To die, to sleep...

Что отличает бактерицидные антибиотики от бактериостатических

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Ribosome is the most complex and sophisticated molecular machines in the cell.
Antibiotics could be bacteriostatic or bactericidal

Bacteriostatic drugs stop bacteria from growing but do not kill them. Bacterial cells resume growth after removal of the antibiotic.

Bactericidal drugs kill bacteria.

Bacteria grow antibiotic incubate
The Macrolide Family

Erythromycin (ERY)

Ketolides: Telithromycin (TEL)
Peptide exit tunnel

Catalytic center
Macrolides bind in the peptide exit tunnel
Common wisdom: Macrolides plug the tunnel and block synthesis of all the proteins at early rounds of translation.
Macrolides act as protein- and context-specific inhibitors of translation.

Different macrolides inhibit synthesis of different proteins.

**No antibiotic**  
**ERY**  
**TEL**
Do different macrolides exhibit different killing activity?

*Streptococcus pneumoniae*
Ery is static, but Tel is highly cidal.
Static ERY and cidal TEL inhibit synthesis of different proteins.
Hypothesis 1: 
Residual translation defines cidality
Does macrolide-induced cidality require protein synthesis?

Tetracycline → Tet (static)
Telithromycin → Tel (cidal)

[Graph showing CFU/mL over incubation time]

- x40 MIC Tet
- no drug
- x40 MIC Tet

KILL TEST

protein synthesis is required for cidality

cidality does not depend of residual translation
Tel remains bactericidal even cells could make no proteins

Hypothesis 1: Residual translation

![Graph showing CFU/mL vs. Incubation time, hrs for Tet and Tel treatments](attachment:graph.png)
Hypothesis 2:
Affinity of the drug for the ribosome defines cidal activity
Ribosome $^{14}C$-Erythromycin

DEAE magnetic beads
Equilibrium affinity of bactericidal SOL is comparable to the affinity of bacteriostatic ERY.

Hypothesis 2: Affinity of the drug for the ribosome defines cidality.
Hypothesis 3:
Kinetics of the drug dissociation defines cidality

Drugs with similar affinities could have significantly different rates of binding and dissociation

\[
[AB] \xrightleftharpoons{k_d}{k_a} [A] + [B] \quad K_d = \frac{[A][B]}{[AB]}
\]

\[K_d = \frac{k_d}{k_a}\]
Measurement of dissociation rate

Bactericidal drug dissociates very slowly from the ribosome!
Slow-dissociating drugs are bactericidal; fast-dissociating drugs are bacteriostatic.

The slow off-rate requires an extended side-chain.
Side chain makes specific contacts with the ribosome
Selection of mutants that can resist killing

Few surviving cells
\((10^{-3} - 10^{-4})\)
Mutant cells are susceptible to solithromycin

Bactericidal Solithromycin stop mutant cells from growing but does not kill them!
Most likely the dissociation of SOL from the mutant ribosomes is fast
WHY DO SLOWLY DISSOCIATING ANTIBIOTICS KILL BACTERIA?
Steady-state level of every protein is defined by the rates of synthesis and degradation.

If the dormant cell runs out of any of the key factors, translation and cell growth cannot be restarted!
Lack of any key component will prevent translation from restarting (or cell from re-growing and dividing)
During prolonged period of dormancy the cell runs out of one or several critical proteins.
Bacteriostatic antibiotics kill cells upon the long enough exposure.
Implications:

- Antibiotics need to be optimized not only for their affinity but also for the proper kinetics.

Questions to address:

- What is the limiting factor?
- Could we convert bacteriostatic antibiotics into bactericidal if we inhibit the ‘limiting factor’?