Activities of amoxicillin, clarithromycin and moxifloxacin towards intracellular forms of *S. pneumoniae* with increasing MICs: attempt at defining an intracellular susceptibility breakpoint

Sandrine Lemaire,[1] Mark P.G. van der Linden,[2] and Paul M. Tulkens[1]

[1] Pharmacologie cellulaire et moléculaire,
Louvain Drug Research Institute,
Université catholique de Louvain, Brussels, Belgium

[2] Nationales Referenzzentrum für Streptokokken Abteilung
Medizinische Mikrobiologie, Universitätsklinikum RWTH,
Aachen, Germany

23d European Congress of Clinical Microbiology
and Infectious Diseases (ECCMID)

Content of the presentation

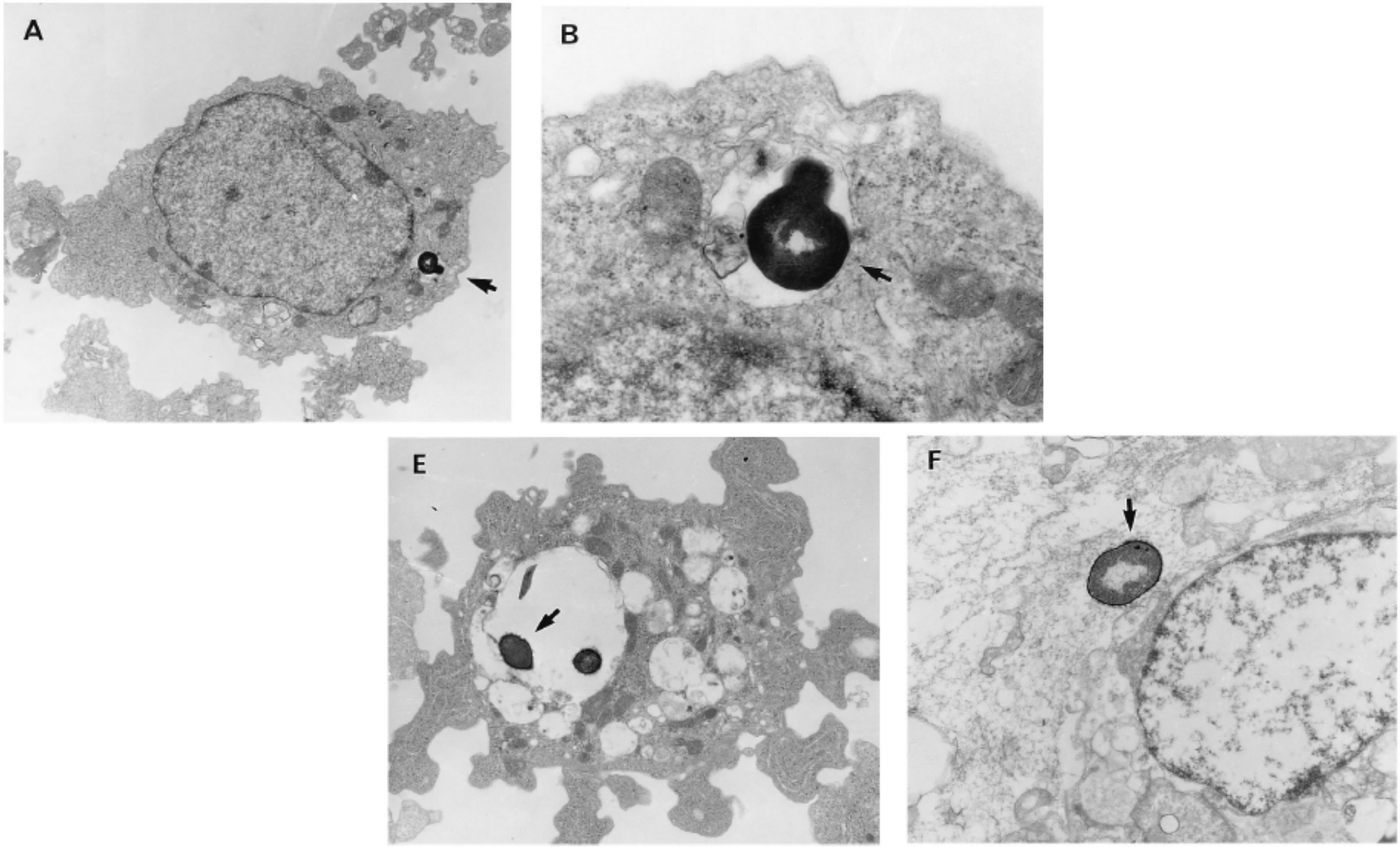
- Intracellular *S. pneumoniae*
- Why do we need a breakpoint ?
- The data …
- Interpretation …
- Our conclusions

Intracellular *S. pneumoniae*…

- *Streptococcus pneumoniae* have evolved strategies for colonization and resistance.
- Among them is the possibility to invade eucaryotic cells, such as monocytes and macrophages (1, 2), dendritic cells (3), or lung epithelial cells (4,5).
- This is also evidenced *in vivo* based on the examination of biopsies from children with chronic otitis (6)
- However, they are variations in internalization and intracellular survival according to capsular serotypes (7).

1. Song *et al.* Transcriptome studies on *Streptococcus pneumoniae*, illustration of early response genes to THP-1 human macrophages. Genomics. 2009 Jan;93(1):72-82. PMID: 18848982.
2. Nibbering *et al.* Deficient intracellular killing of bacteria by murine alveolar macrophages. Am J Respir Cell Mol Biol. 1989 Nov;1(5):417-22. PMID: 2534678.
3. Littmann *et al. Streptococcus pneumoniae* evades human dendritic cell surveillance by pneumolysin expression. EMBO Mol Med. 2009 Jul;1(4):211-22. PMID: 20049723
4. Mandell & Coleman. Activities of antimicrobial agents against intracellular pneumococci. Antimicrob Agents Chemother. 2000 Sep;44(9):2561-3. PubMed PMID: 10952618
5. Talbot *et al.* Uptake of *Streptococcus pneumoniae* by respiratory epithelial cells. Infect Immun. 1996 Sep;64(9):3772-7. PMID: 8751928
6. Coates *et al.* The role of chronic infection in children with otitis media with effusion: evidence for intracellular persistence of bacteria. Otolaryngol Head Neck Surg. 2008 Jun;138(6):778-81. PMID: 18503854.
7. Peppoloni *et al.* The encapsulated strain TIGR4 of *Streptococcus pneumoniae* is phagocytosed but is resistant to intracellular killing by mouse microglia. Microbes Infect. 2010 Nov;12(12-13):990-1001. PMID: 20615478.

Intracellular *S. pneumoniae*…

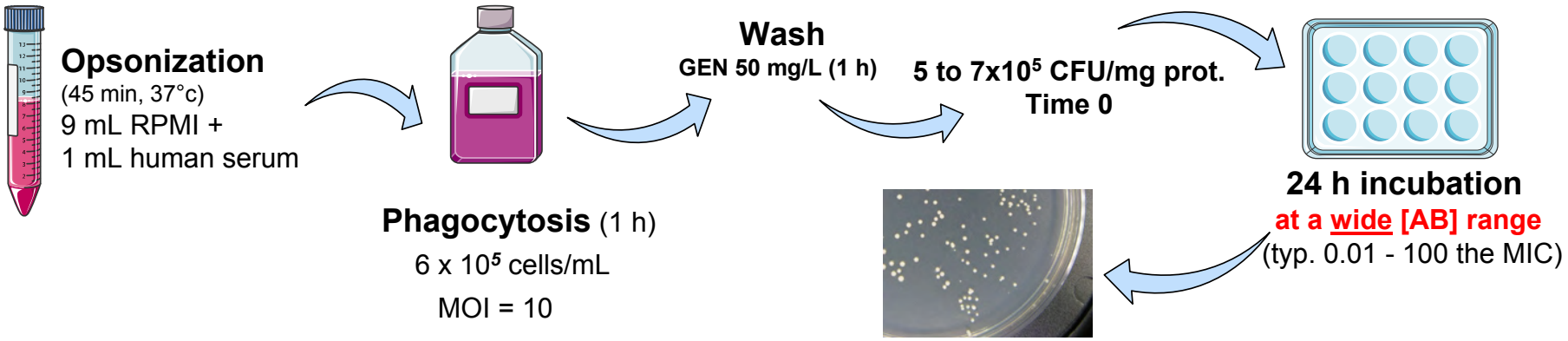


Talbot *et al.* Uptake of *Streptococcus pneumoniae* by respiratory epithelial cells. Infect Immun. 1996 Sep;64(9):3772-7. PMID: 8751928

Intracellular *S. pneumoniae*

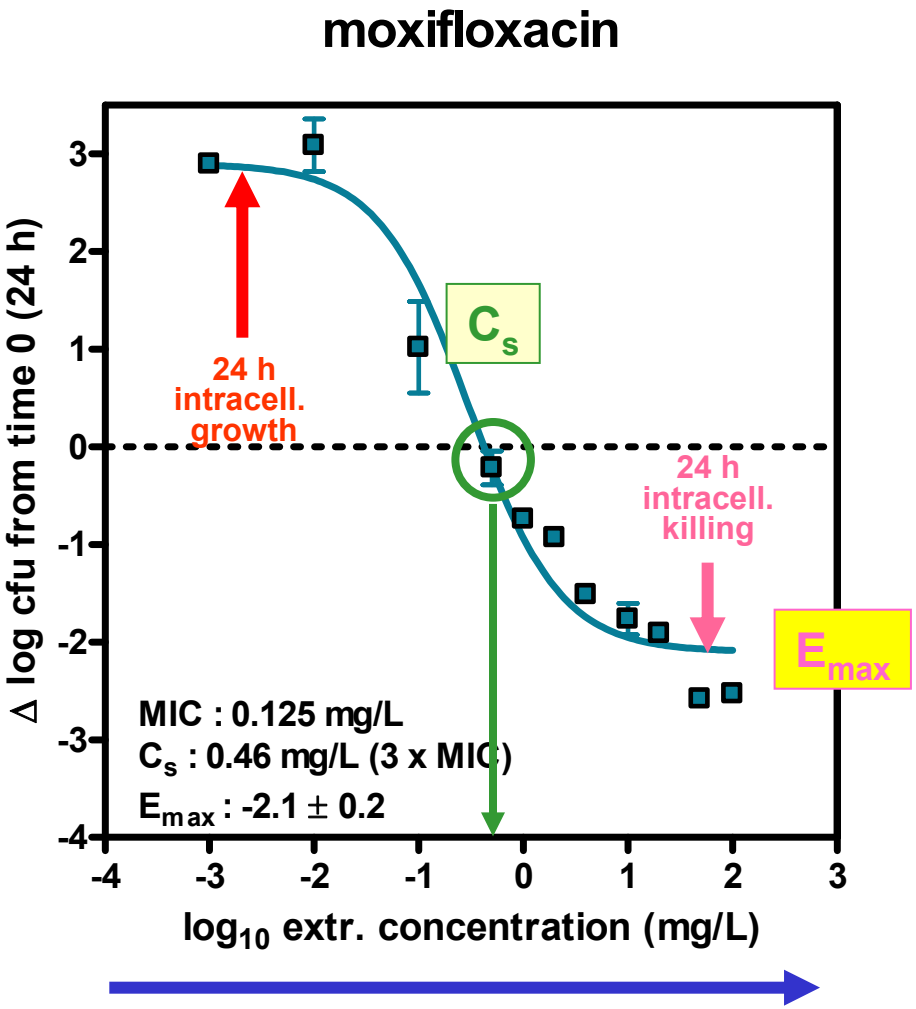
- Intracellular forms may play a critical role in chronic character of streptococcal infections (persistent foci) and in their subsequent dissemination (relapses and recurrence) because
  - ➢ they are sheltered from immune mechanisms of defense
  - ➢ they are poorly susceptible to most antibiotics (ECCMID 2011)

Pharmaodynamic model to study the intracellular activity of antibiotics



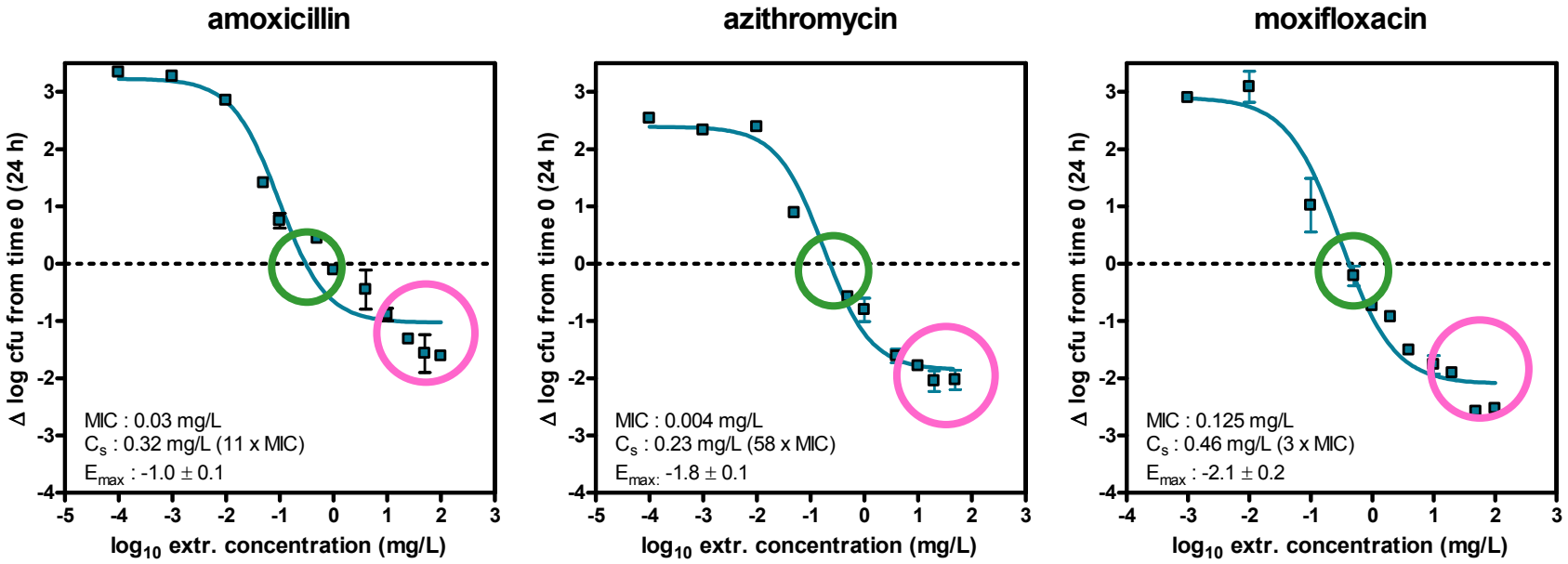
Lemaire et al. Activity of 9 antibiotics against intracellular forms of *S. pneumoniae,* ECCMID 2011: Poster P781

Pharmacodynamic intracellular *S. pneumoniae* model: typical results (1)



Lemaire et al. Activity of 9 antibiotics against intracellular forms of *S. pneumoniae*, ECCMID 2011: Poster P781

Pharmacodynamic intracellular *S. pneumoniae* model: typical results (2)



C_s (mg/L or x MIC

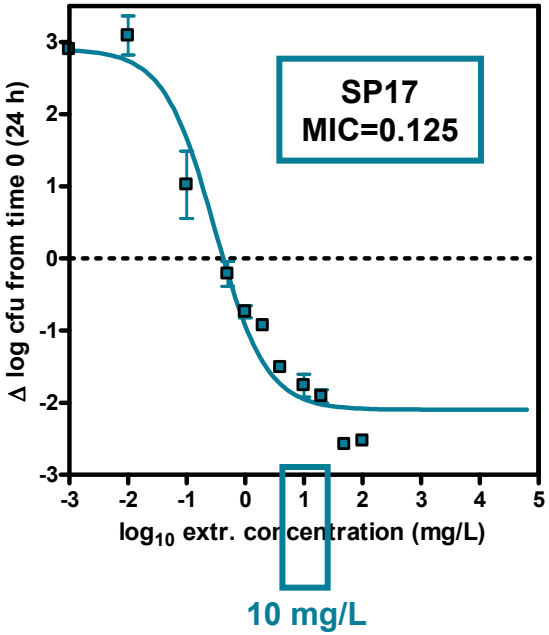
Emax (Δ log10 CFU/mg prot.)

Lemaire et al. Activity of 9 antibiotics against intracellular forms of *S. pneumoniae*, ECCMID 2011: Poster P781

Why do we need breakpoint ?

- A breakpoint is a **susceptibility value** (e.g., an MIC) which, applied to a **population of a micro-organisms of decreasing susceptibility** , will separate those that have a high likelihood of showing a useful response from those that will not.

simulation



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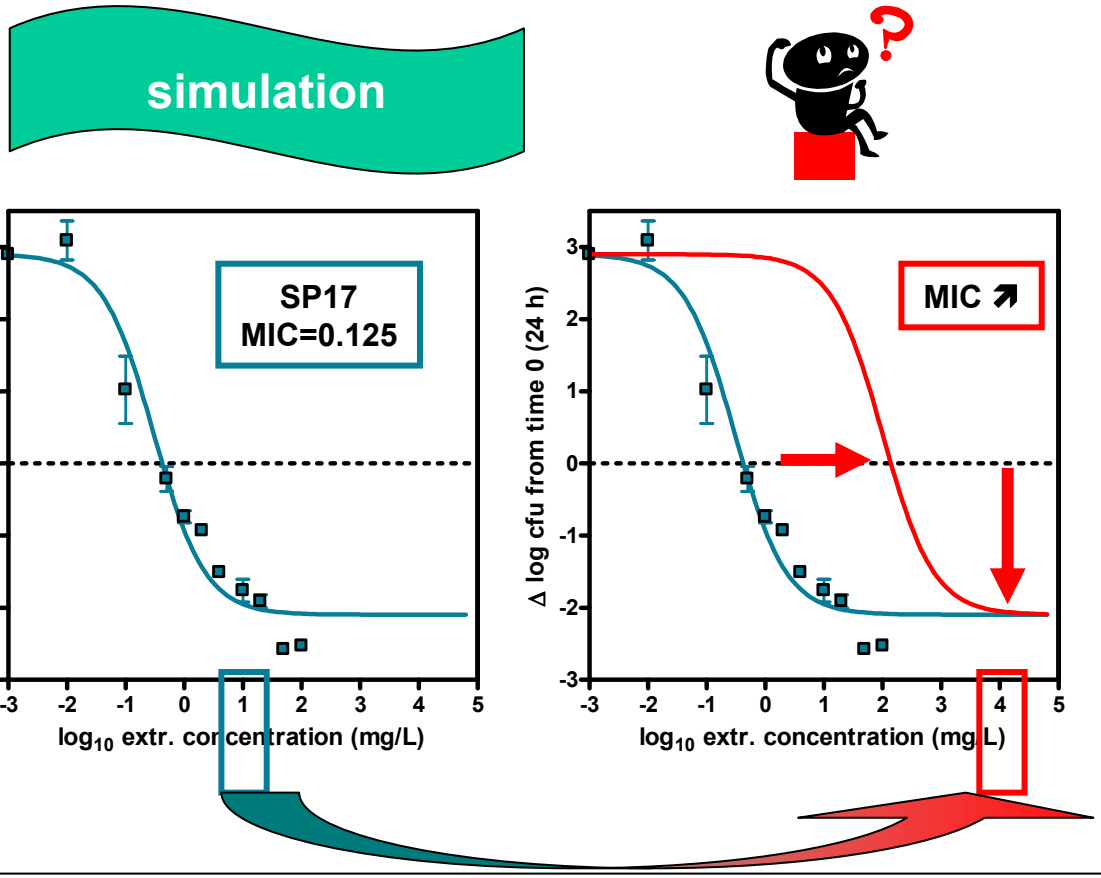

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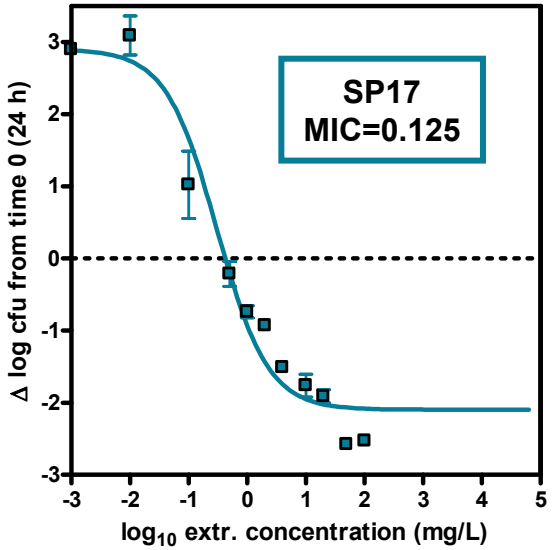
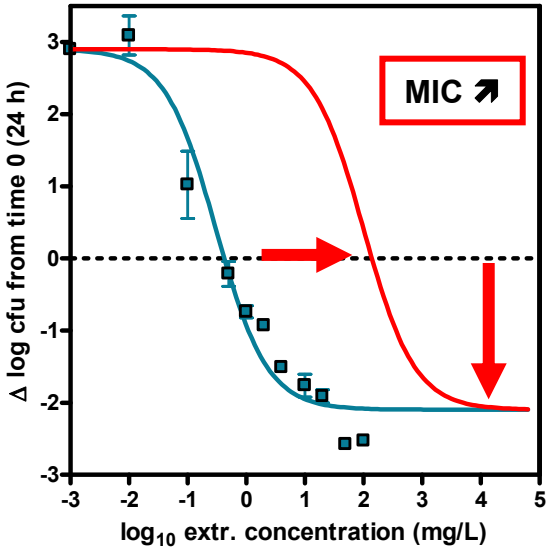
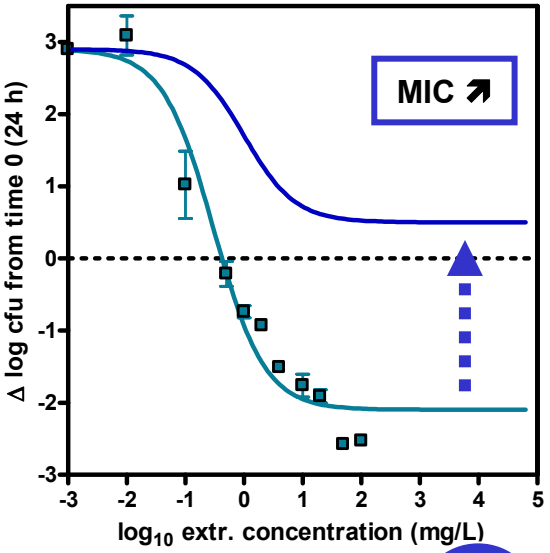
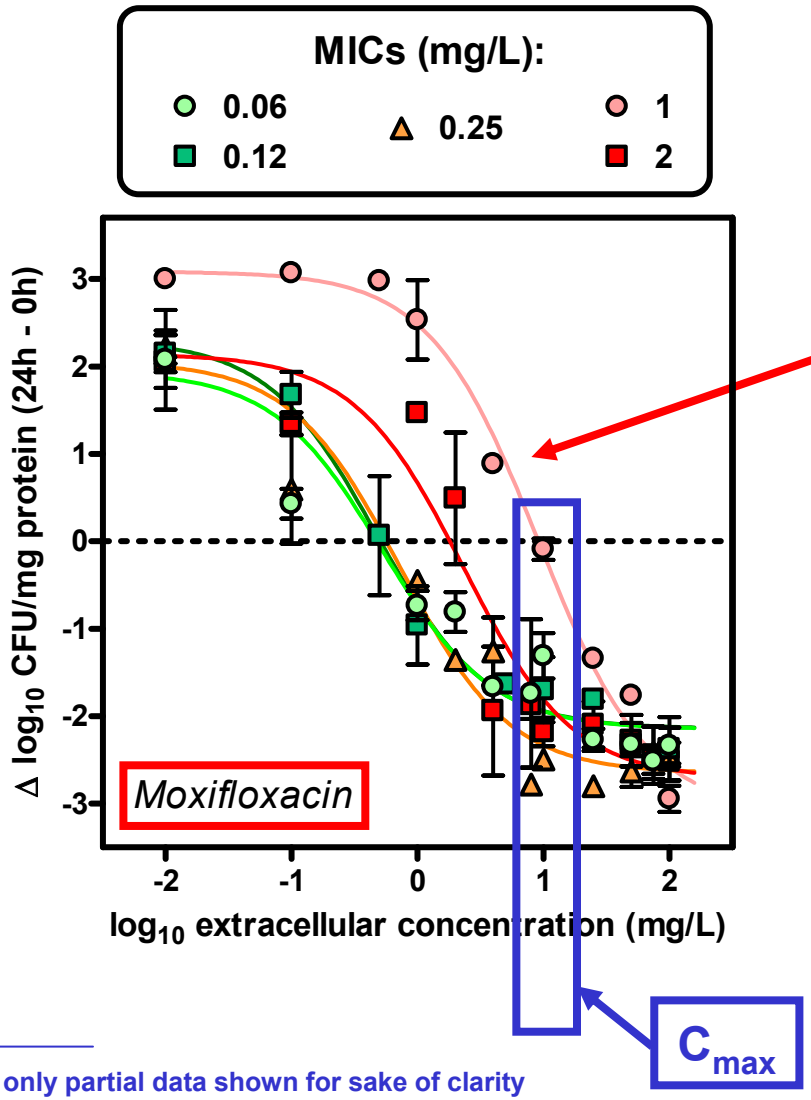




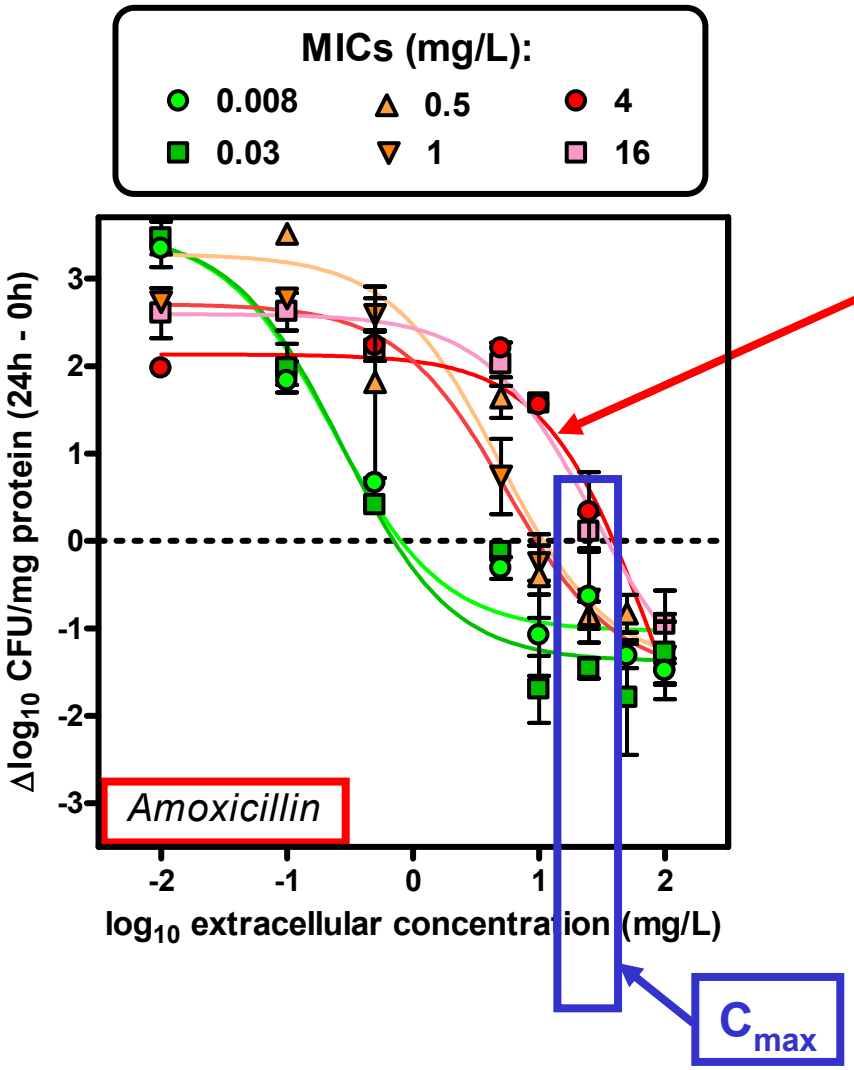


Actual data: moxifloxacin… *



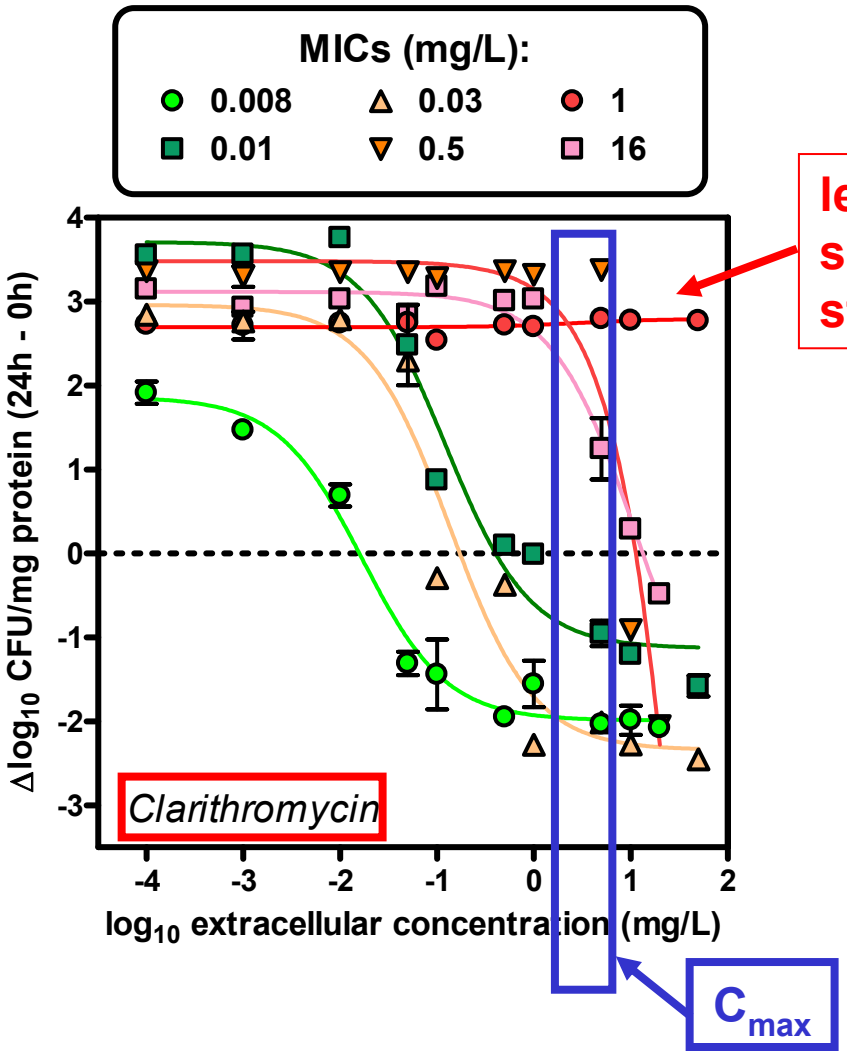
* only partial data shown for sake of clarity

Actual data: amoxicillin… *



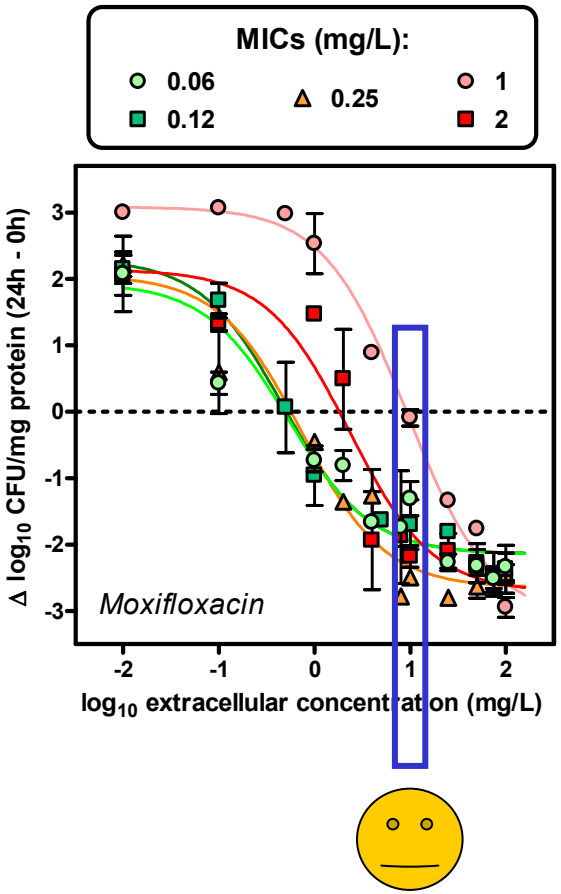
* only partial data shown for sake of clarity

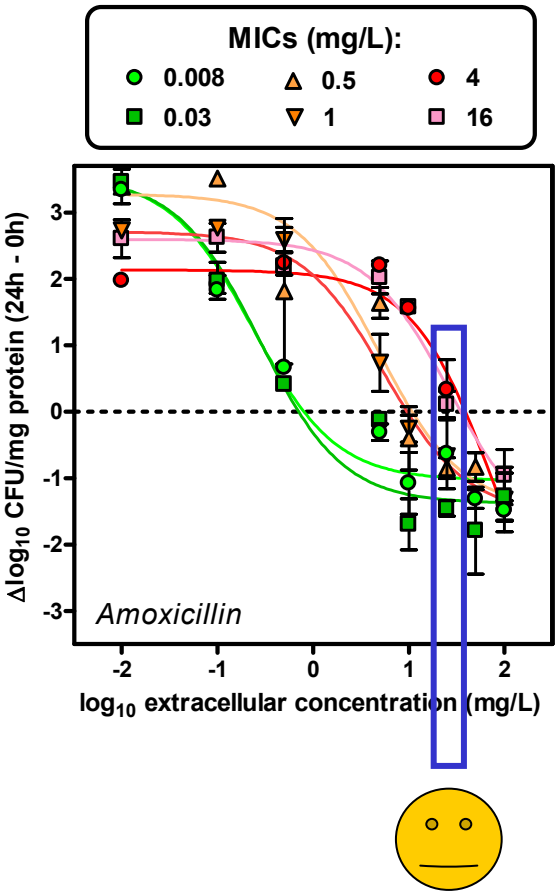
Actual data: clarithromycin… *

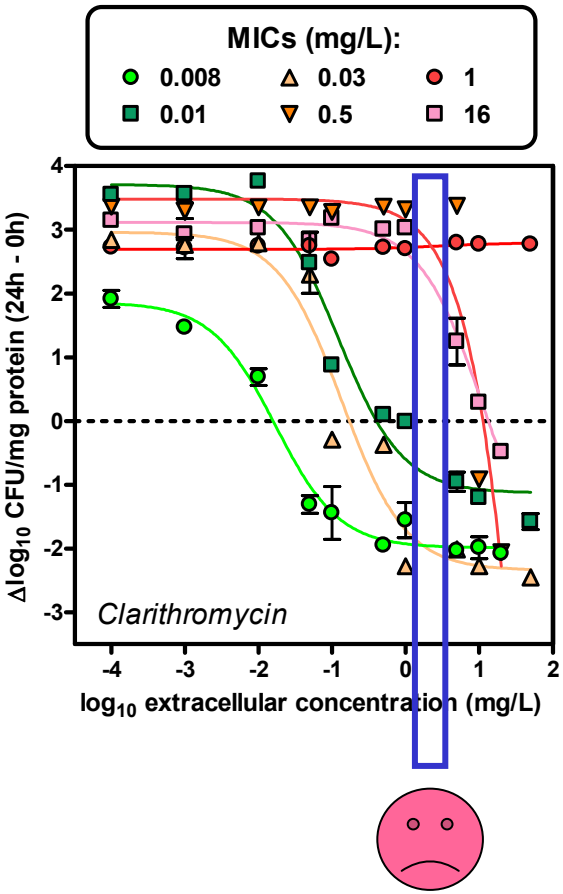


* only partial data shown for sake of clarity

Actual data: putting all together…







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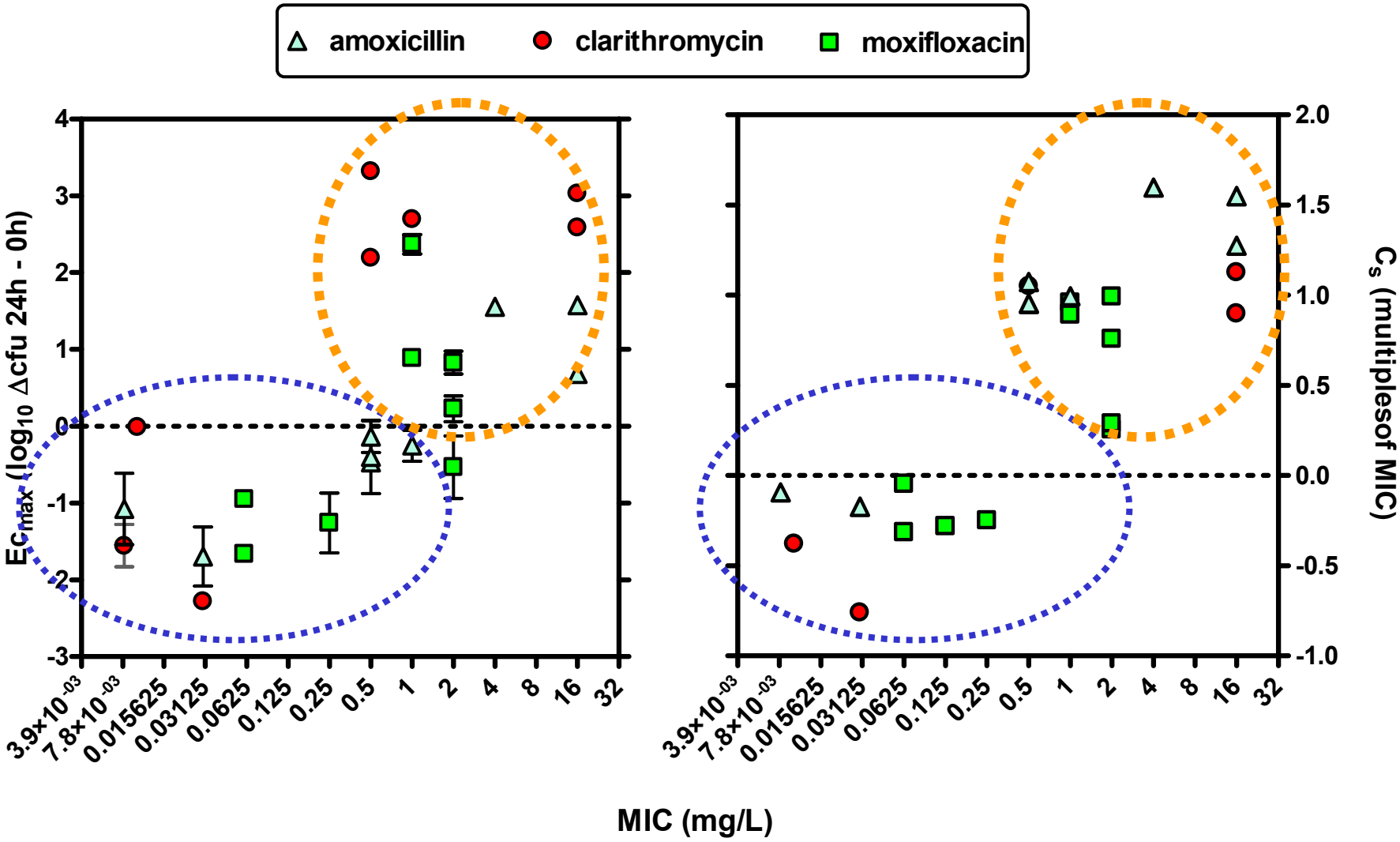
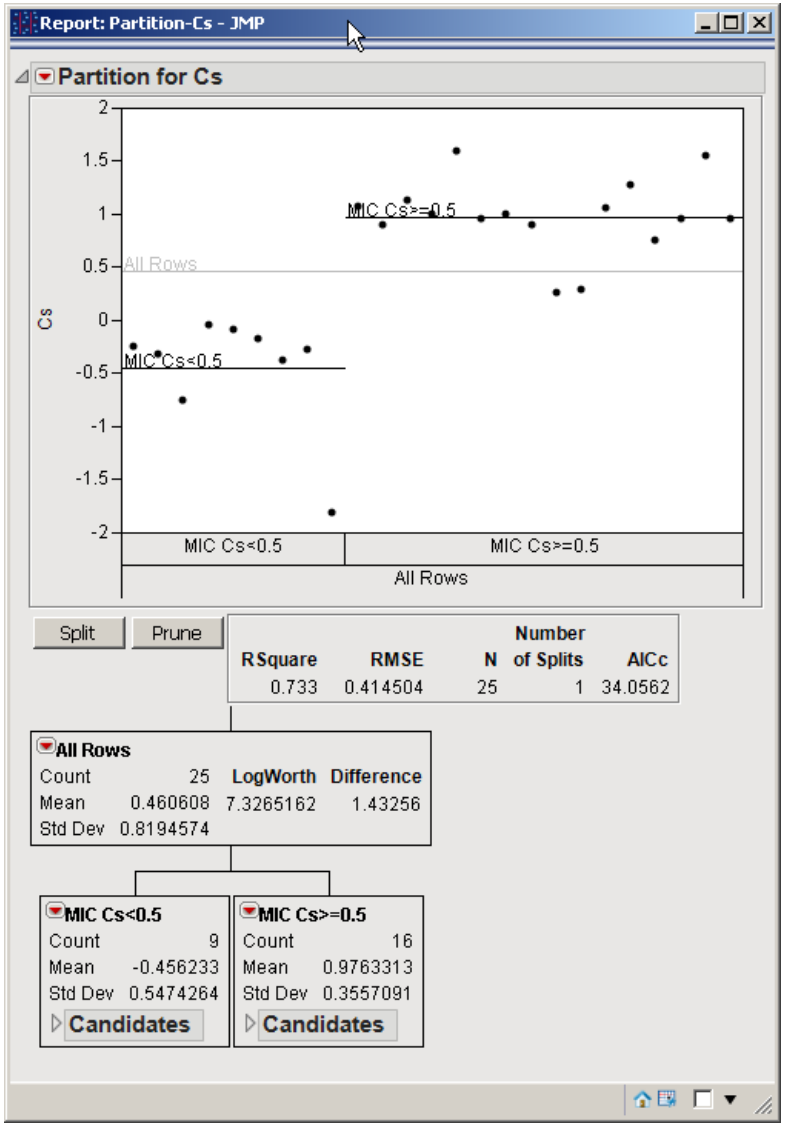
Analyzing all data globally



Towards a breakpoint…

Partition analysis

- turning a data table into a hierarchy of questions
- The factor columns (X's) can be continuous.
- Then the platform fits means.
- The split is chosen **to maximize the difference in the responses between the two branches of the split.**



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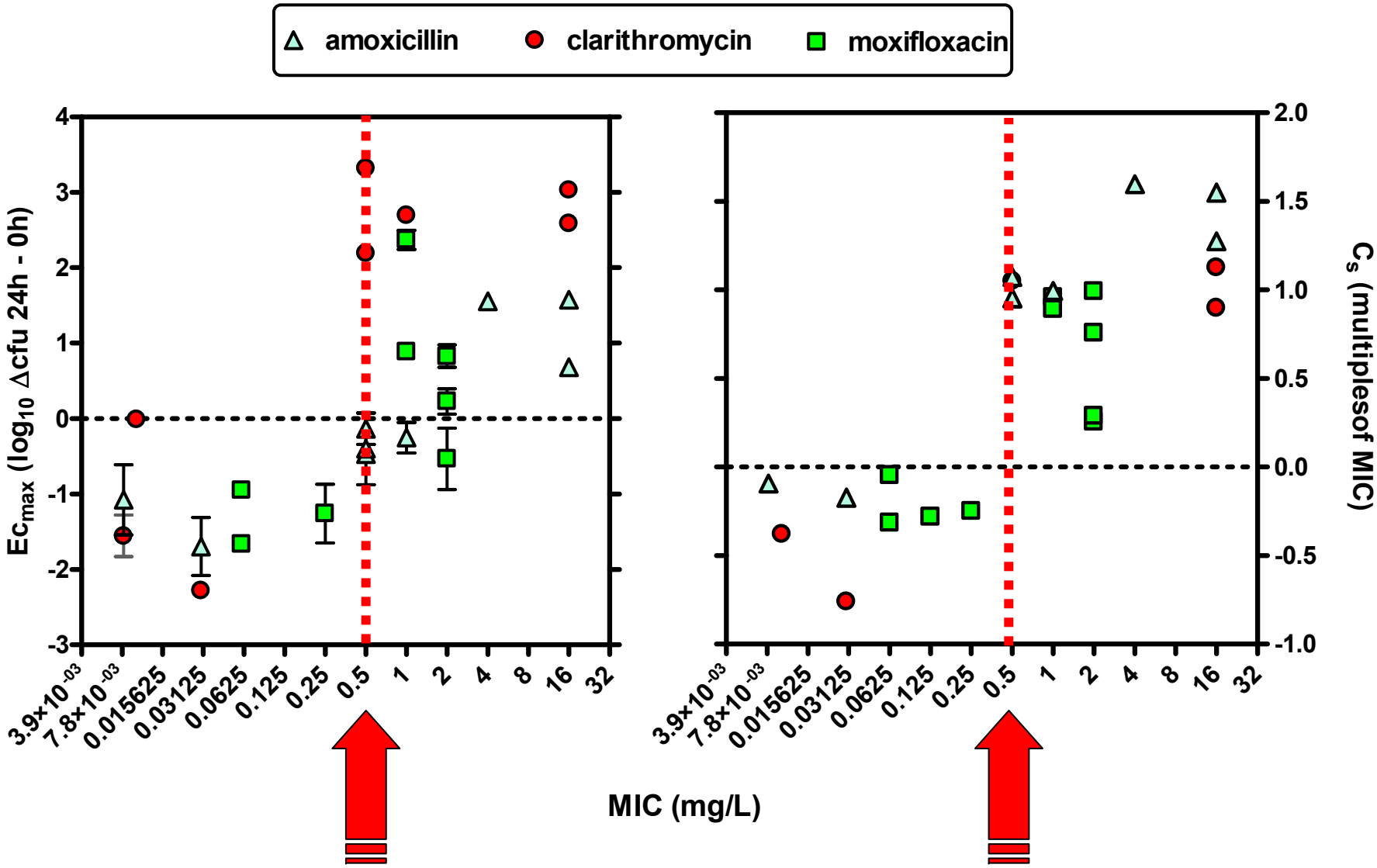
"Recursive partitioning" breakpoints



Main messages and Food for thought…

- It is possible to **measure** the intracellular activity of antibiotics against *S. pneumoniae* using pharmacological tools (pharmacodynamics)
- There is a gradual shift of the concentration-dependent responses of phagocytized isolates as a function their MICs, with E_{max} moving from negative to positive values and C_s increasing
- For both parameters, recursive partitioning indicated a dichotomous split at an MIC of 0.5 mg/L
- This "intracellular breakpoint" is close to the clinical "S" susceptibility breakpoints of EUCAST for these antibiotics…
  (clarithromycin = 0.25 – amoxicillin and moxifloxacin = 0.5)

A word of caution, however…

A breakpoint should not be considered as wall …

… but as a help to the clinician

… who may decide to knowingly overpass it !

Thank you for any suggestion !