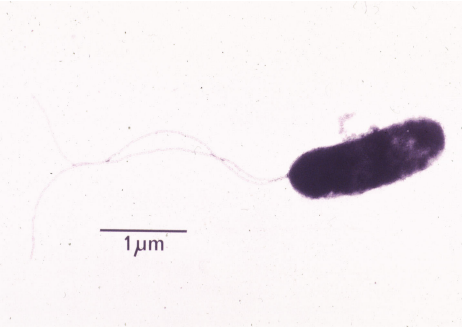
Pseudomonas bacteria

- **Very widespread in the environment**
- **Many species are harmless**
- **Some major plant pathogens eg. PSA**

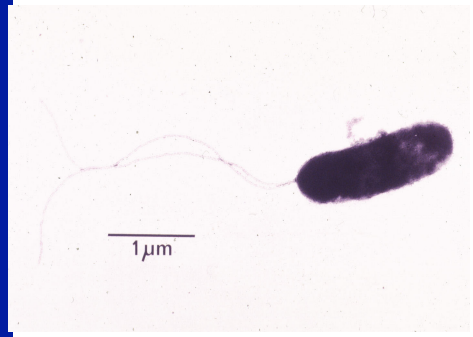


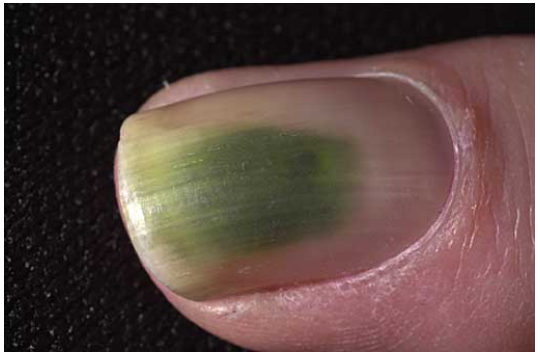
Pseudomonas aeruginosa

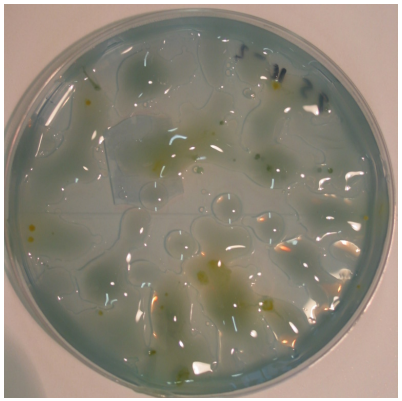
Common in moist environments

Infects animals eg. horses, dogs, cats, sheep

Infects patients with predisposing conditions – chronic and acute infections

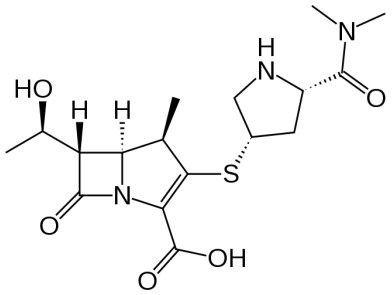


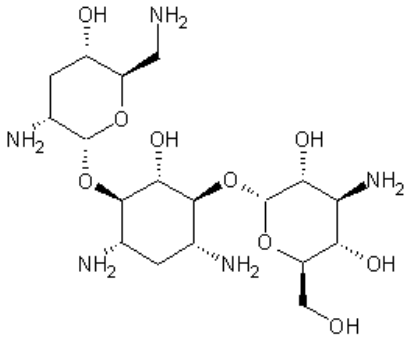


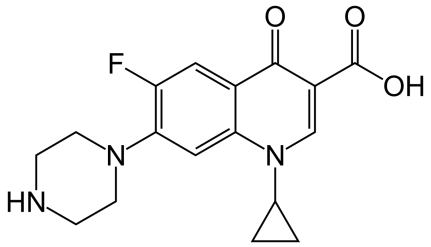


Treating infection - antibiotics

- *P. aeruginosa* has intrinsic low-level resistance







- **Antibiotic resistant strains are now a major problem**

WHO Global priority list of antibiotic resistant bacteria

World Health Organization

Priority 1: CRITICAL

Acinetobacter baumannii, carbapenem-resistant

Pseudomonas aeruginosa, carbapenem-resistant

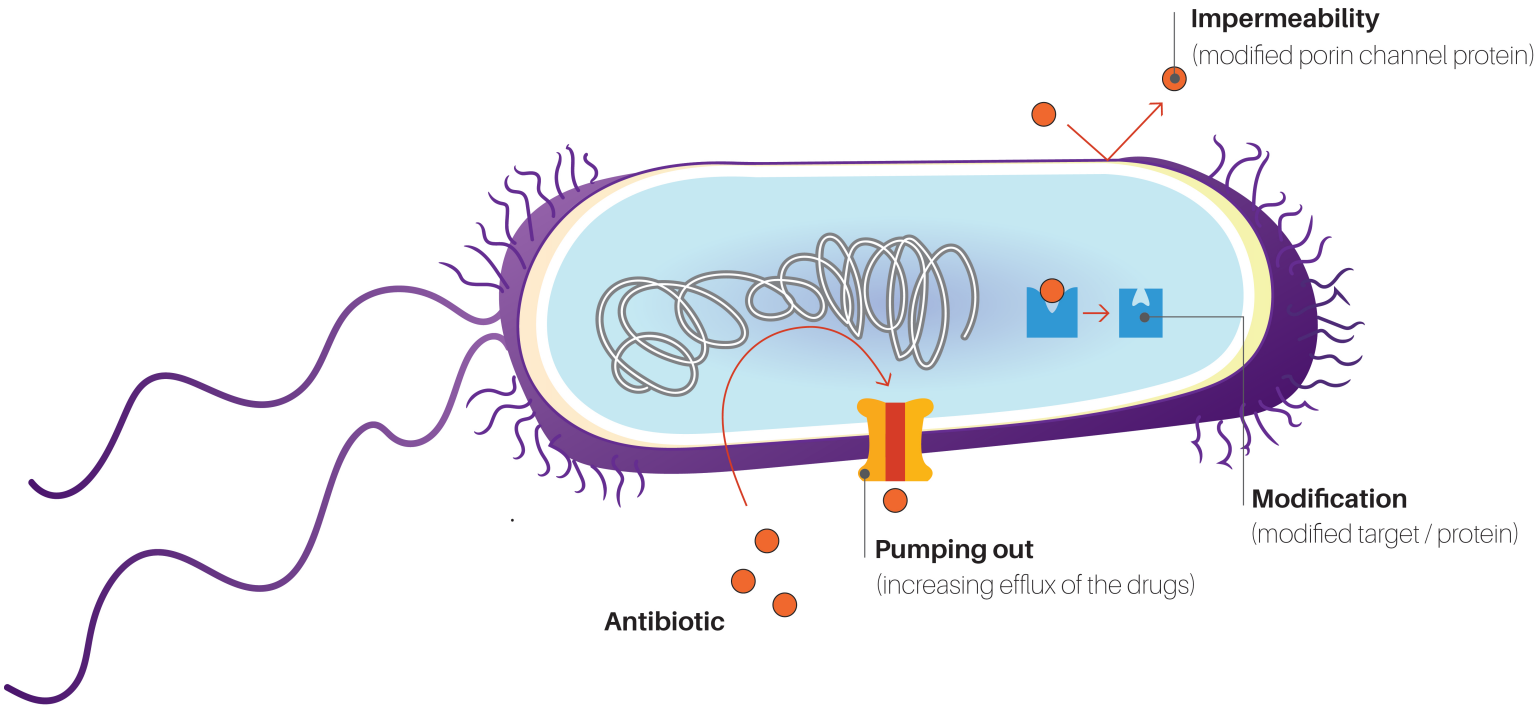
***Enterobacteriaceae*,** carbapenem-resistant, 3rd generation cephalosporin-resistant

How do bacteria become antibiotic resistant?

- Acquire antibiotic resistance genes from other bacteria
- Acquire mutations that confer resistance

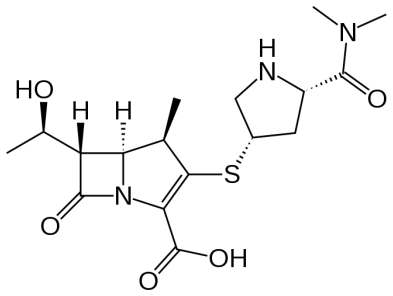
In *Pseudomonas* resistance arises mainly through mutations

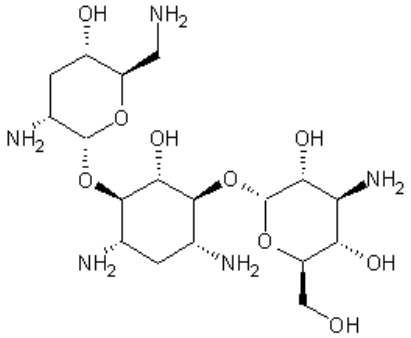
- Mutations reduce uptake, increase efflux or alter target proteins

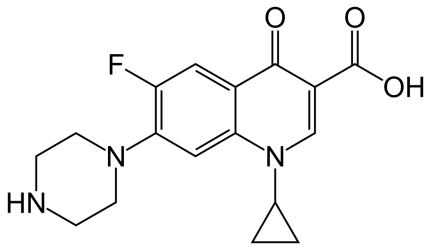


Major goal of our research programme

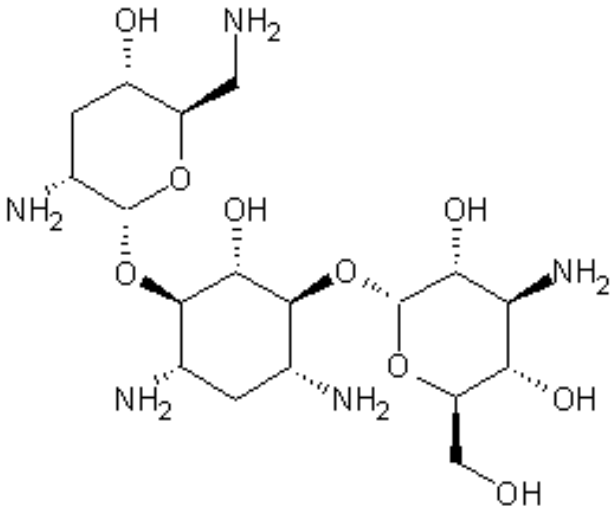
- To understand how *Pseudomonas aeruginosa* survives antibiotic treatment and causes infection





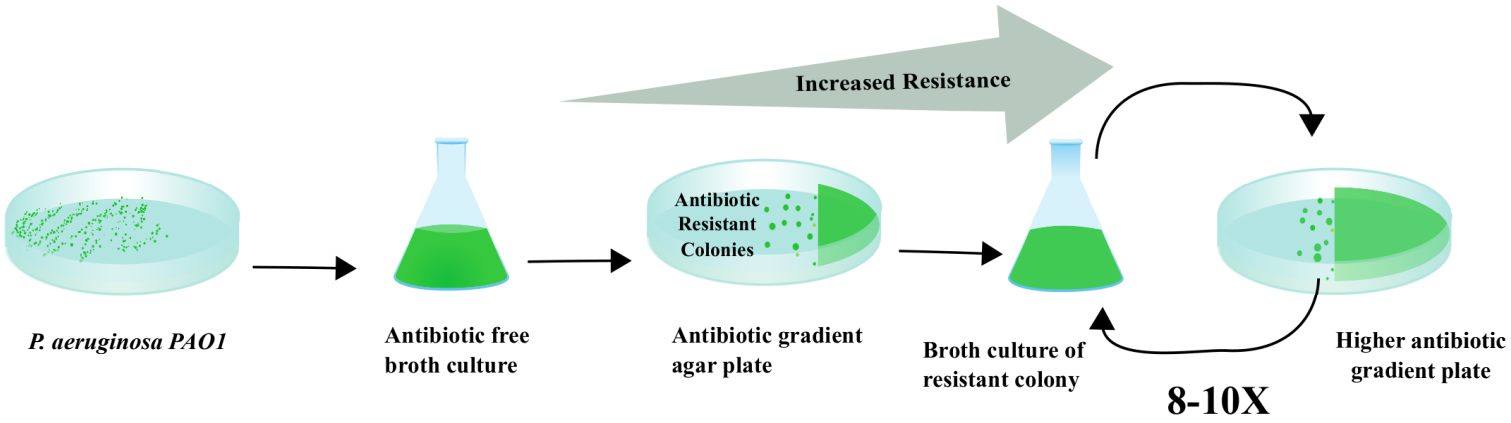


Example: obtaining a complete picture of mutations contributing to antibiotic resistance



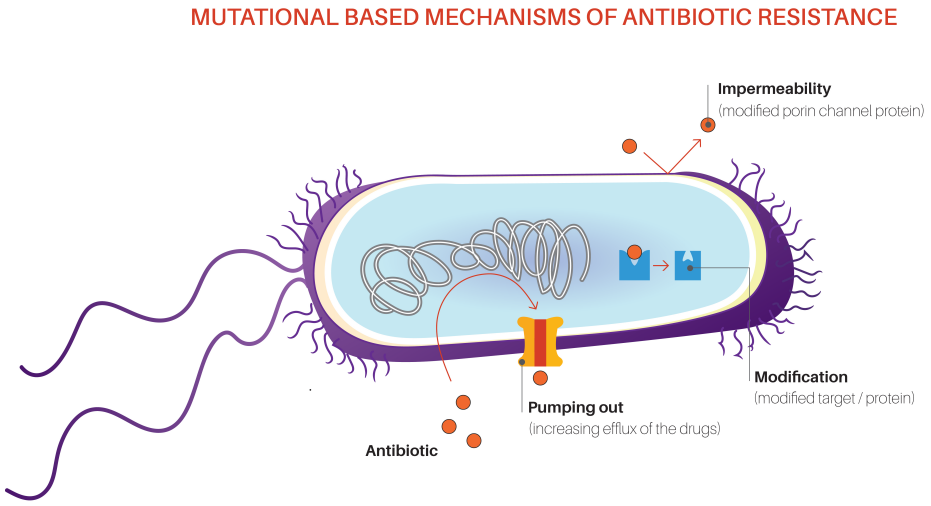
Research strategy

- Evolve highly resistant mutants in the lab
- Use whole genome sequencing to identify the mutations conferring resistance



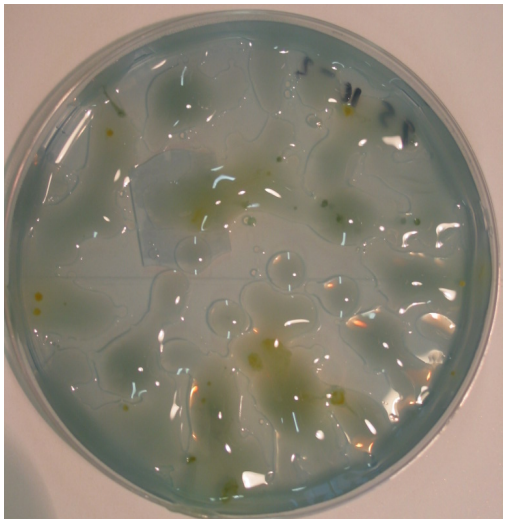
Results

- 15 mutants, at least 32-fold more resistant than wild-type
- Mutated genes are consistent with earlier studies



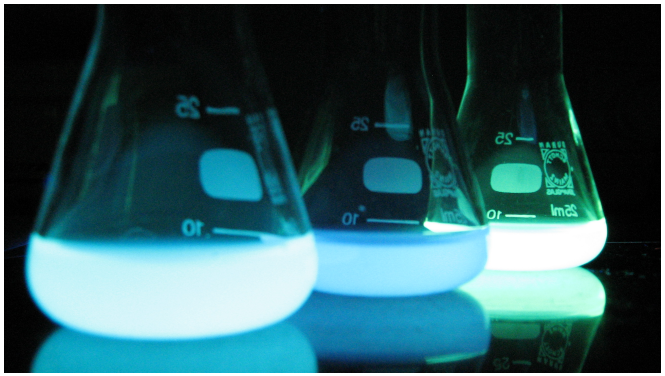
Clinical relevance?

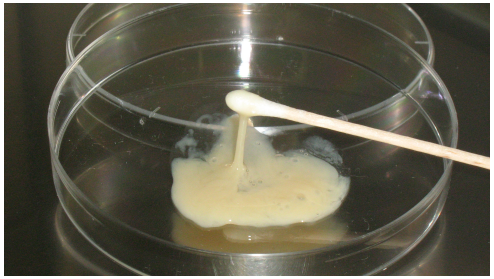
- Analysed 185 clinical isolates for resistance alleles in lab mutants
- Mutations in our *in vitro* study also present in clinical isolates



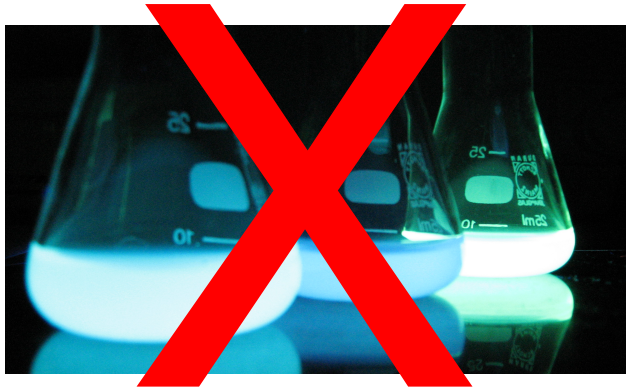
Example two: bacterial physiology during infection

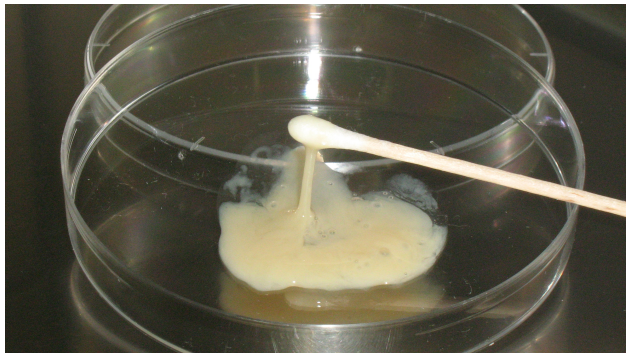
- How active are antibiotic resistance genes during infection?
- Infection: cystic fibrosis





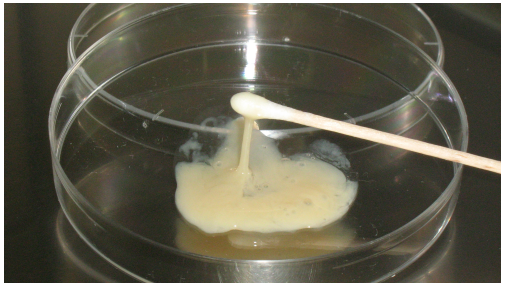
Approach: analyse antibiotic resistance gene activity IN PATIENT SPUTUM





Sputum obtained directly from patients represents infection in the lung

Outcome

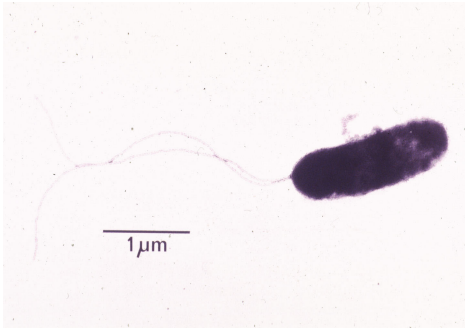


Can quantify antibiotic resistance gene activity in infection (sputum)

WIDE variation in gene expression between patients

Conclusions

- Identified a number of genes not previously associated with resistance
- Mutations in lab-evolved bacteria reflect those that occur during infection
- Antibiotic resistance gene expression shows remarkable variation during infection



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