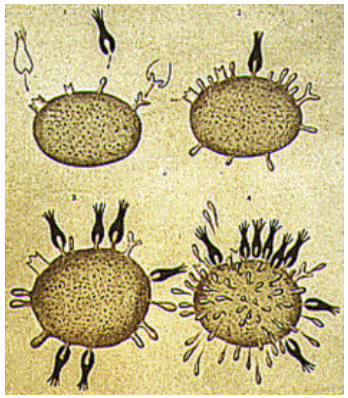
“corpora non agunt nisi fixata”



Ehrlich’s “magic bullet” theory

Antibiotic accumulation and efflux in eukaryotic cells:

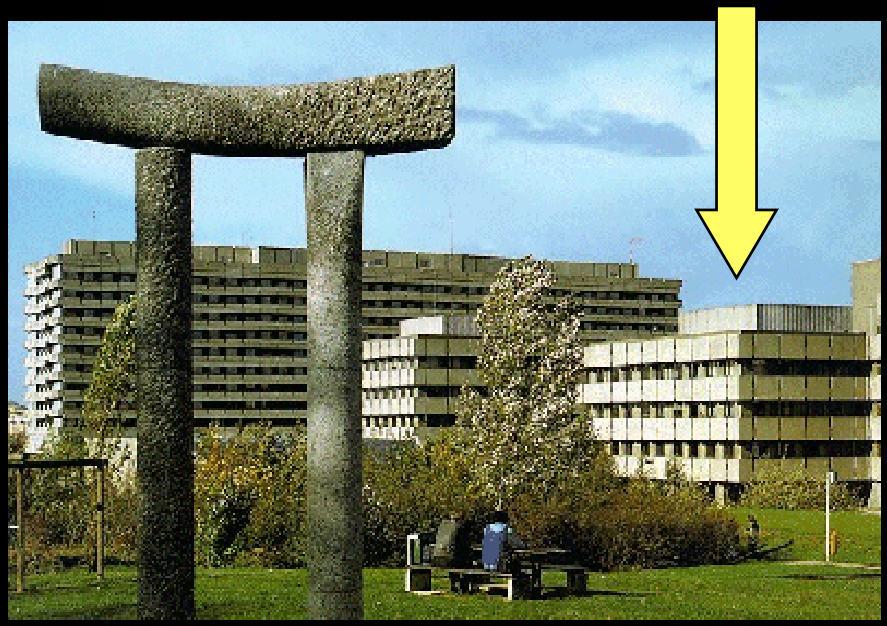
a journey at the frontier of pharmacokinetics and pharmacodynamics

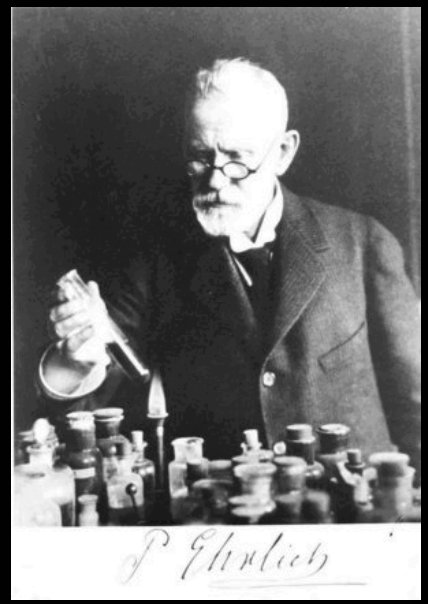
Unité de Pharmacologie
cellulaire et moléculaire

F. Van Bambeke

Magic bullets need to reach their target

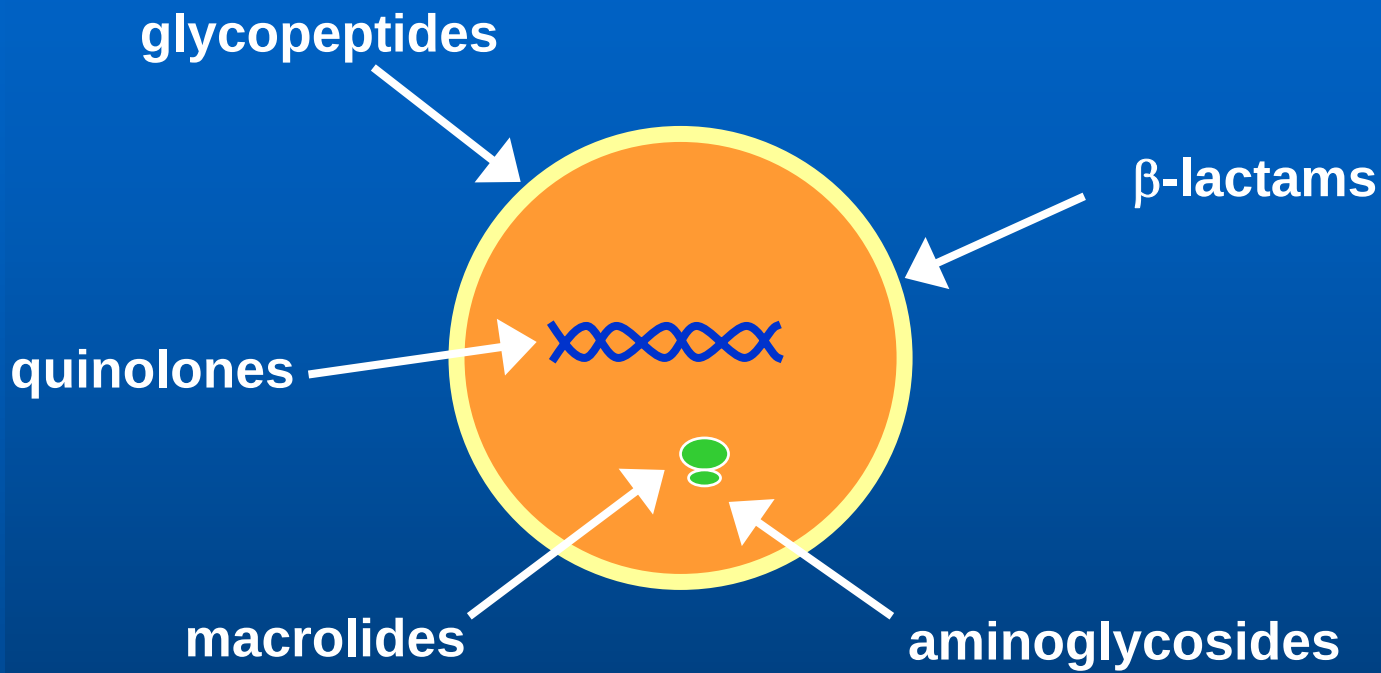
Paul Ehrlich (1854–1915)





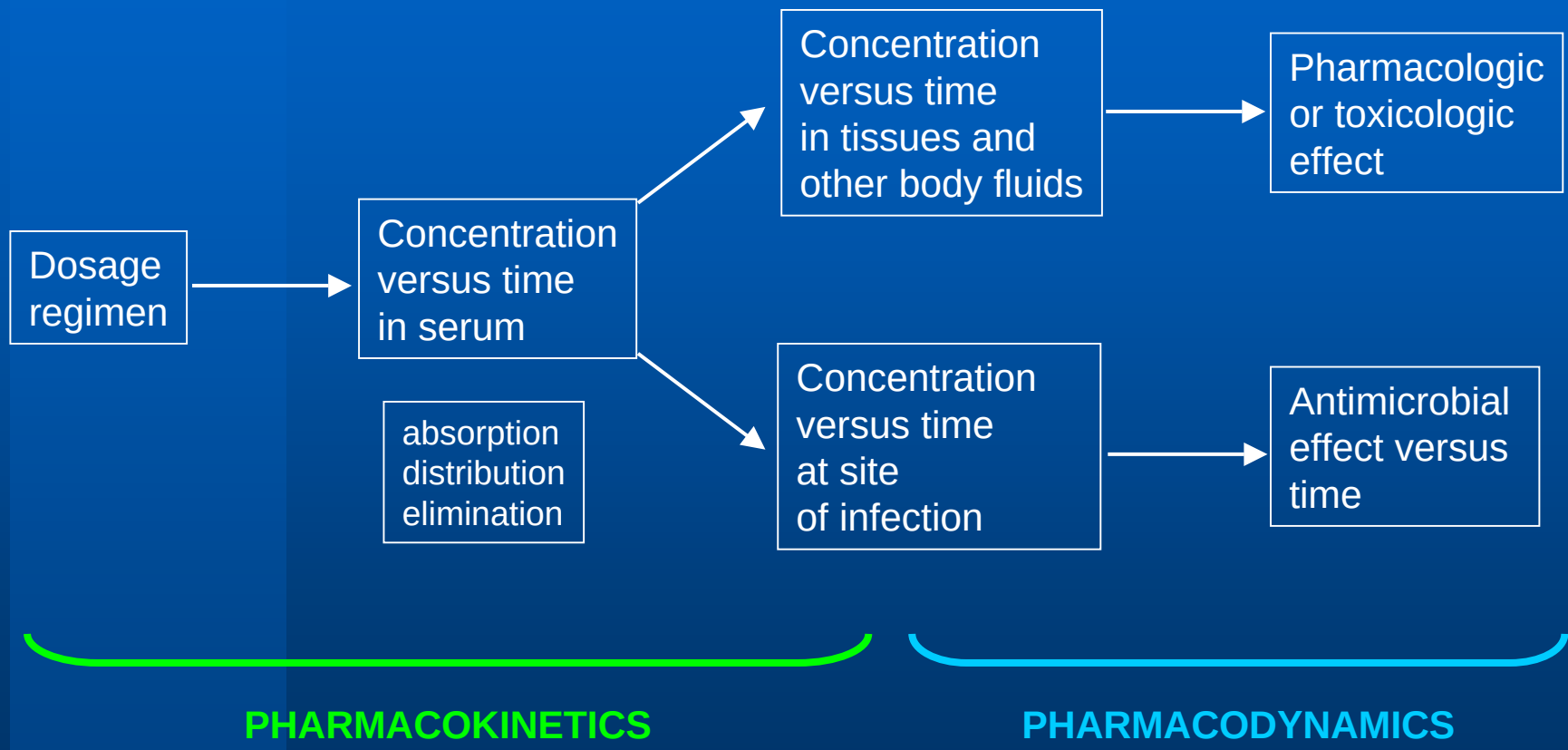
"…the goal is…to find chemical substances that have special affinities for pathogenic organisms and that, like *magic bullets*, go straight to their targets..."

Magic bullets need to reach their target



for appropriate time
and in sufficient concentration …

Birth of antibiotic “PK-PD”



Craig (1998) CID 26:1-10

ISAP classical view of PK/PD

PPT Slide - Netscape

File Edit View Go Bookmarks Tools Window Help

http://www.isap.org/2002/Workshop-San-Diego/dias-part-2/Craig/sld011.htm

Home Radio Netscape Search Bookmarks

PK/PD Parameters Correlating with Efficacy in Murine Thigh and Lung Infections

Time Above MIC	AUC (Peak)
Penicillins	Aminoglycosides
Cephalosporins	Fluoroquinolones
Carbapenems	Metronidazole
Monobactams	Daptomycin
Tribactams	Ketolides
	Azithromycin
	Streptogramins
	Glycopeptides
	Tetracyclines
	Macrolides

Slide 11 of 31

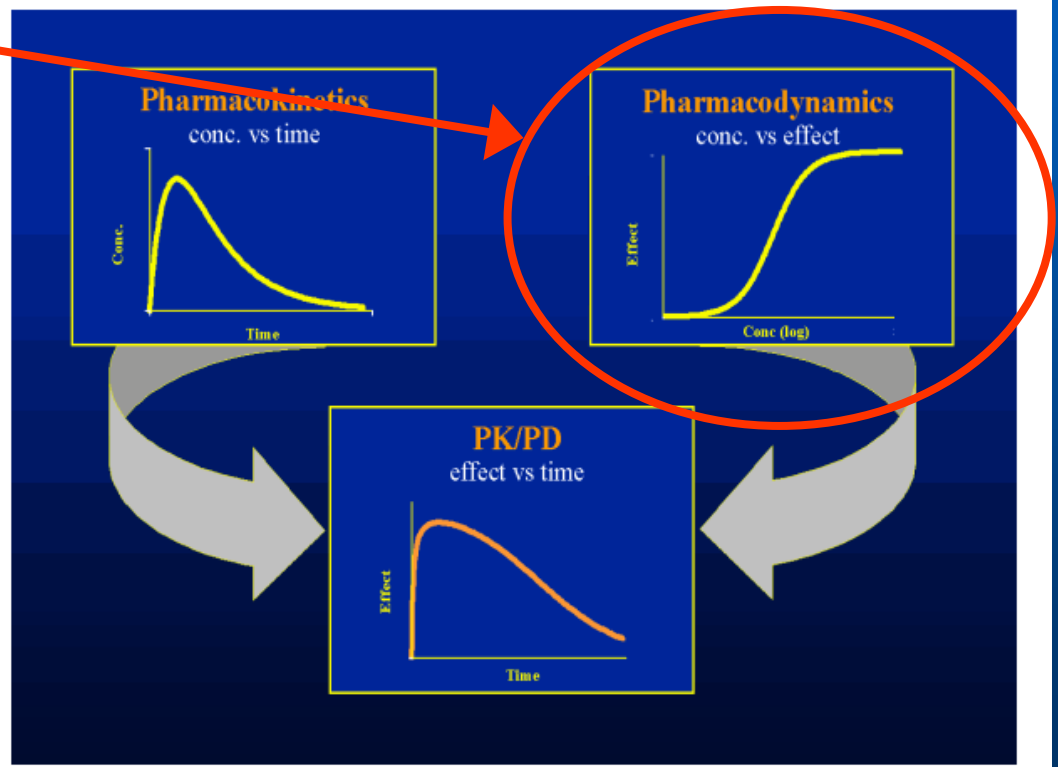
but classical PD predicts concentration-effects for all drugs ….

PPT Slide - Netscape

File Edit View Go Bookmarks Tools Window Help

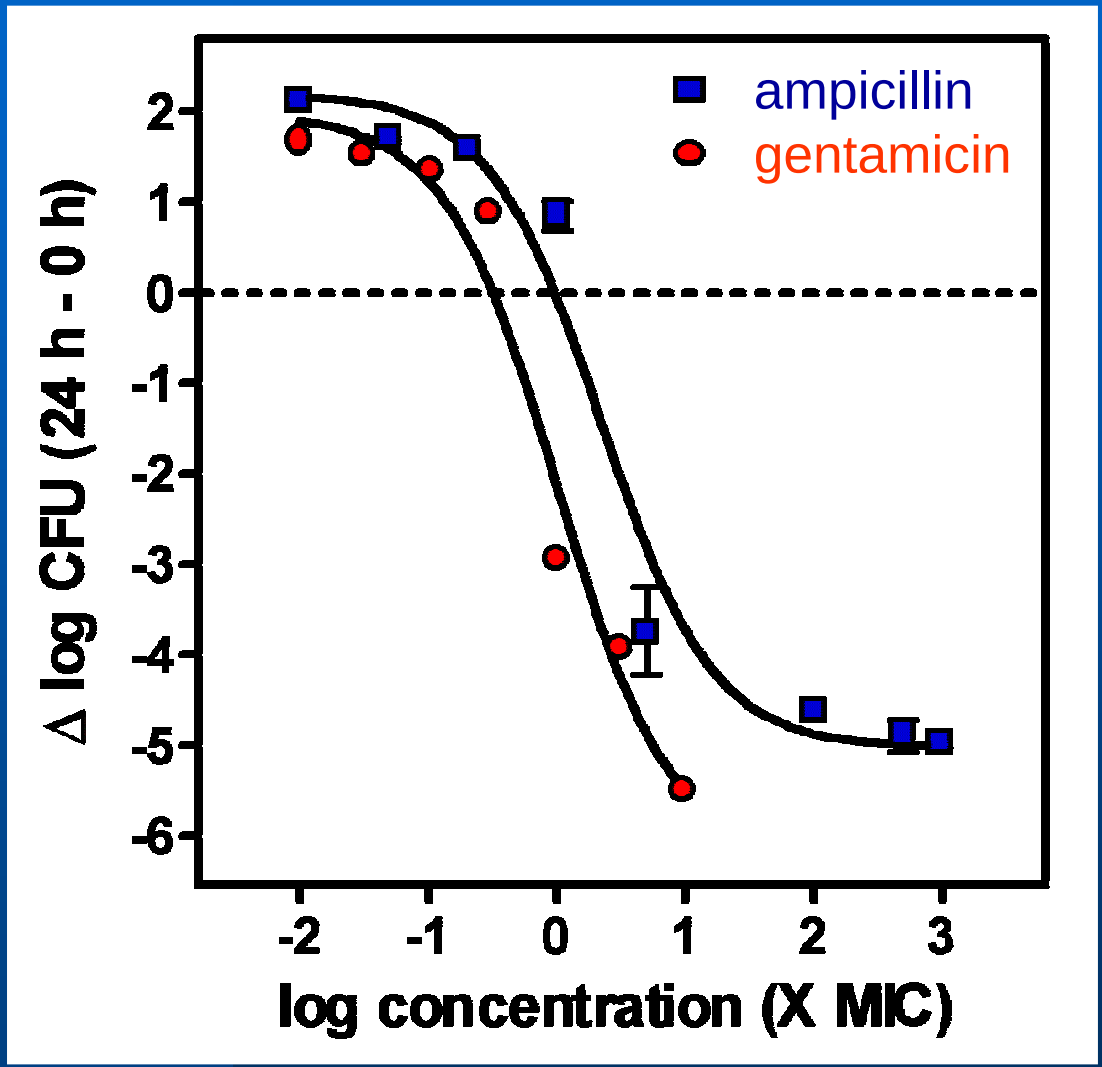
http://www.isap.org/2003/Workshop-Glasgow/Derendorf-1/sld003.htm

Home Radio Netscape Search Bookmarks



Slide 3 of 55

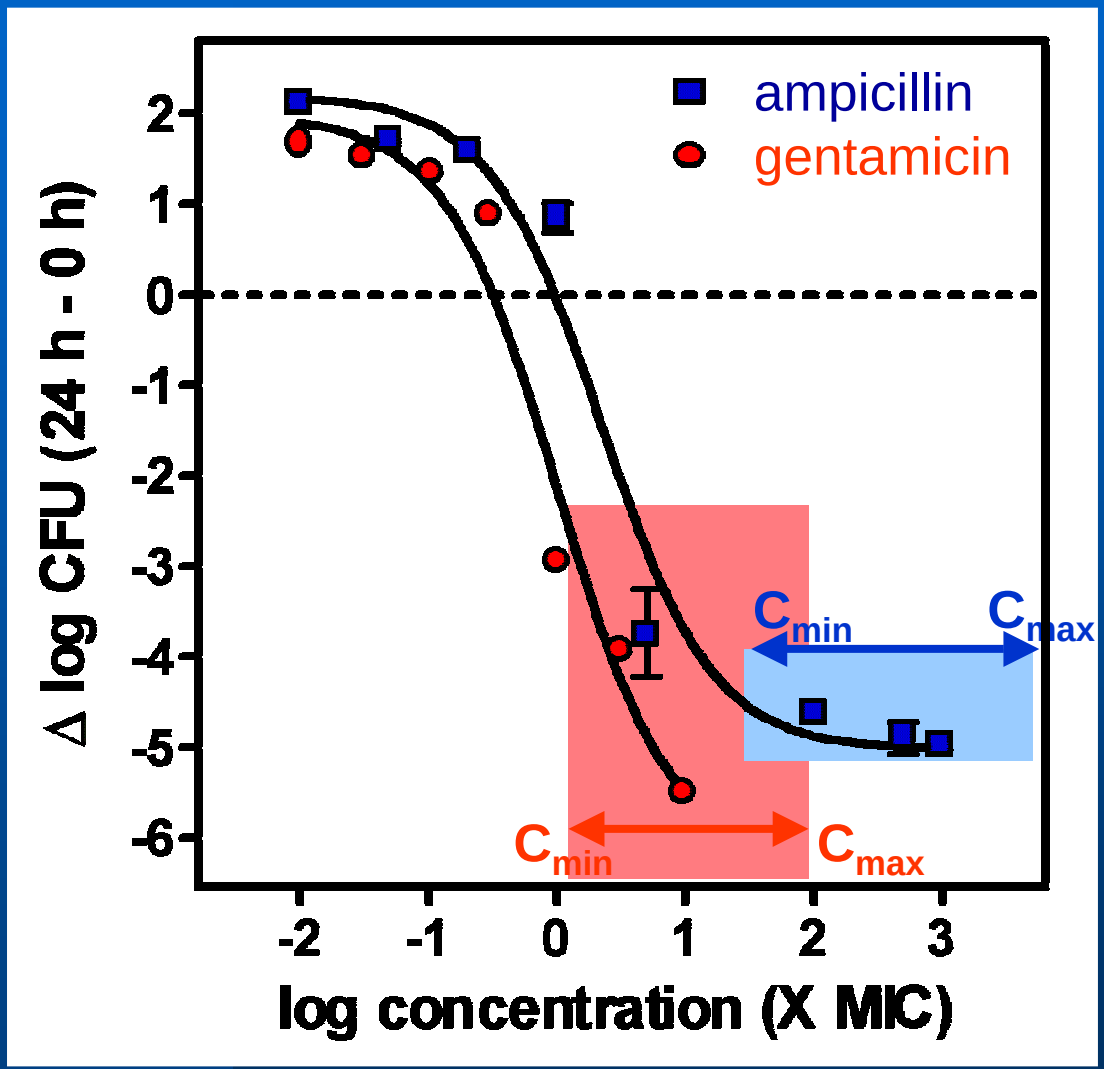
but classical PD predicts concentration-effects for all drugs ….



S. aureus; 24 h

Barcia-Macay *et al,* submitted; Lemaire *et al* (2005) JAC in press

Can we conciliate both theories ?



time -dependent

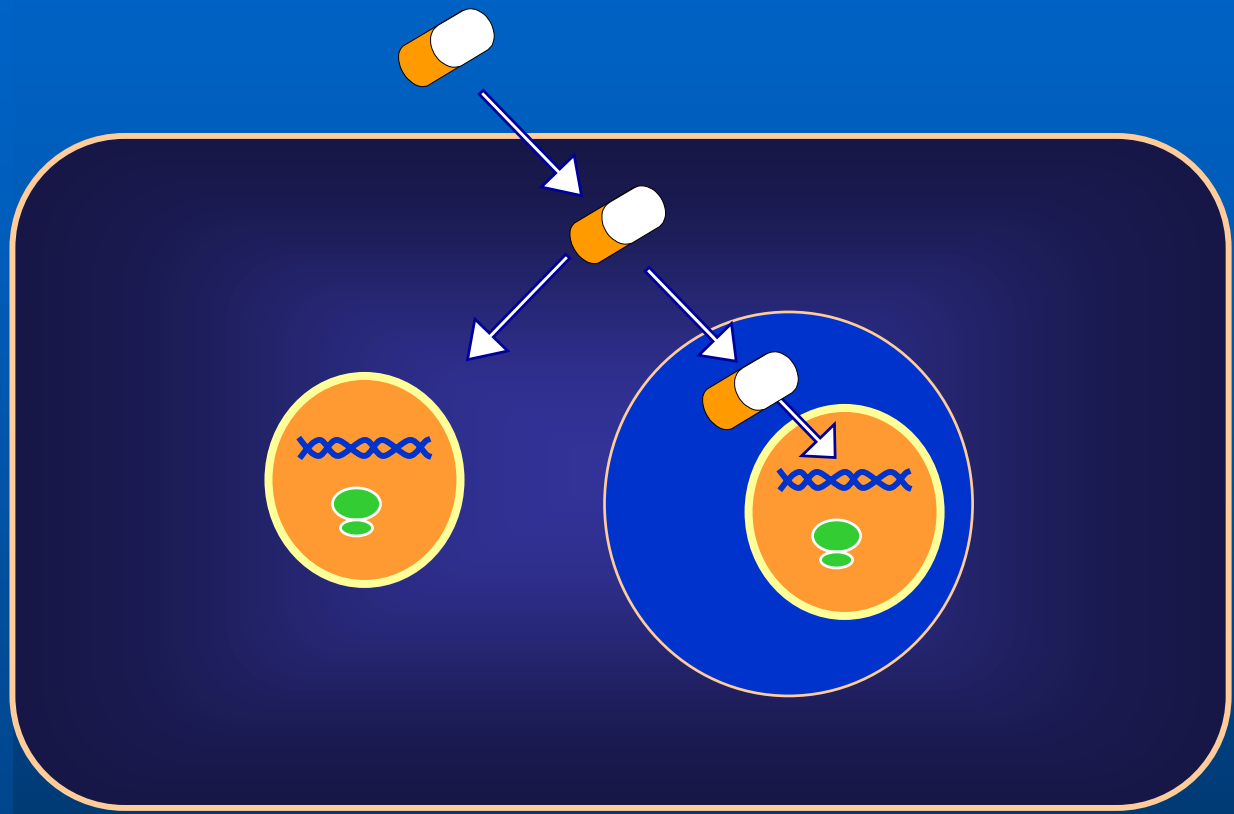
conc.-dependent

PD profile
in the clinics

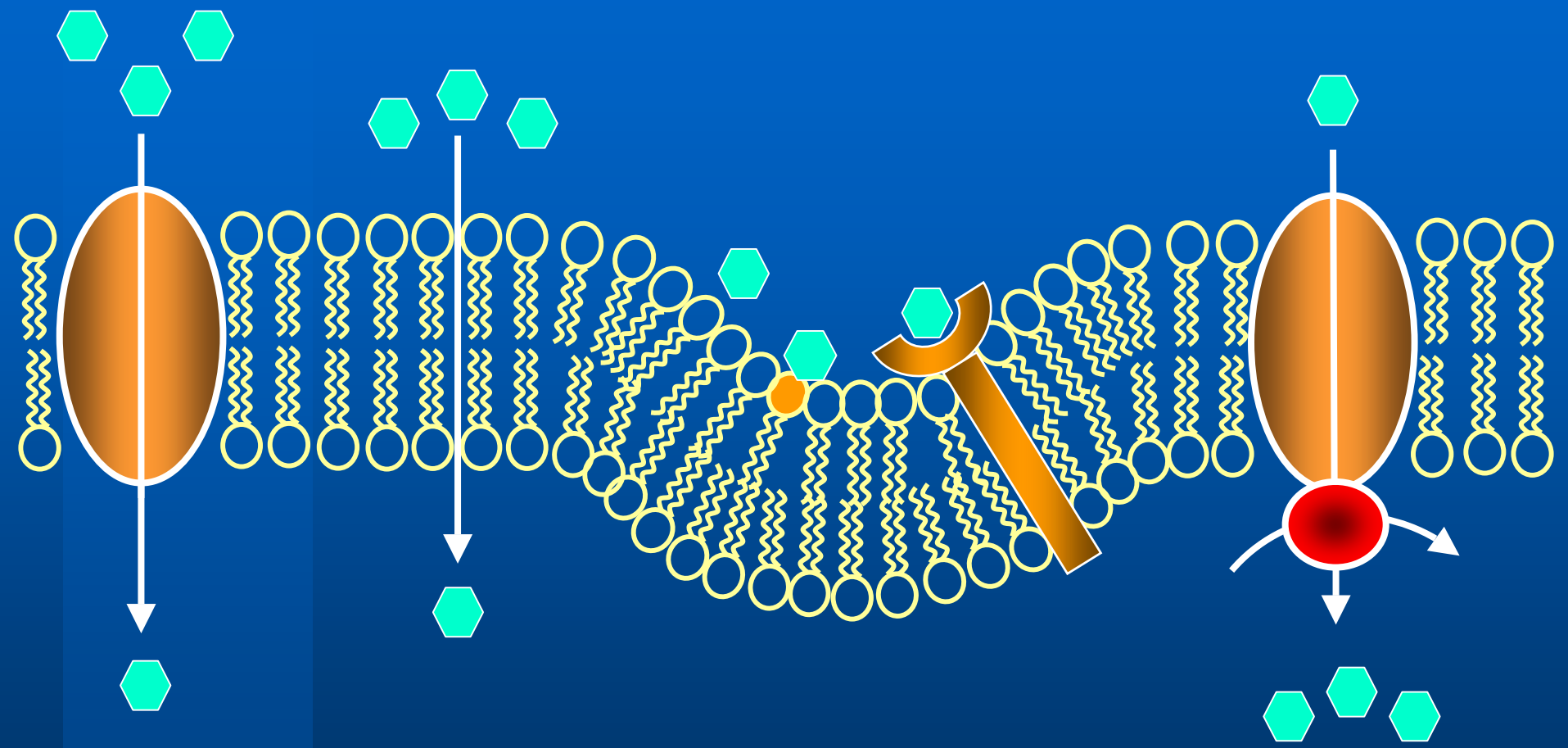
S. aureus; 24 h

Barcia-Macay *et al*, submitted; Lemaire *et al* (2005) JAC in press

Target accessibility becomes critical for intracellular activity



Main routes of drug entry in cells



channel

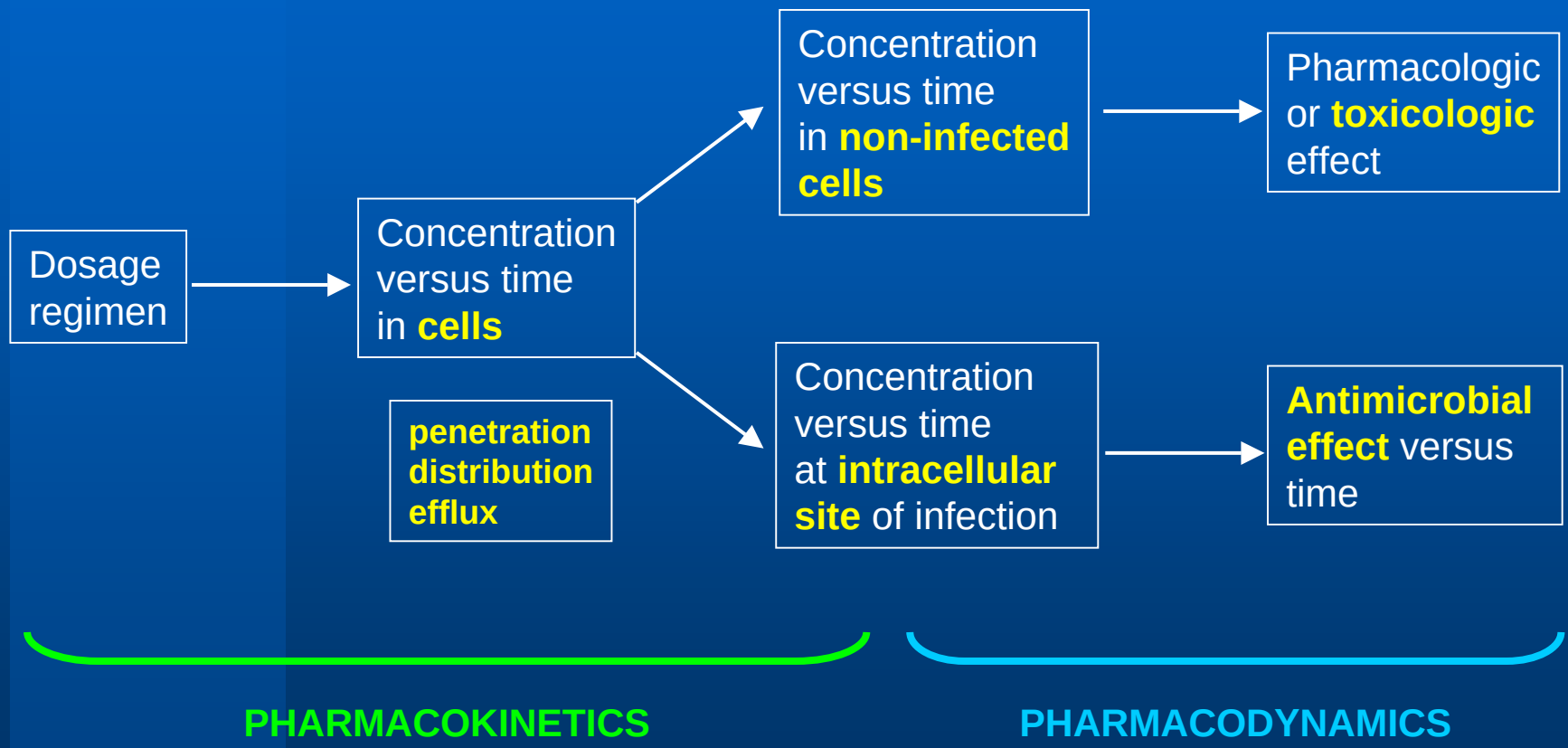
transporter diffusion

endocytosis transporter

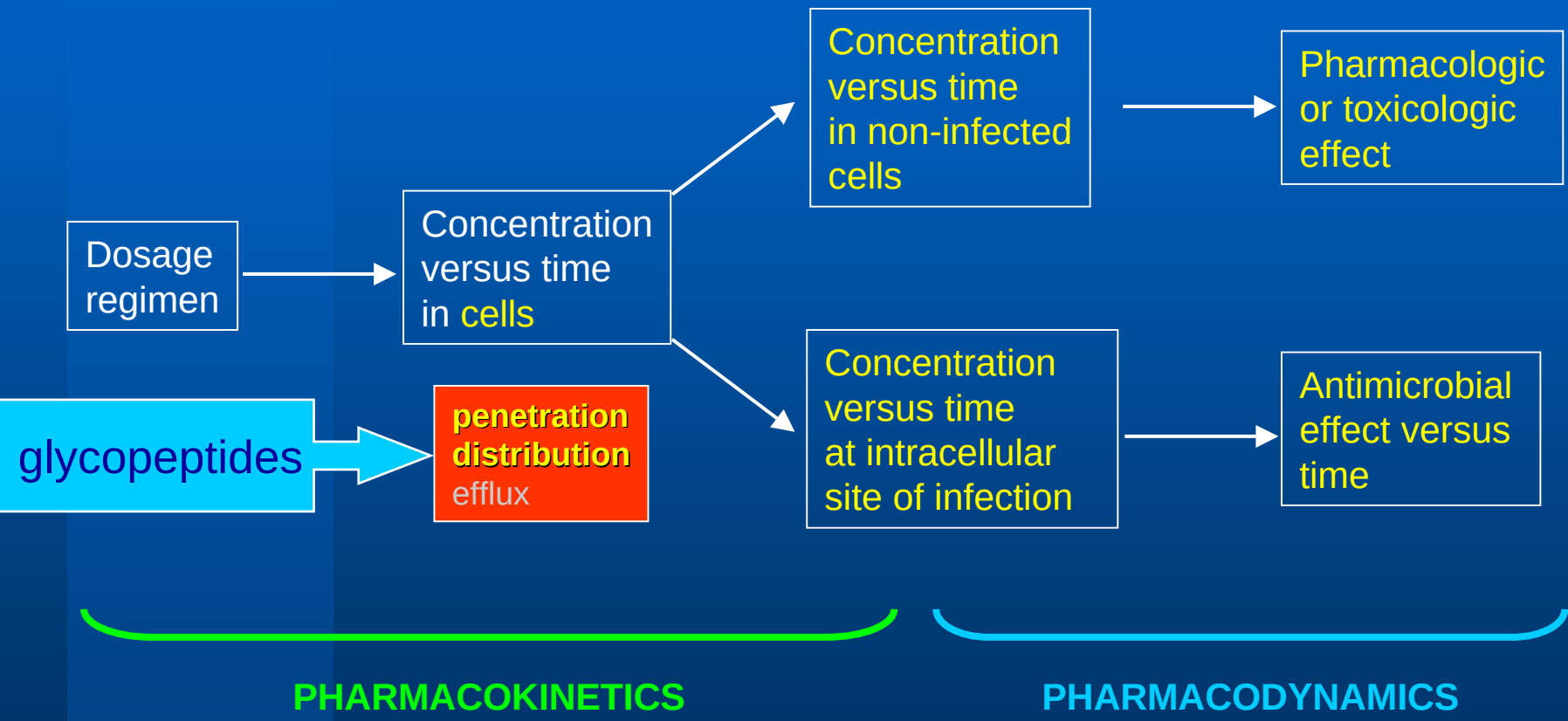
passive

active

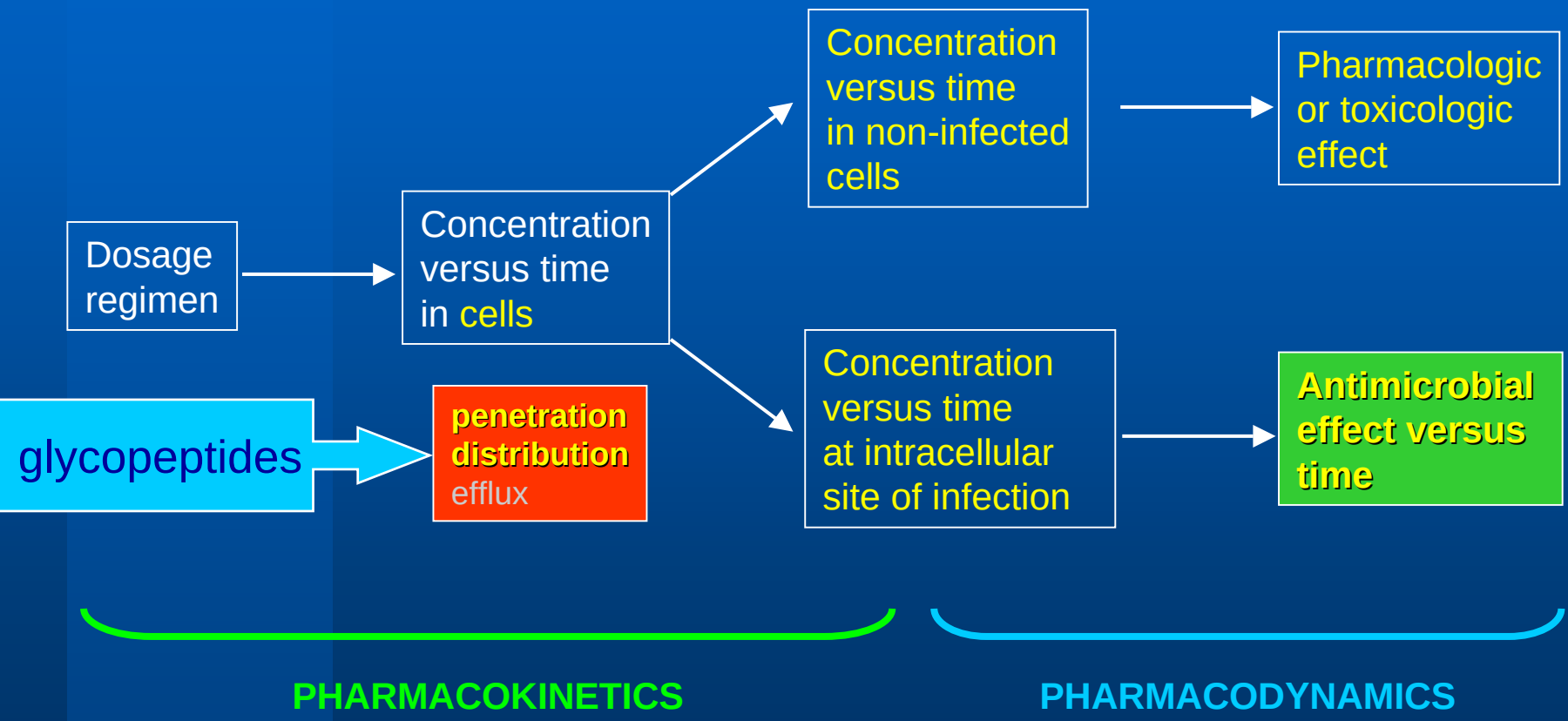
Intracellular “PK-PD”



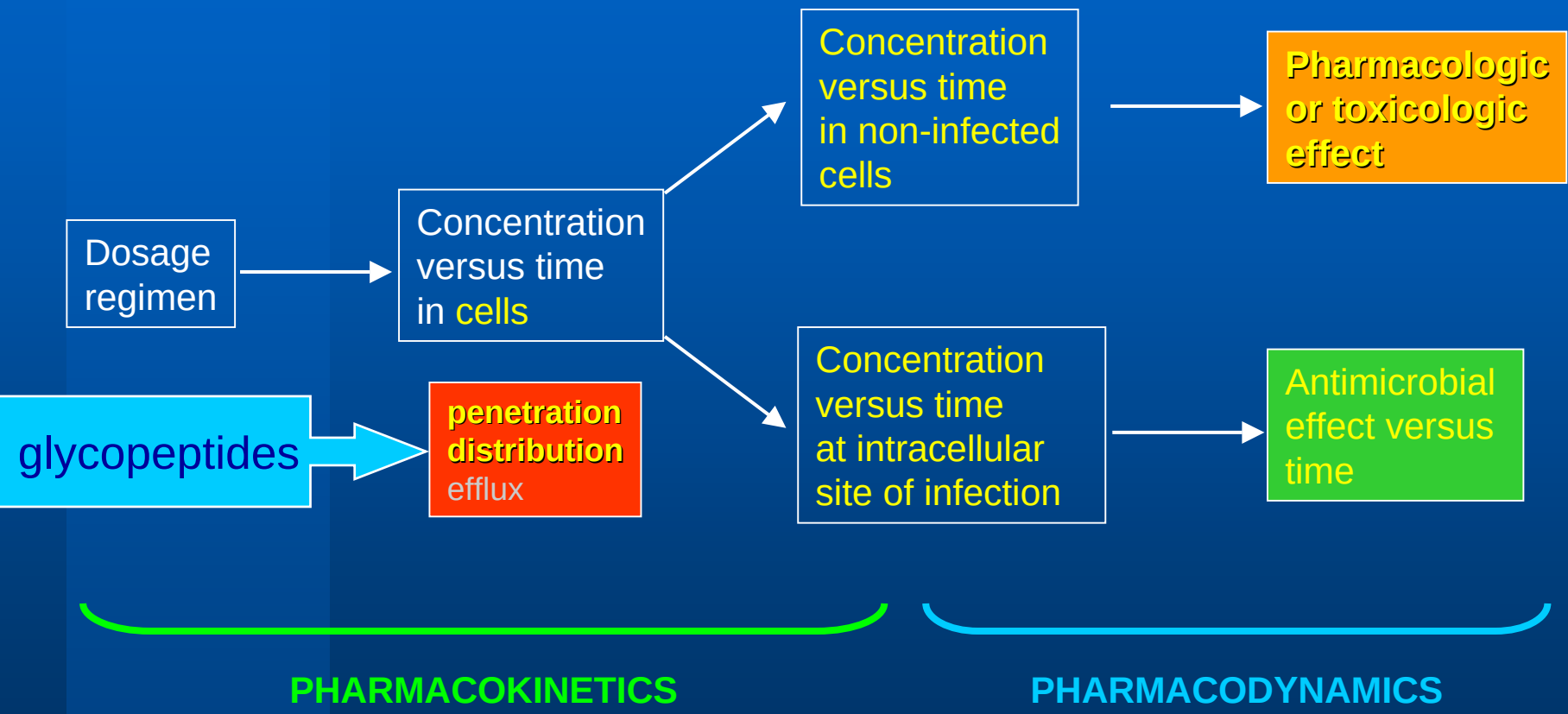
Intracellular “PK-PD”



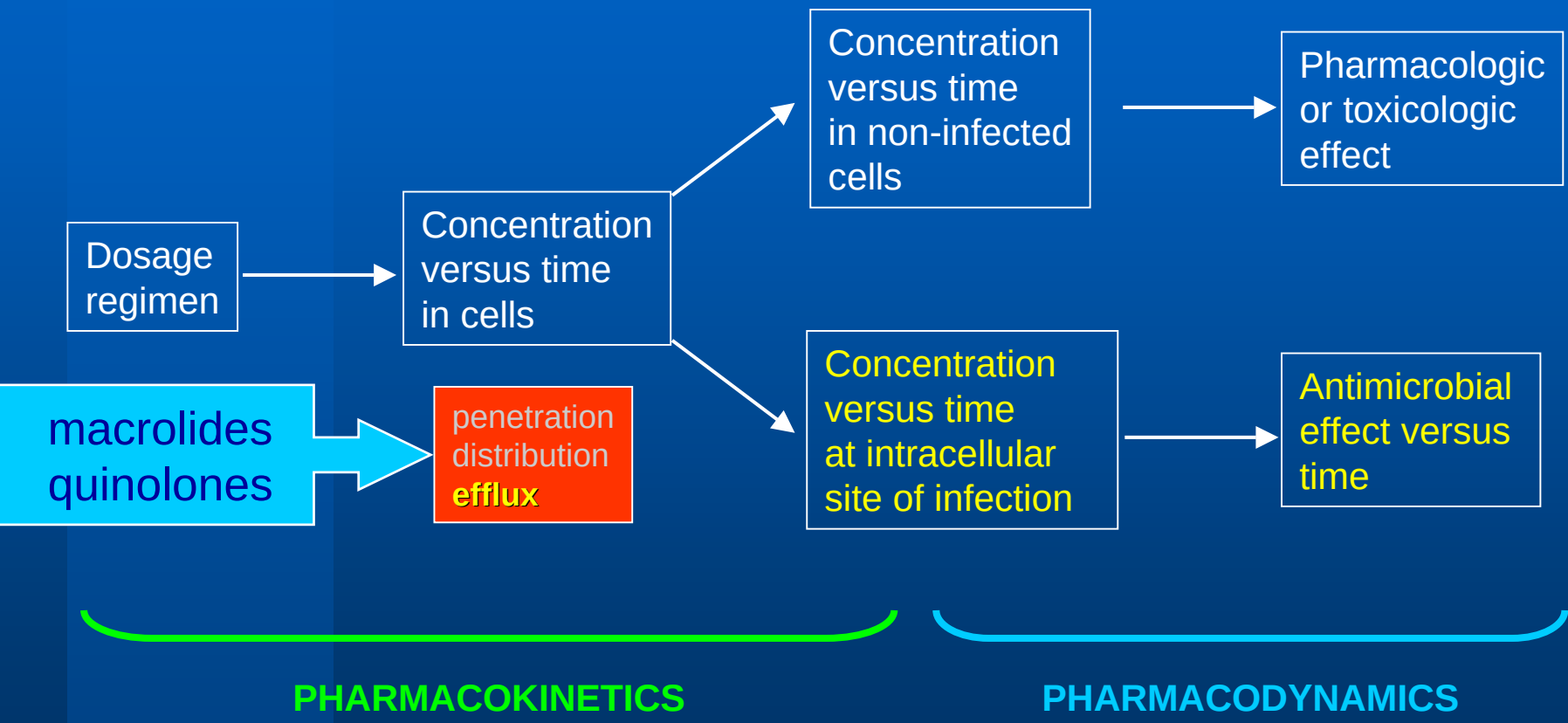
Intracellular “PK-PD”



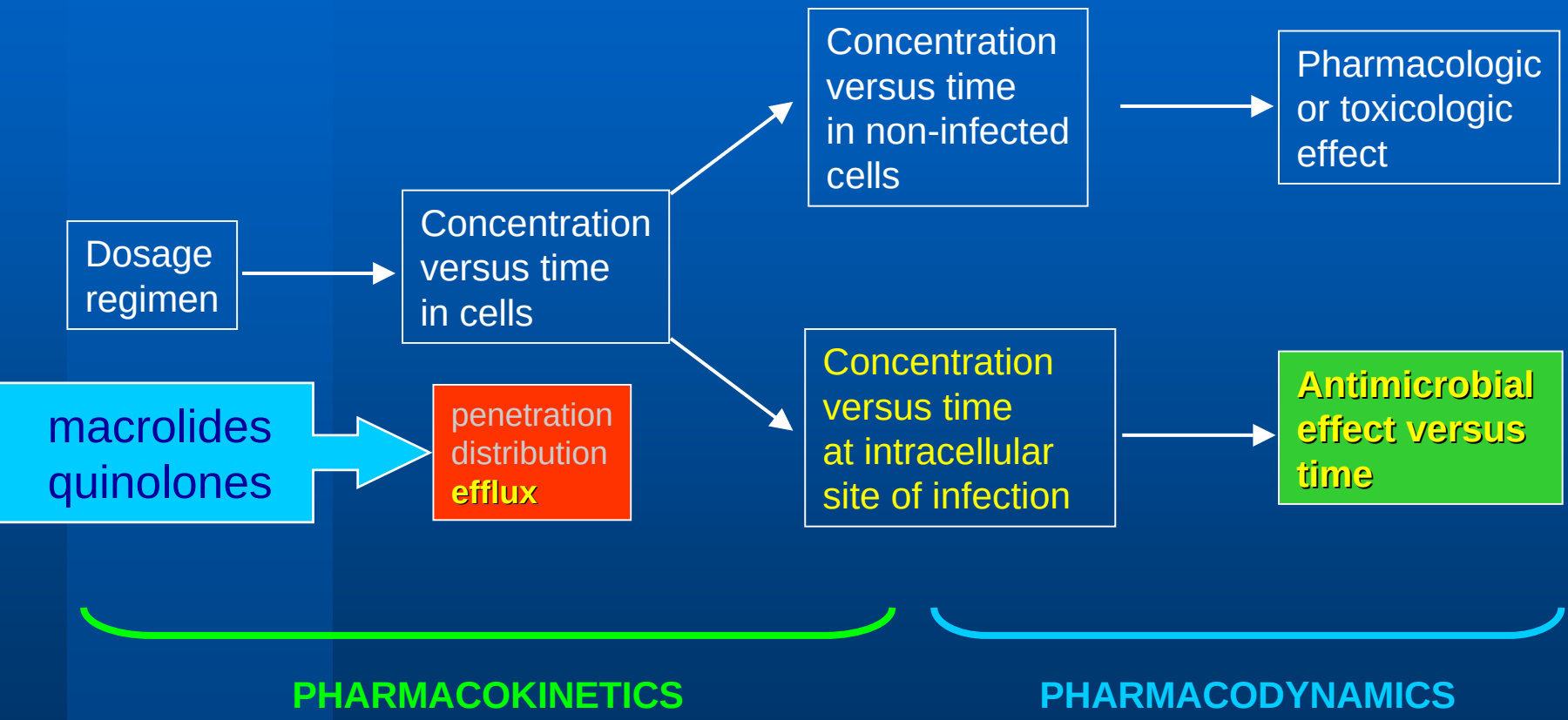
Intracellular “PK-PD”



Intracellular “PK-PD”



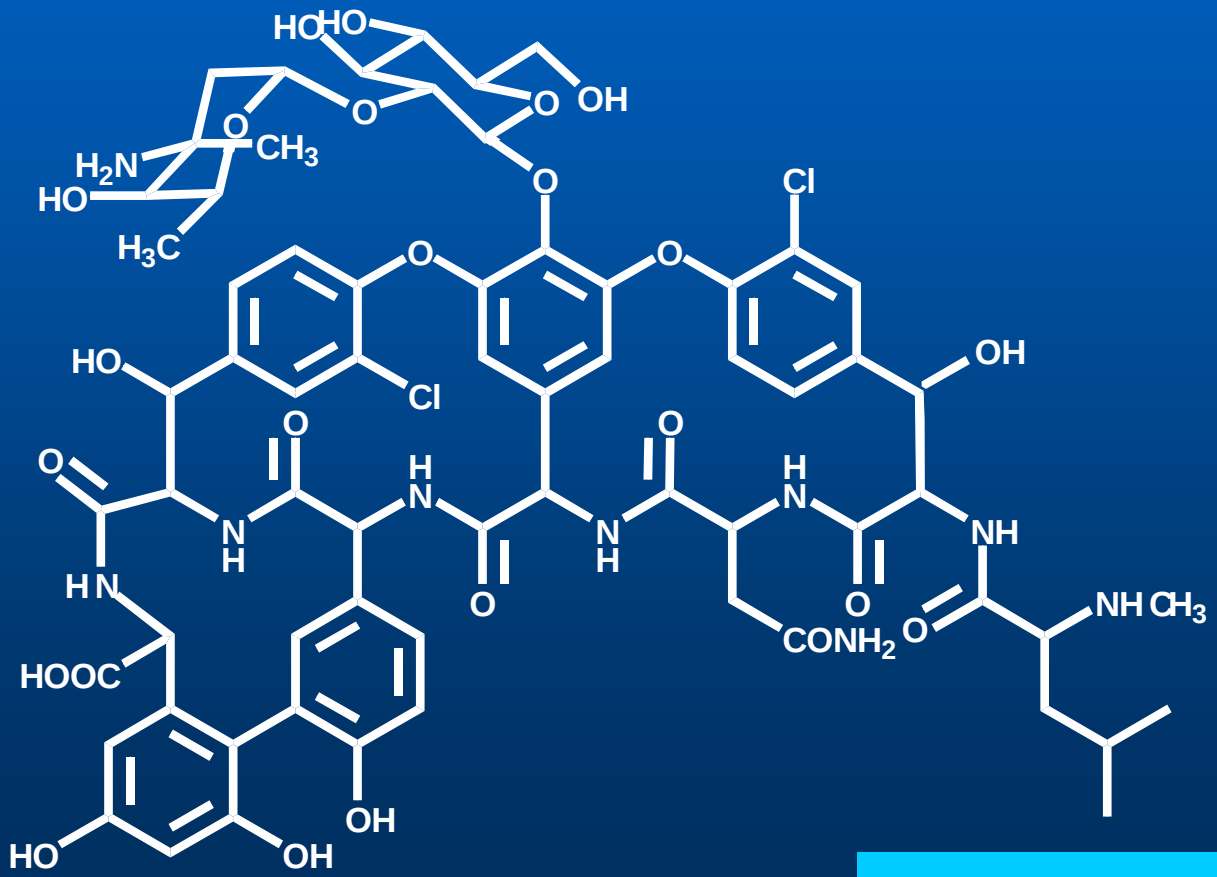
Intracellular “PK-PD”



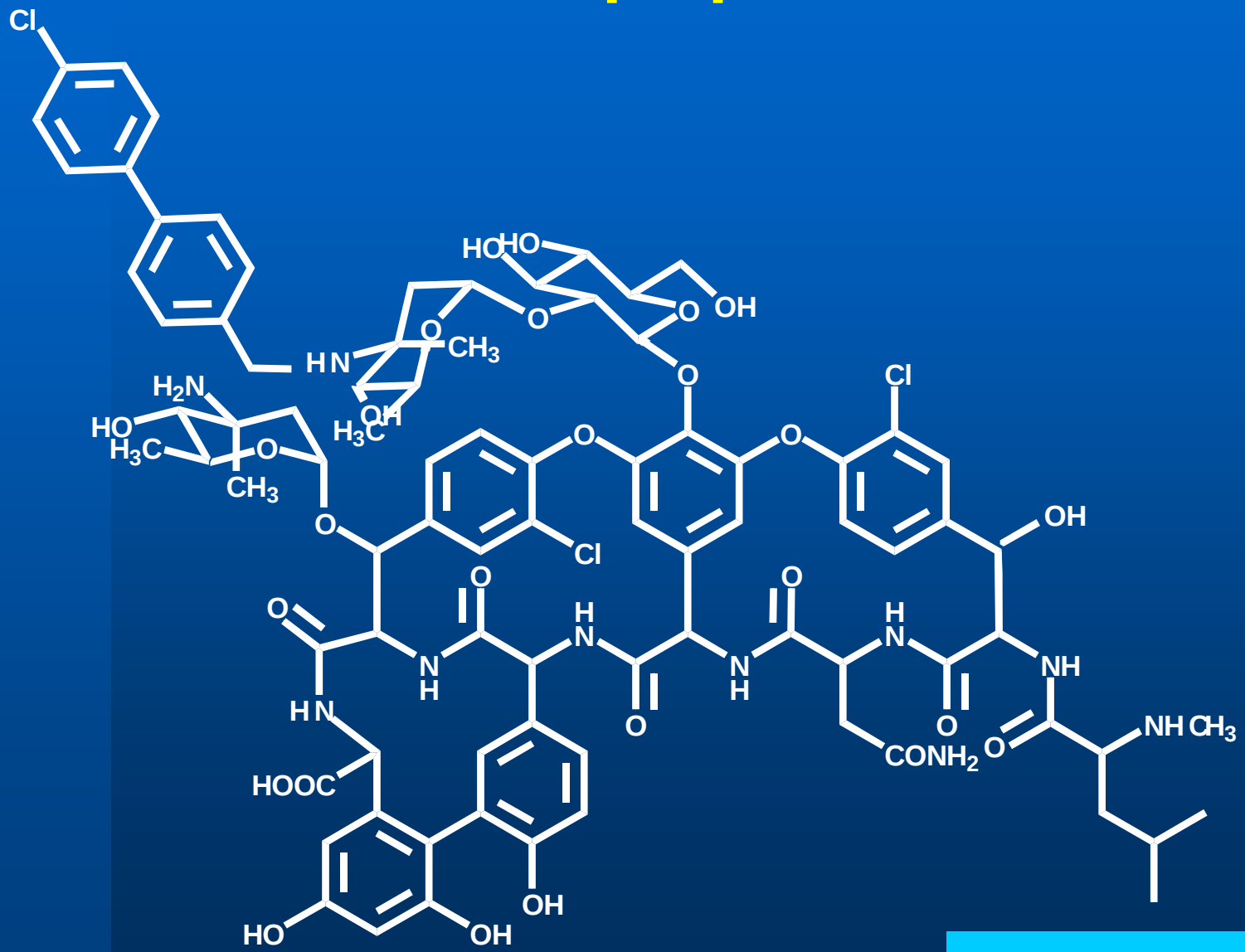
Accumulation of magic bullets in eukaryotic cells

glycopeptides

Vancomycin, the parent compound

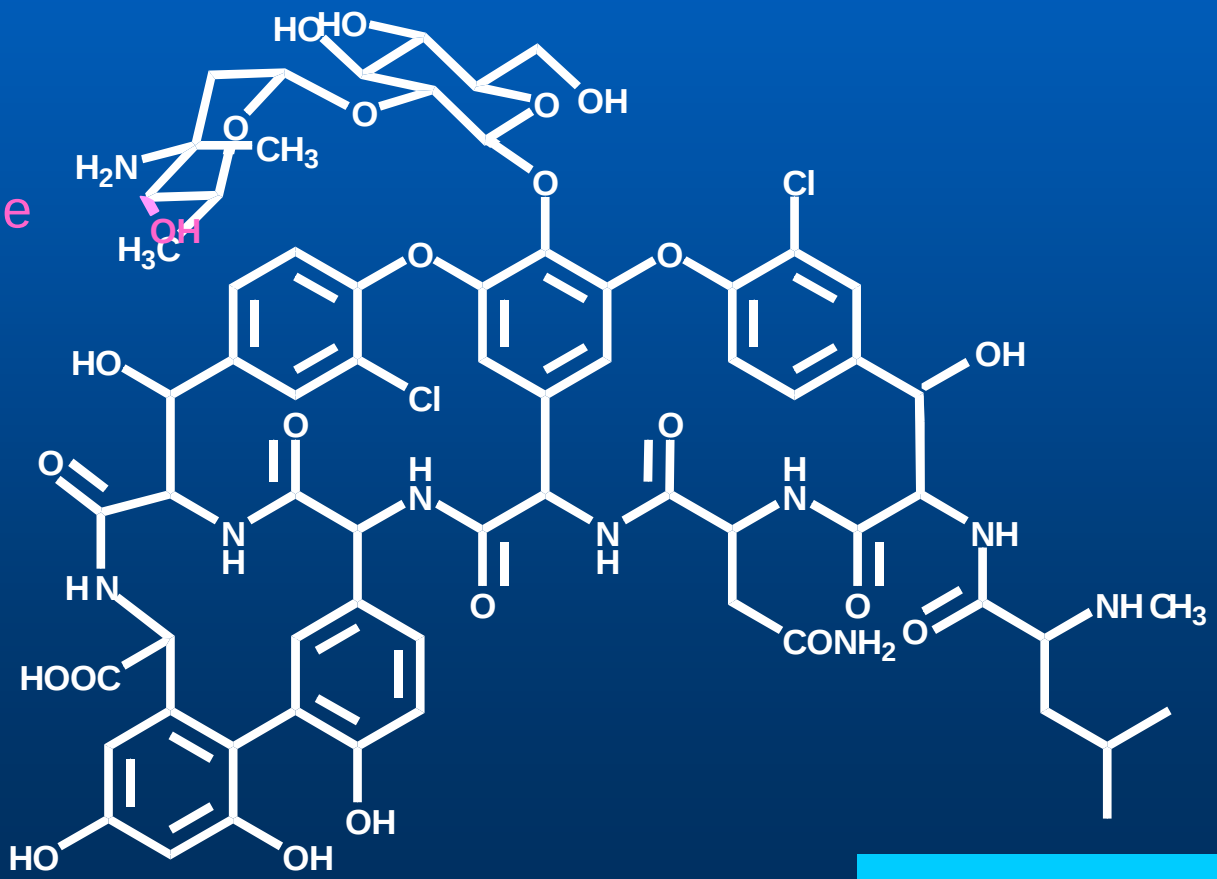


The glycopeptide oritavancin, a voluminous, amphiphilic molecule

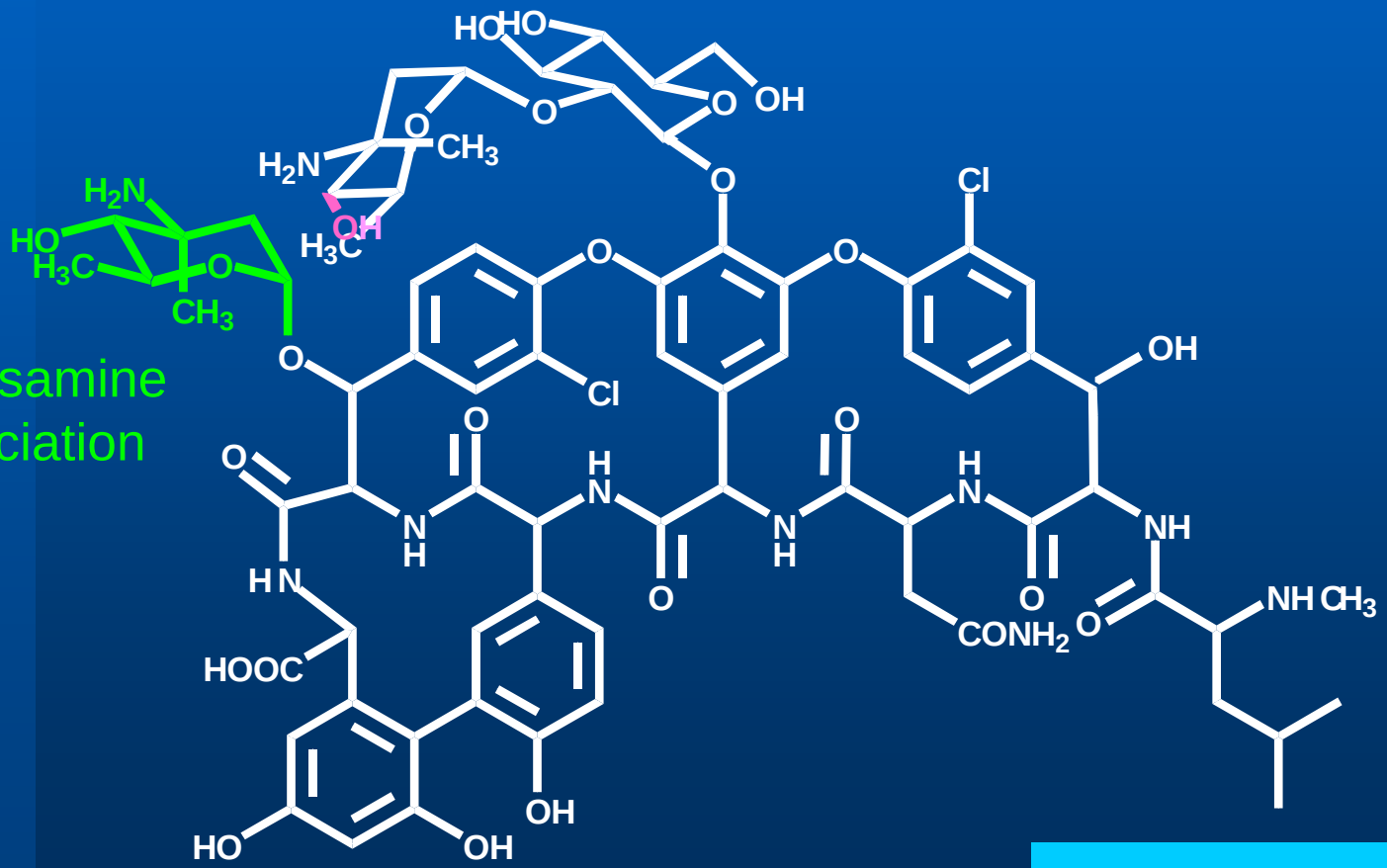


From vancomycin to oritavancin

epi-vancosamine



From vancomycin to oritavancin



4-*epi*-vancosamine
↗ self-association capacity

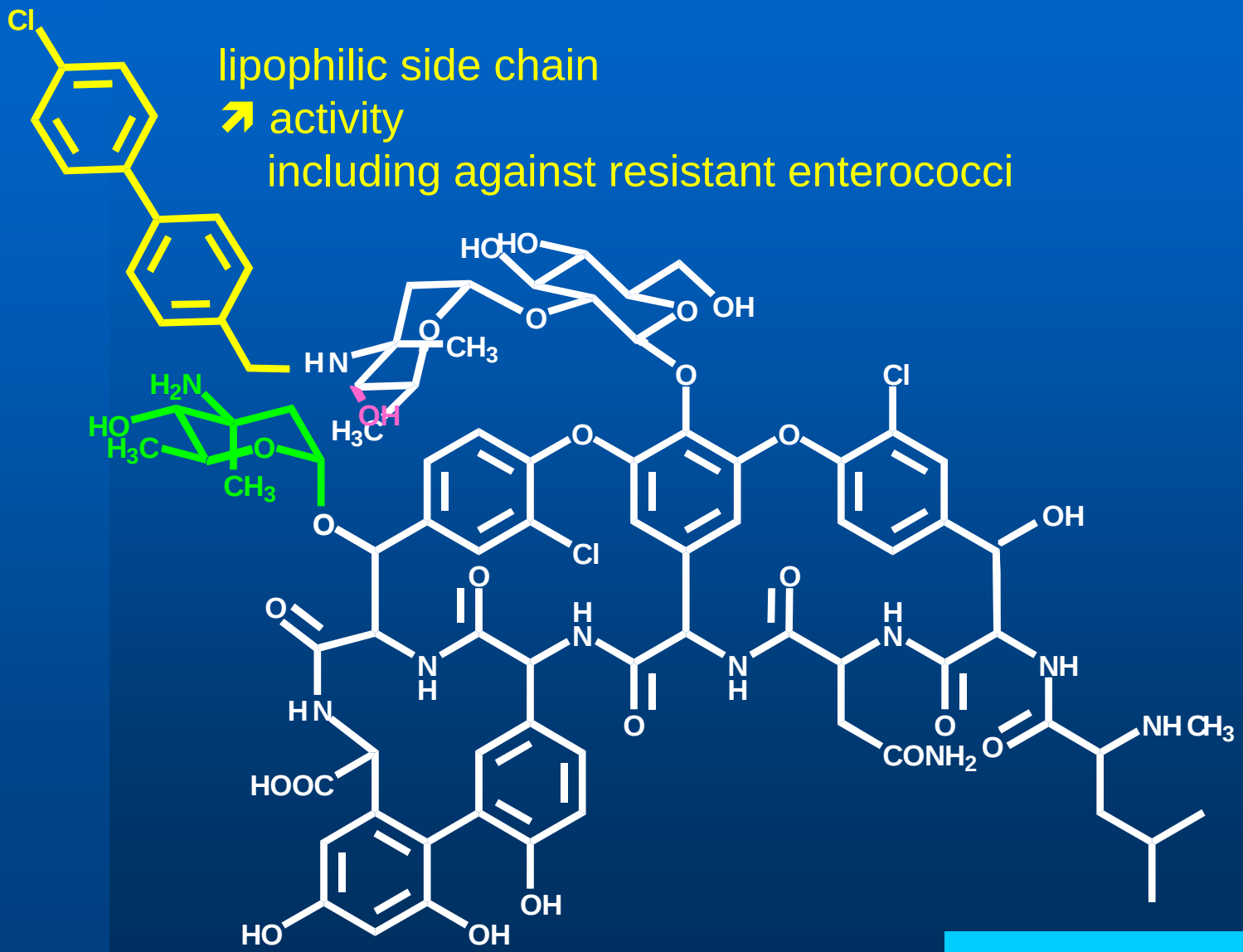
LY264626

From vancomycin to oritavancin

lipophilic side chain

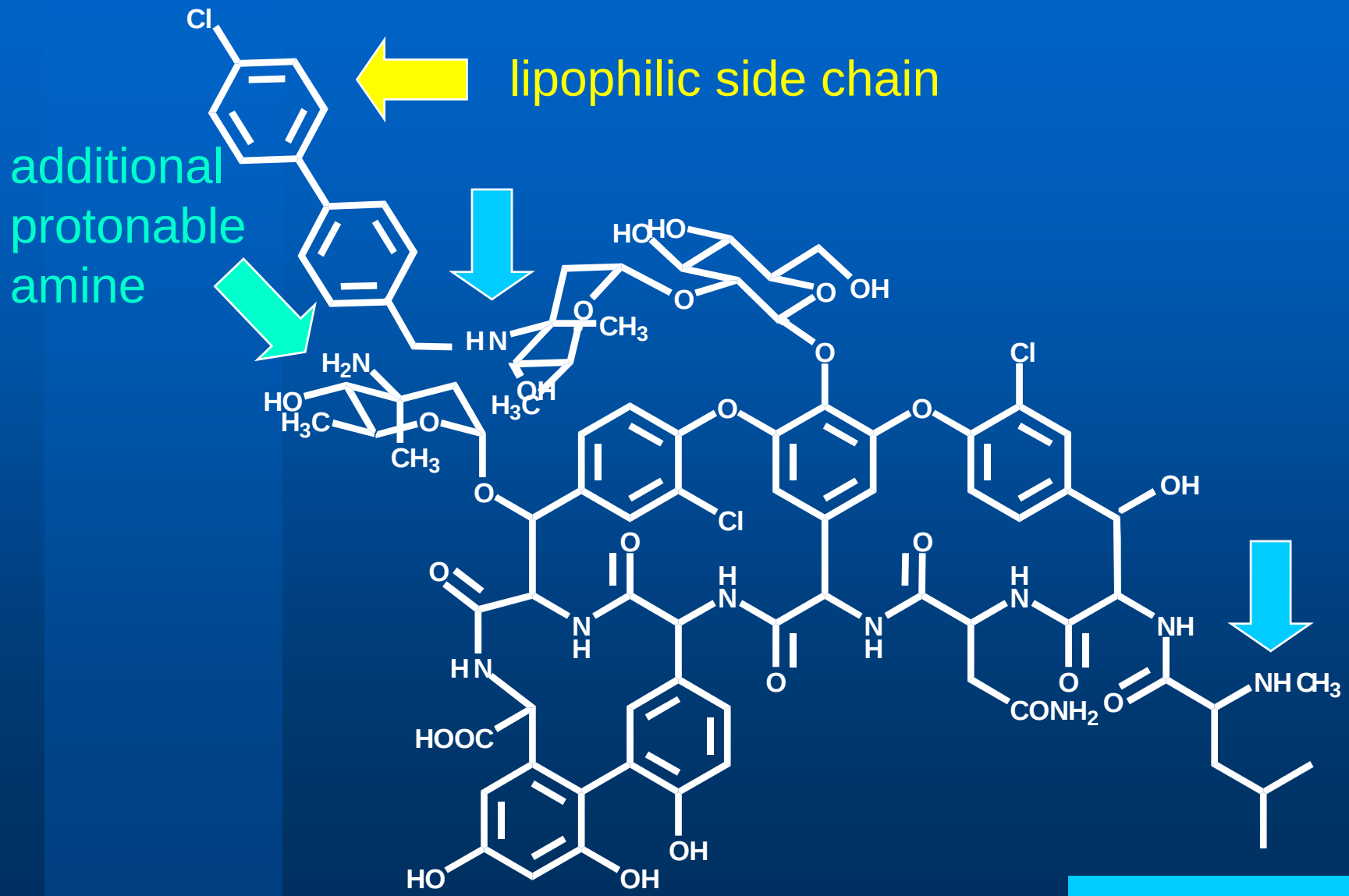
↗ activity

including against resistant enterococci



Cooper *et al* (1996) J Antibiot 49: 575-81

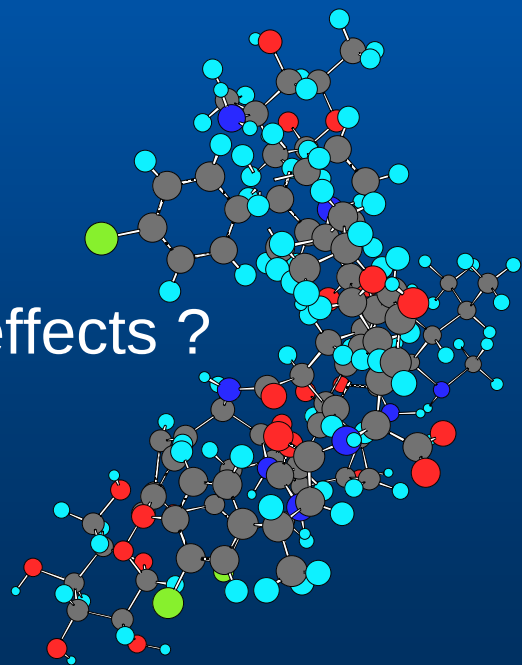
Oritavancin, a cationic amphiphile



Oritavancin, a cationic amphiphile

New chemical entity

- New pharmacodynamic properties ?
- New pharmacokinetic profile ?
- But also ... new potential side effects ?



Spectrum of activity

bacteria	resistance	vancomycin	oritavancin
enterococci	susc.	1-2	0.06-0.25
	VanA	>128	1-4
	VanB	8-128	0.125
S. aureus	Methi-S	1-2	1
	Methi-R	1-4	1-2
	GISA	8	1-8
	GRSA	> 128	0.5

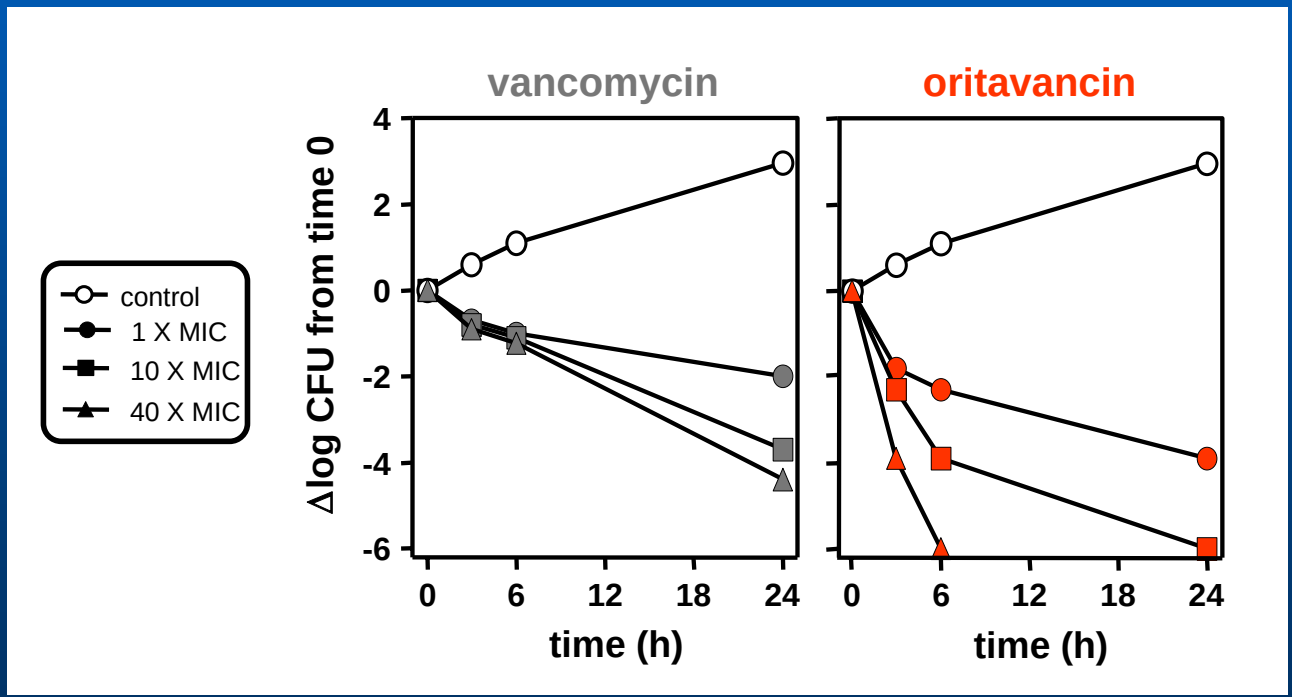
- **highly active on susc. enterococci**
- **active on VAN-resistant strains**

Van Bambeke *et al*. (2004) Drugs 64:913-36

Pharmacodynamic profile

VANCOMYCIN:
modestly bactericidal
- **slow**
- **no or little conc. effect**

ORITAVANCIN:
highly bactericidal:
- **rapid**
- **conc. dependent**



S. aureus

Barcia-Macay *et al*. submitted

Pharmacokinetic properties

parameter	Vancomycin (15 mg/kg)	Oritavancin (3 mg/kg)
peak (mg/L)	20-50	31
trough (mg/L)	5-12 (24 h)	1.7 (24 h)
protein binding	10-55 %	90 %
terminal t½ (h)	4-8	360

- **daily administration**
- **retention in the organism ?**

Van Bambeke *et al*. (2004) Drugs 64:913-36

Aim of the study

- activity against (multi-resistant) Gram-positive (*S. aureus*)
- rapid bactericidal activity
- retention in the organism

any place for intracellular infections?

cellular pharmacokinetics:

accumulation and subcellular distribution in eukaryotic cells

cellular pharmacodynamics:

activity against intracellular *S. aureus*

cellular toxicity:

morphological and biochemical alterations

Aim of the study

- activity against (multi-resistant) Gram-positive (*S. aureus*)
- rapid bactericidal activity
- retention in the organism

any place for intracellular infections?

cellular pharmacokinetics:

accumulation and subcellular distribution in eukaryotic cells

cellular pharmacodynamics:

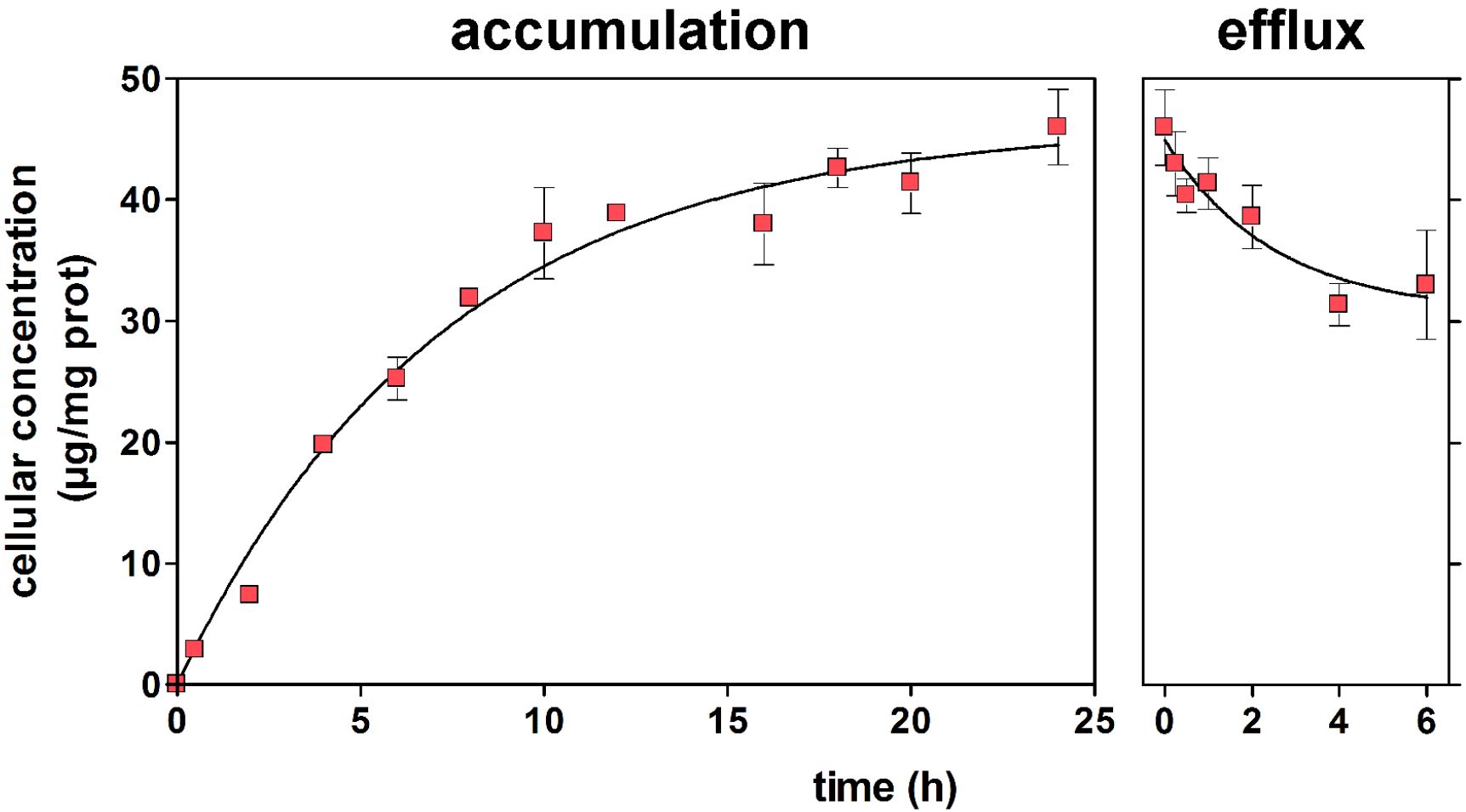
activity against intracellular bacteria

cellular toxicity:

morphological and biochemical alterations

Kinetics of accumulation-efflux

Oritavancin accumulation and release are slow

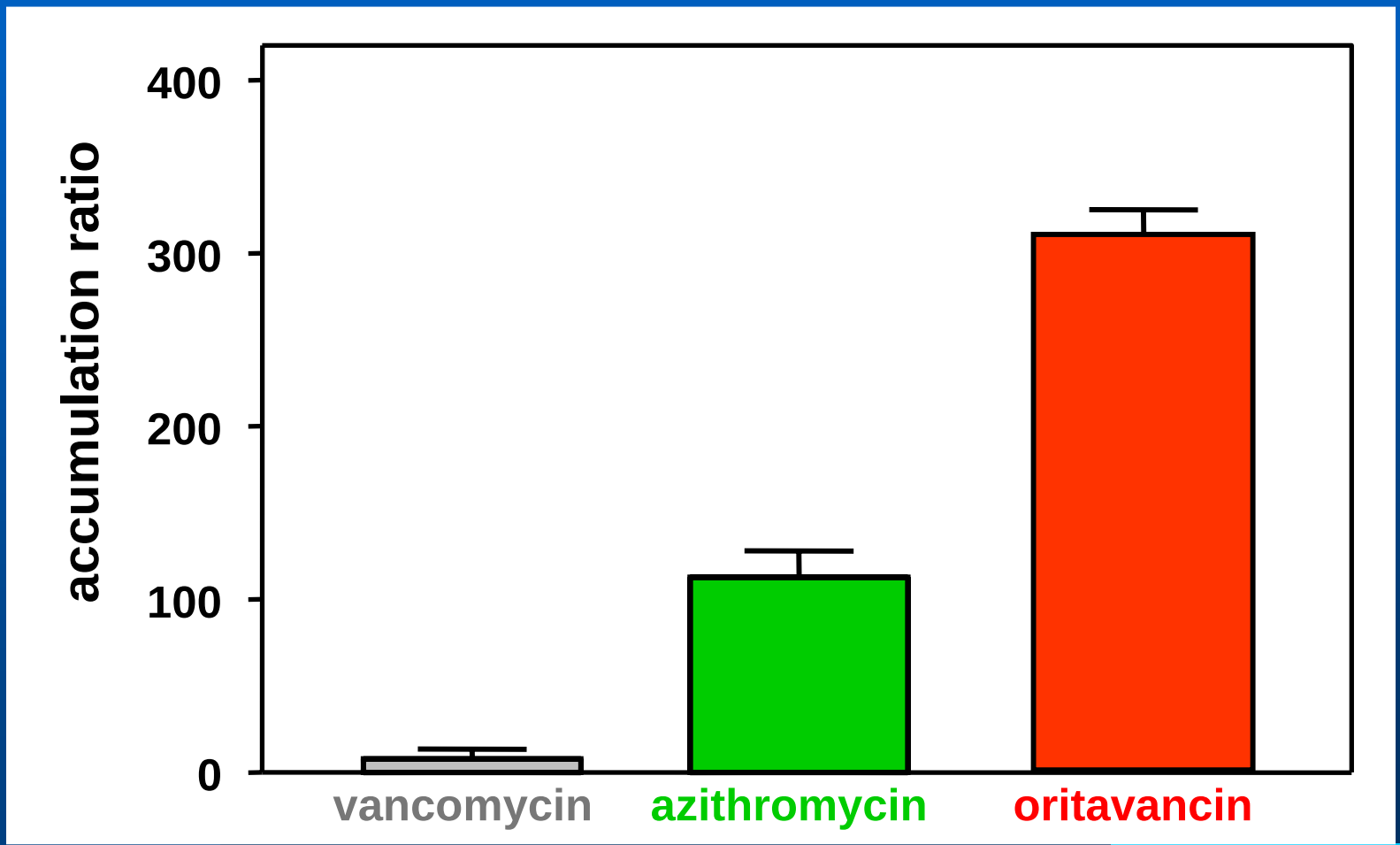


J774 macrophages; extracell. conc. 25 mg/L; 24 h

Van Bambeke *et al*. (2004) AAC 48:2853-60

Comparison with other antibiotics

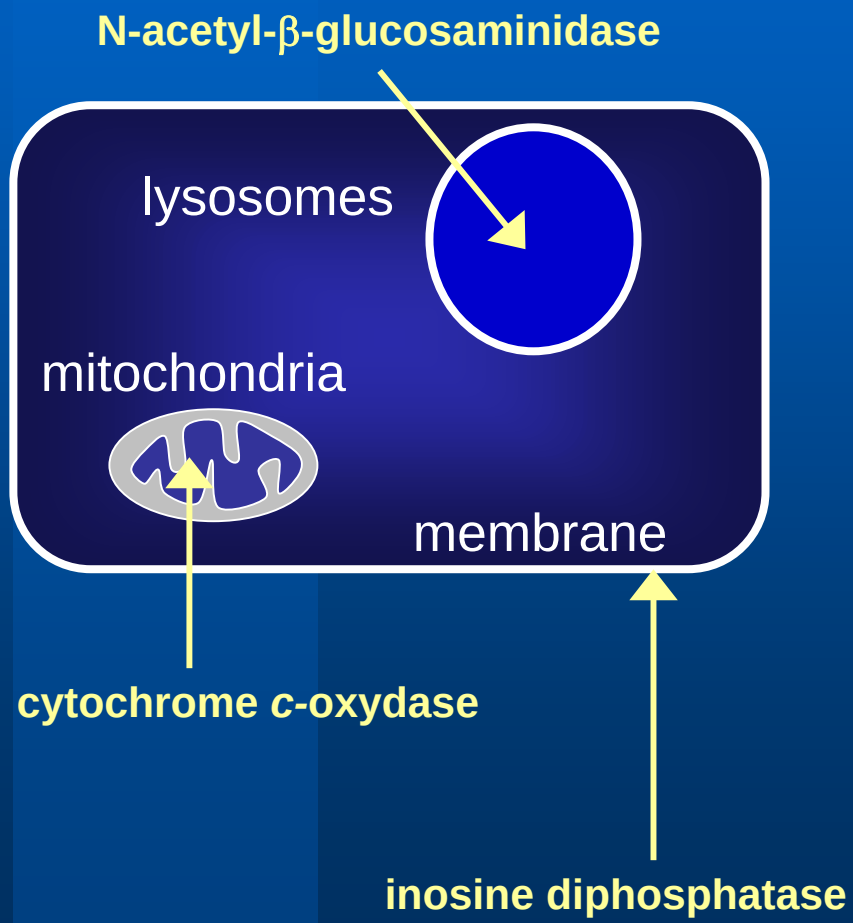
Oritavancin reaches exceptional cellular accumulation levels

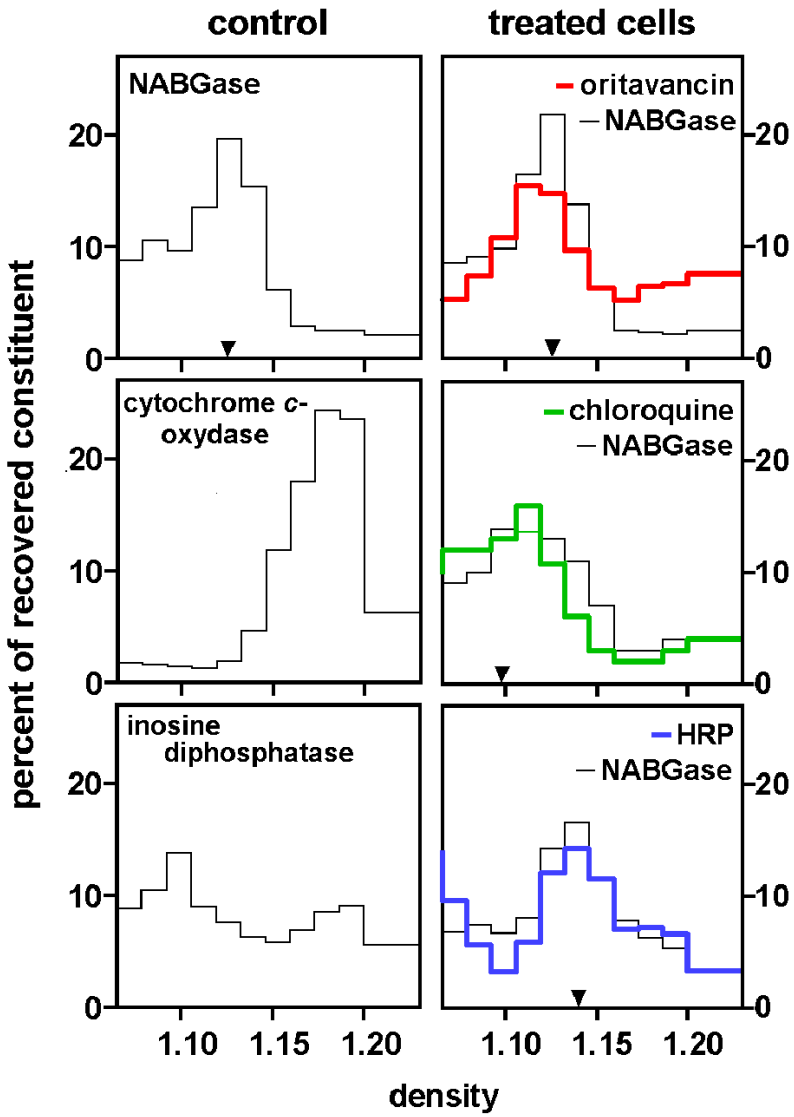


J774 macrophages; extracell. conc. 25 mg/L; 24 h

Van Bambeke *et al*. (2004) AAC 48:2853-60

Subcellular localization

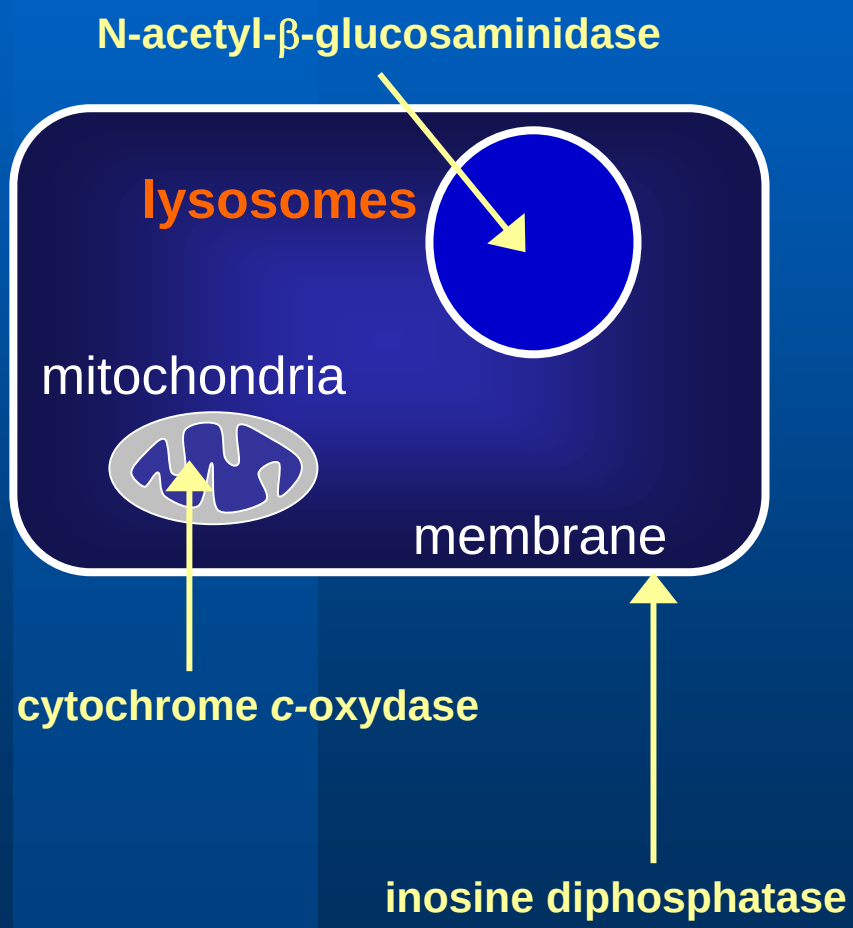


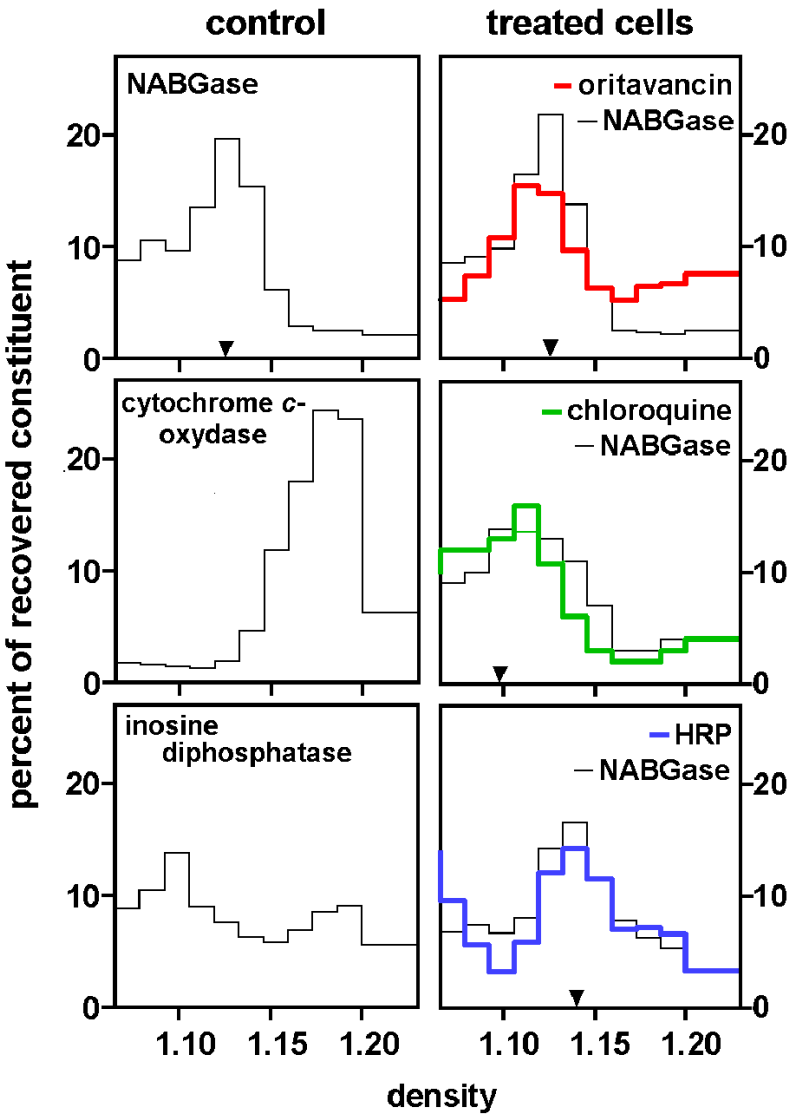


Van Bambeke *et al*. (2004) AAC 48:2853-60

Subcellular localization

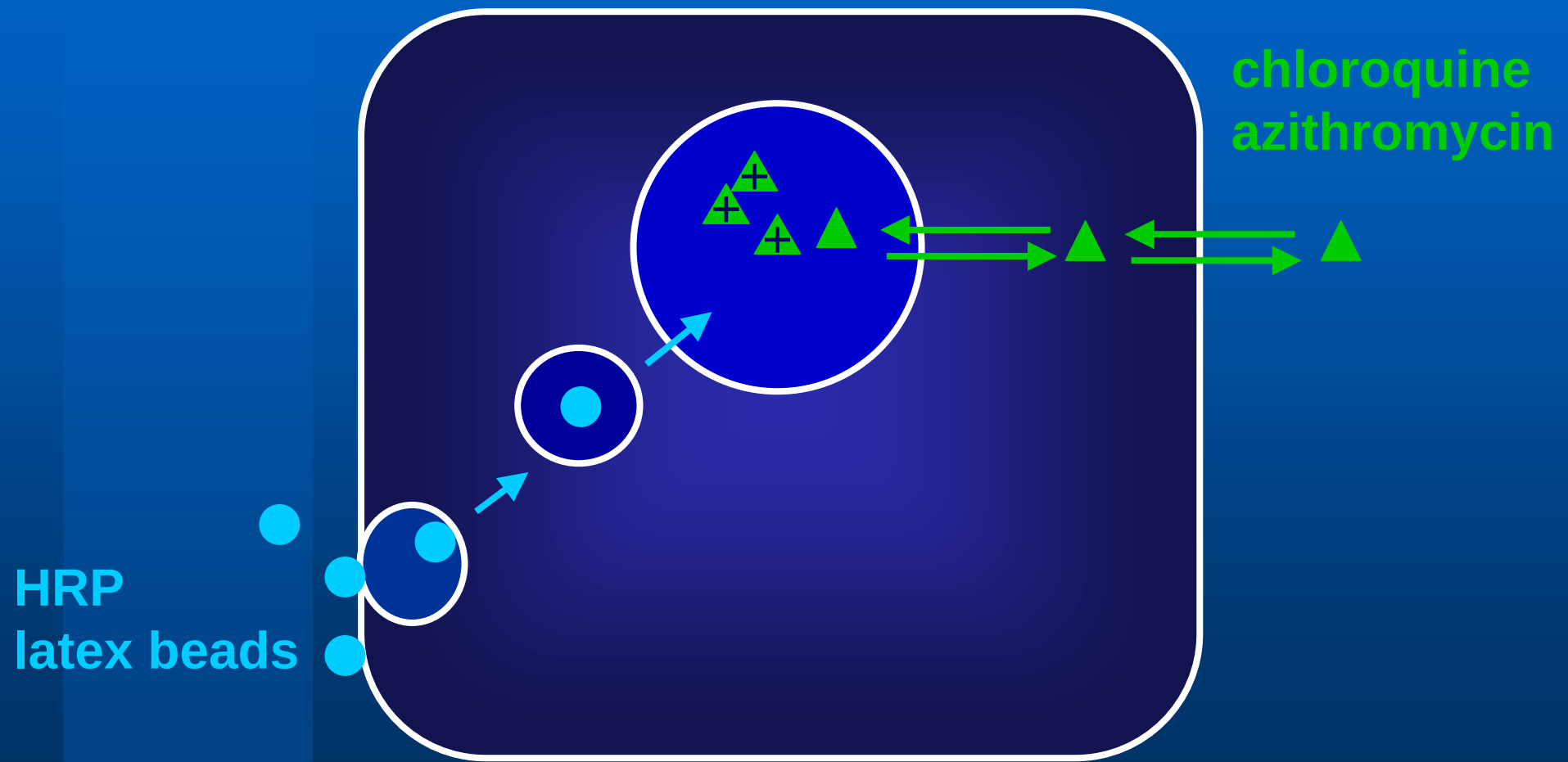
Oritavancin is a lysosomotropic antibiotic





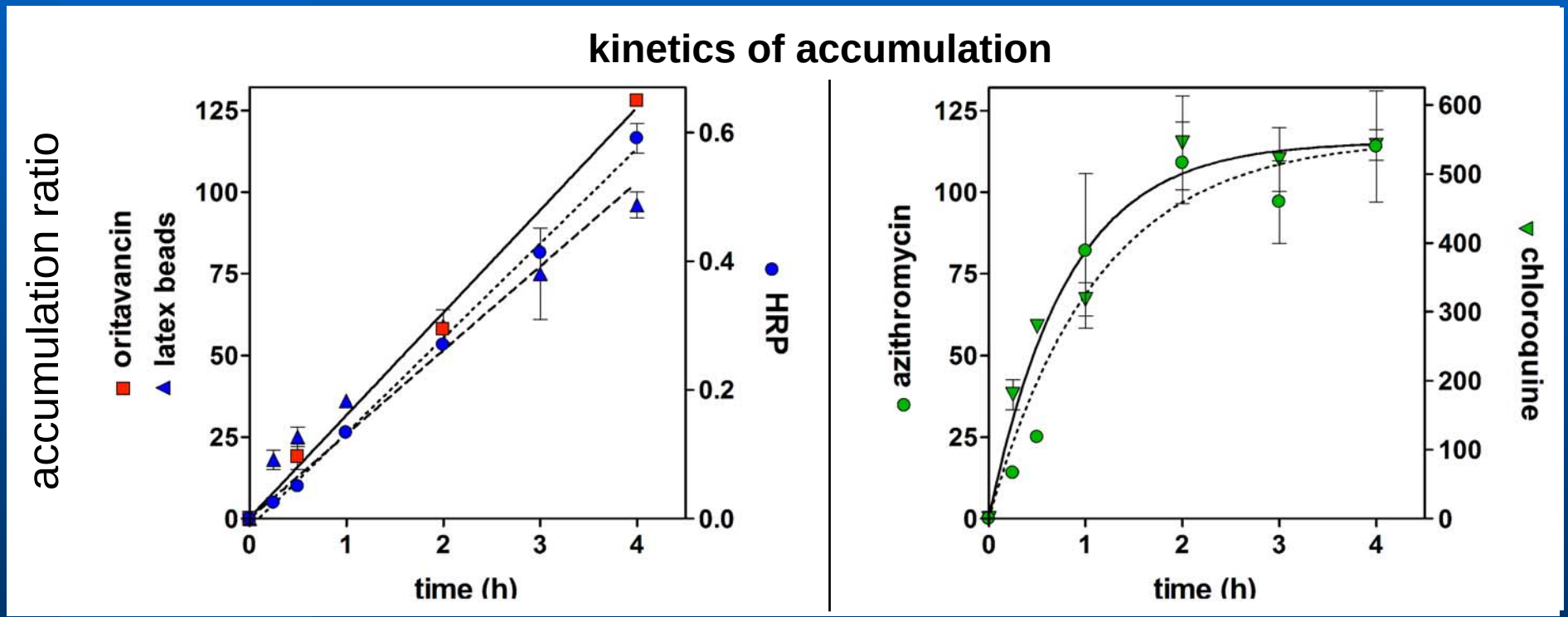
Van Bambeke *et al*. (2004) AAC 48:2853-60

Mechanism of cellular accumulation



Mechanism of cellular accumulation

Oritavancin kinetics of accumulation are very similar to those of tracers of (adsorptive) endocytosis



Van Bambeke *et al*. (2004) AAC 48:2853-60

Aim of the study

- activity against multi-resistant Gram-positive (*S. aureus*)
- rapid bactericidal activity
- retention in the organism

any place for intracellular infections?

cellular pharmacokinetics:

accumulation and distribution in eukaryotic cells

cellular pharmacodynamics:

activity against intracellular bacteria

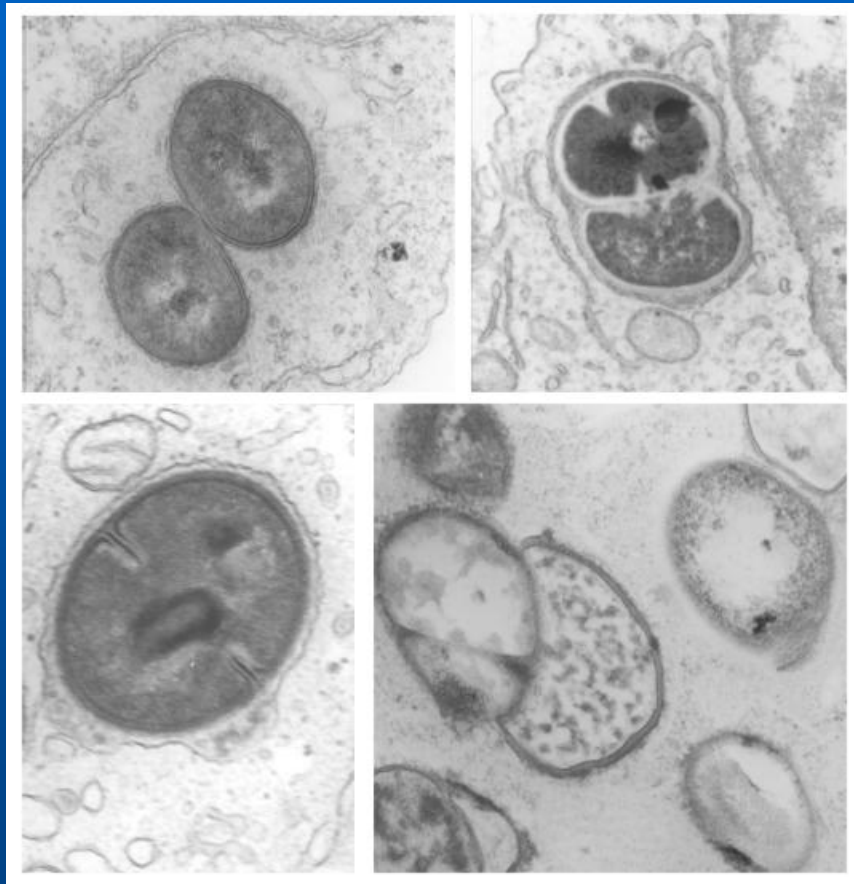
cellular toxicity:

morphological and biochemical alterations

Intracellular activity on *S. aureus*

control

oritavancin



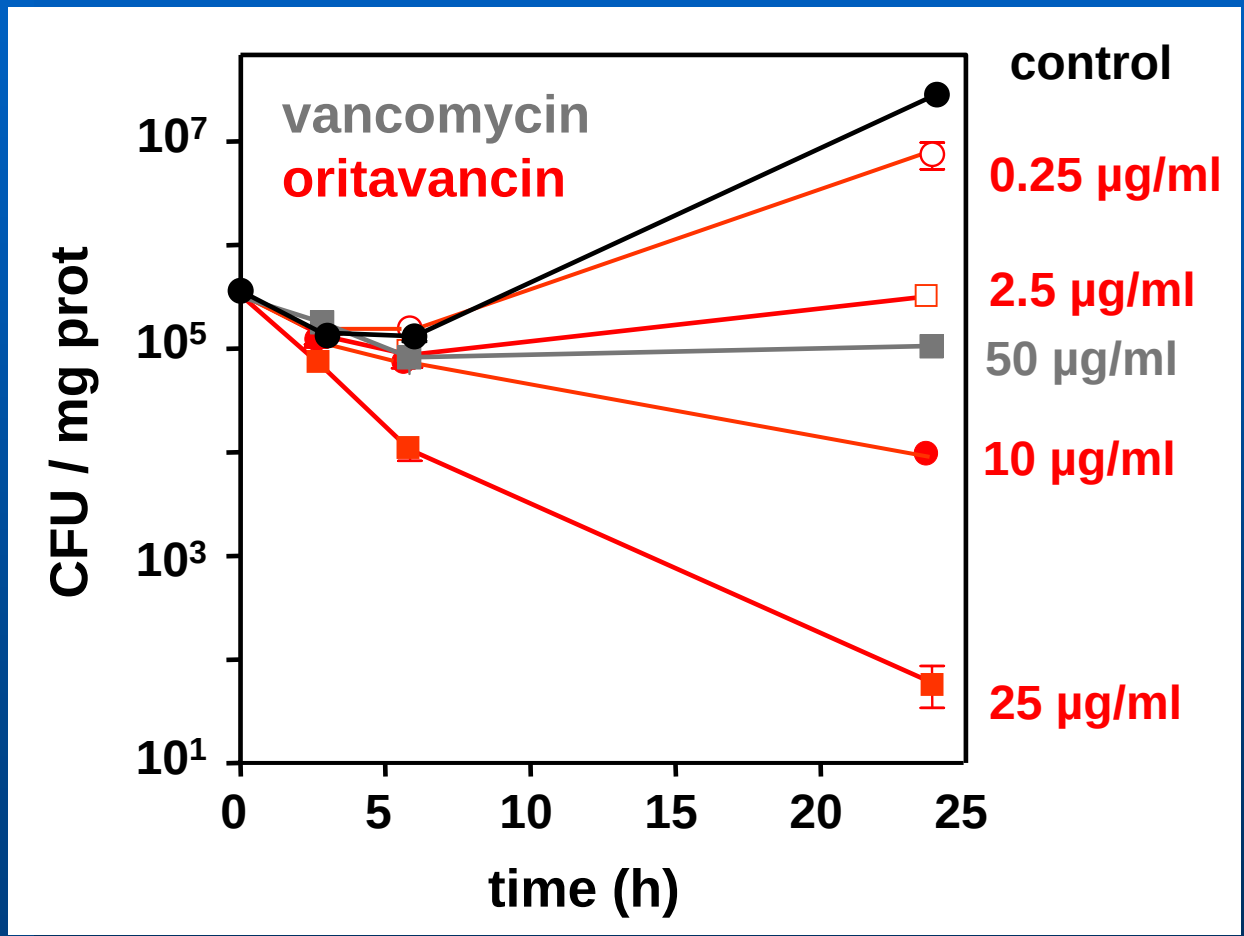
Oritavancin can destroy intracellular bacteria

THP-1 macrophages; extracell. conc. 25 mg/L; 24 h

Barcia-Macay *et al*. submitted

Time-effect for intracellular activity

Oritavancin shows time- and concentration-dependent intracellular bactericidal effects

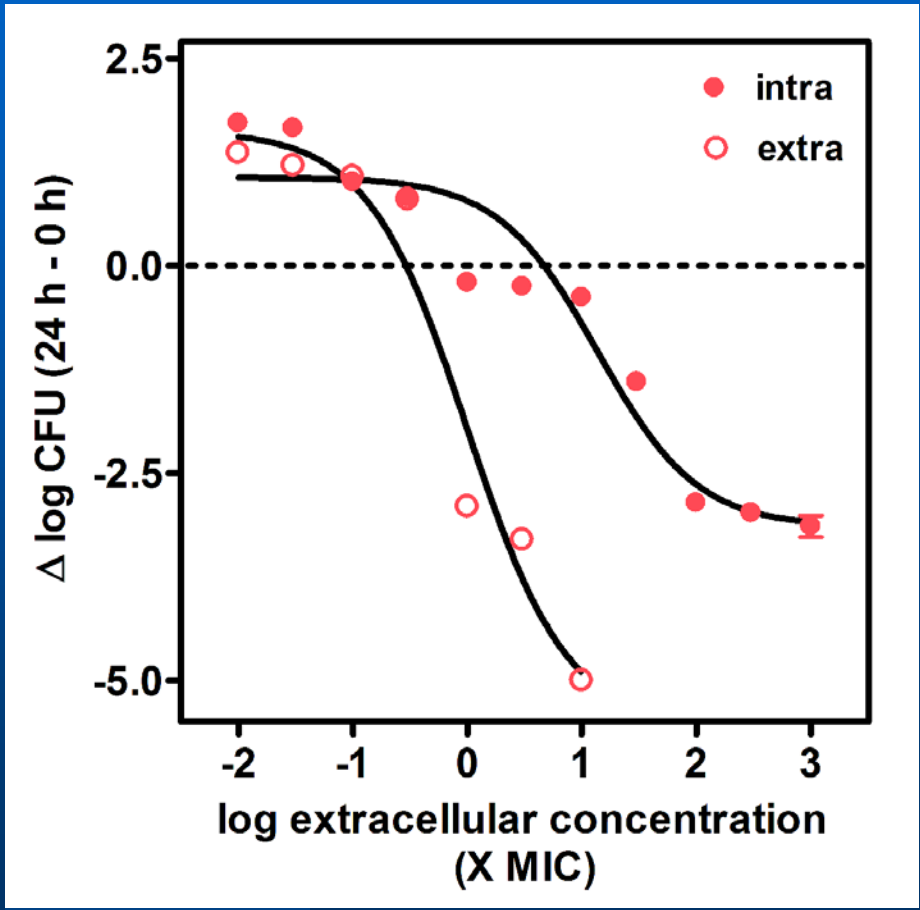


J774 macrophages

Seral *et al* (2003) AAC 47: 2283-92

Dose-effect for extracell. *vs* intracell. activity

intracellular activity < extracellular activity



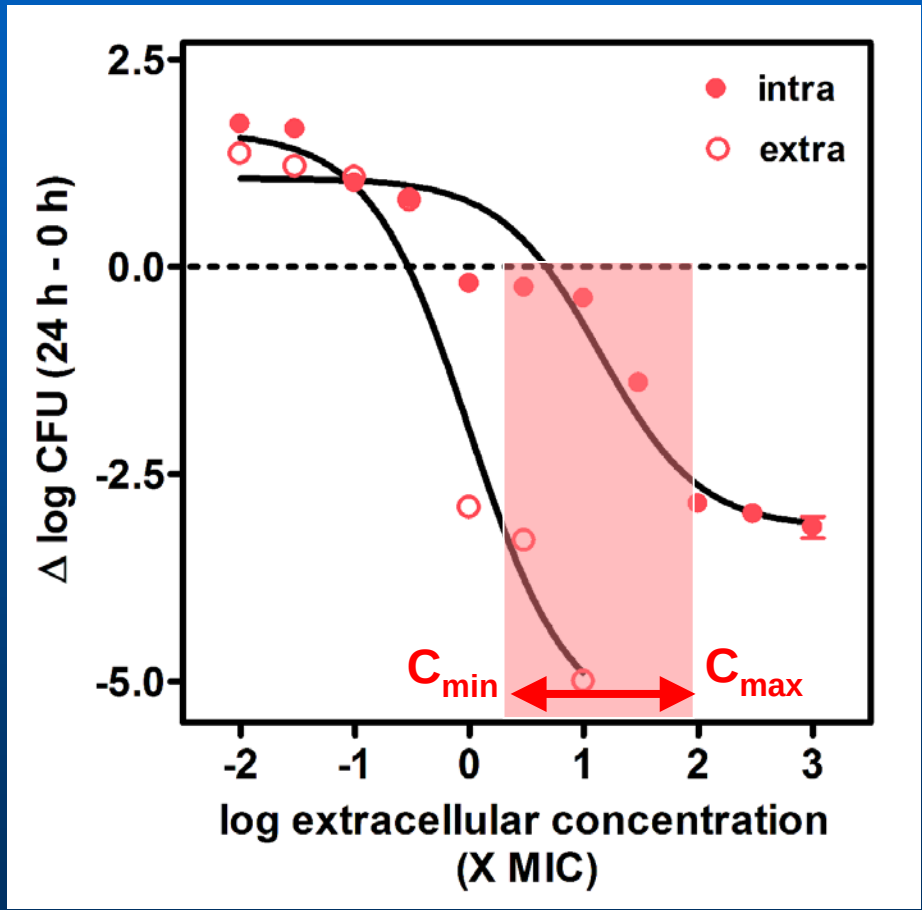
	extra	intra
static conc.	0.3 X MIC	4.8 X MIC
max. effect	- 5.55 log	- 3.15 log

THP-1 macrophages; 24 h

Barcia-Macay *et al*. submitted

Dose-effect

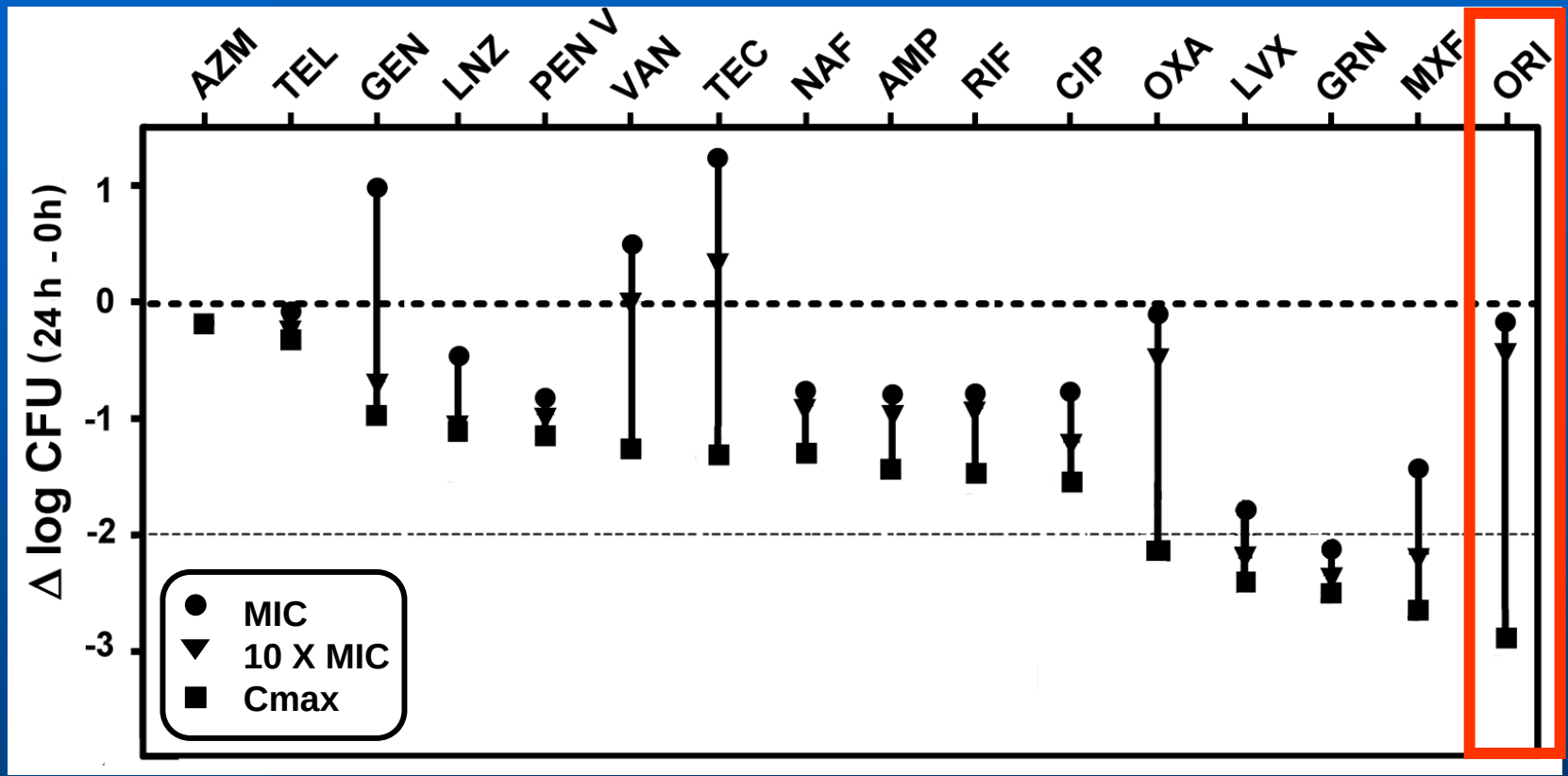
intracellular activity < extracellular activity,
but bactericidal effects reached at clinically-relevant concentrations



Barcia-Macay *et al*. submitted

Comparison with other antibiotics

oritavancin is one of the most active drugs against intracellular *S. aureus*



THP-1 macrophages; 24 h

Barcia-Macay *et al*. submitted

Aim of the study

- activity against multi-resistant Gram-positive (*S. aureus*)
- rapid bactericidal activity
- retention in the organism

any place for intracellular infections?

cellular pharmacokinetics:

accumulation and subcellular distribution in eukaryotic cells

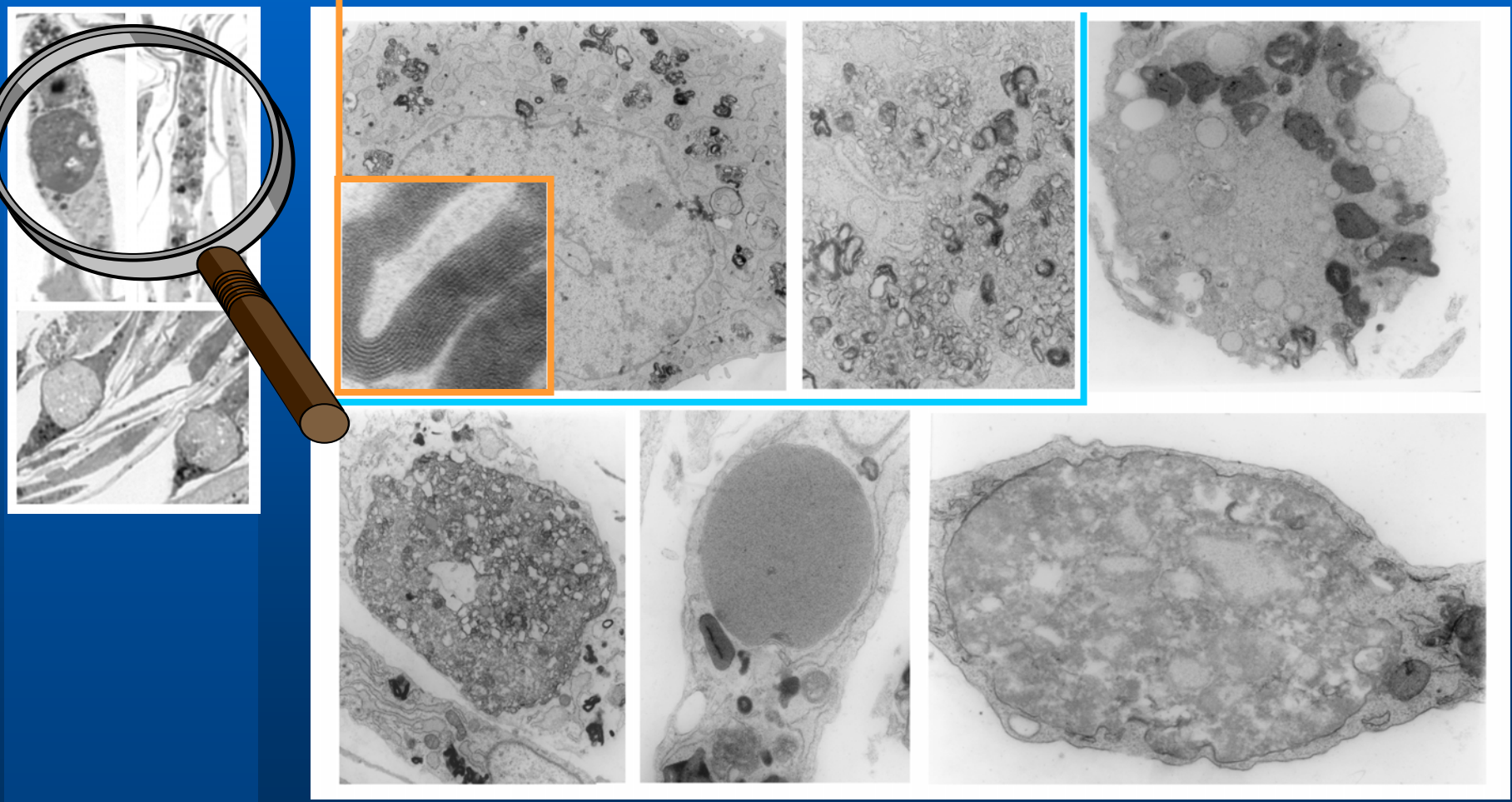
cellular pharmacodynamics:

activity against intracellular bacteria

cellular toxicity:

morphological and biochemical alterations

Morphological studies

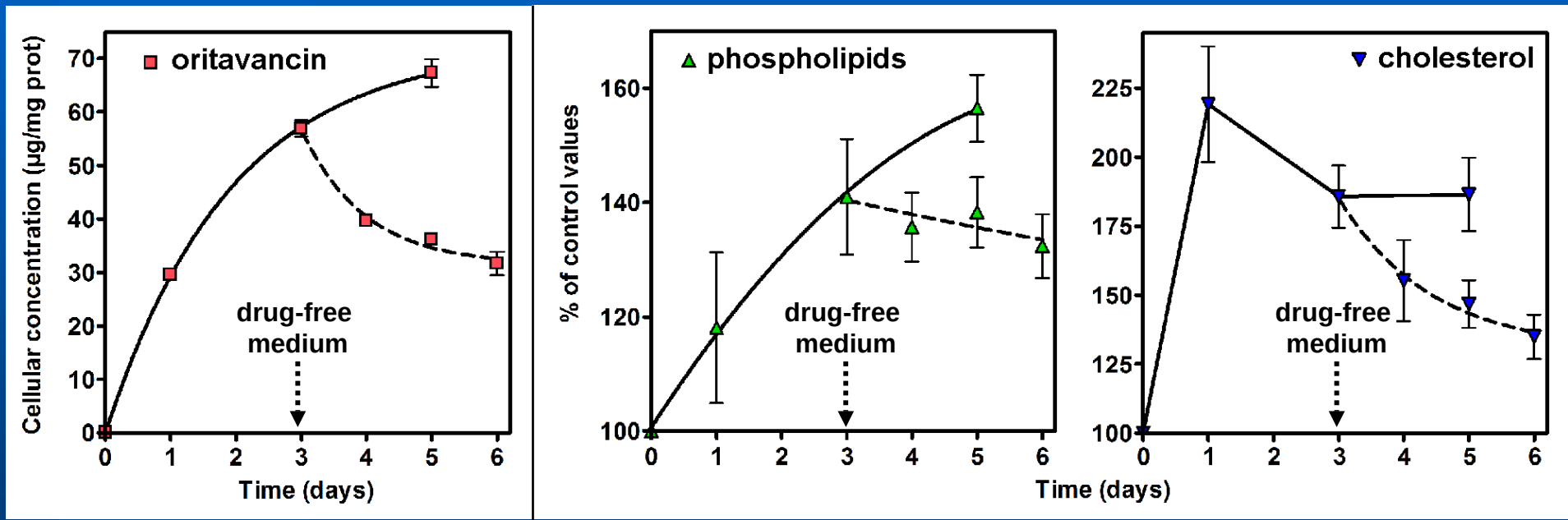


Rat embryo fibroblasts; 25 mg/L; 3 days
J774 macrophages, 25 mg/L; 1 day

Van Bambeke *et al*. (2005) AAC – in press

Biochemical studies : time-effects

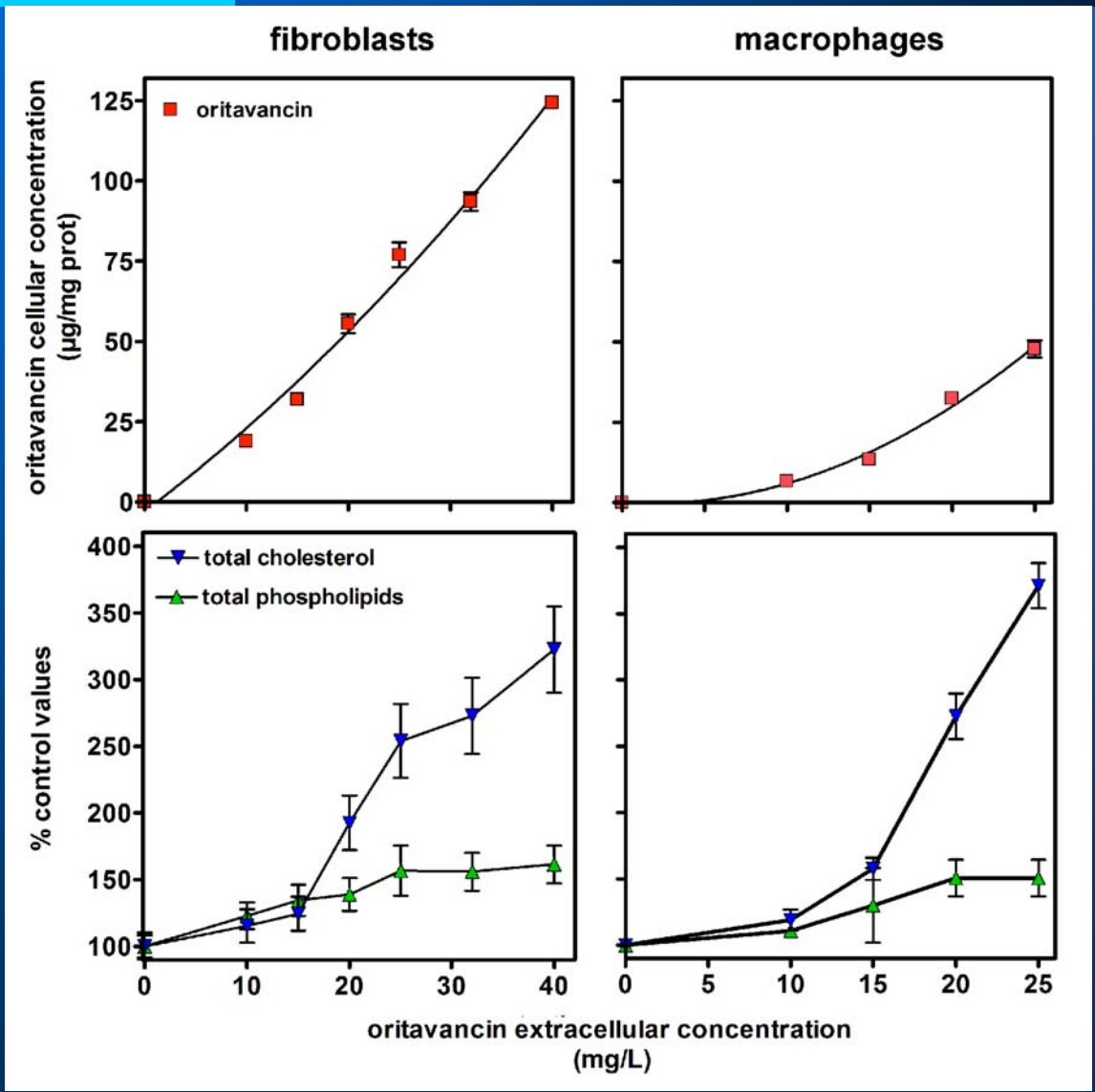
accumulation of phospholipids and cholesterol develops in parallel with oritavancin cellular concentration



Rat embryo fibroblasts; 25 mg/L

Van Bambeke *et al*. (2005) AAC – in press

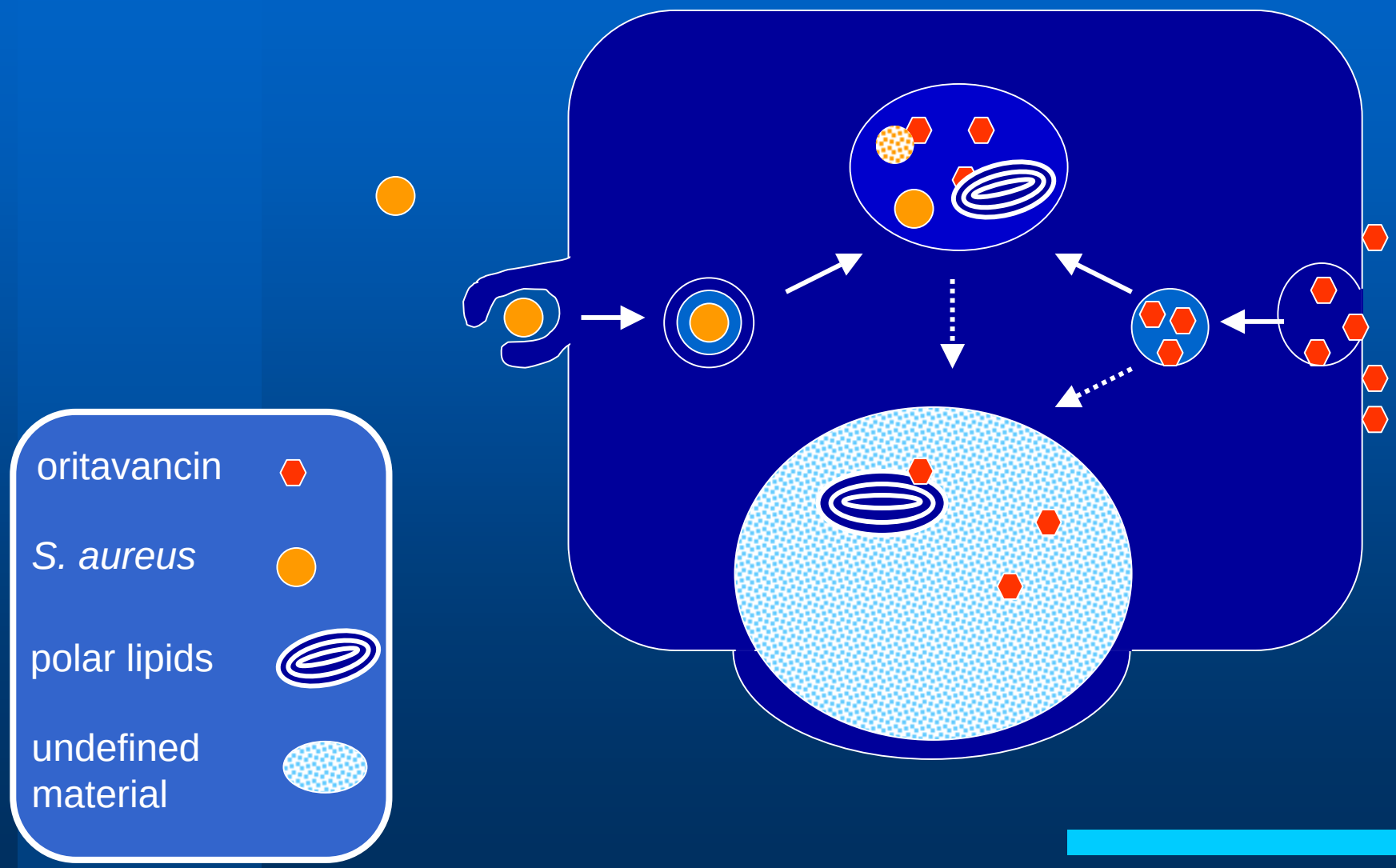
Biochemical studies : dose-effects



Rat embryo fibroblasts, 3 days
J774 macrophages, 1 day

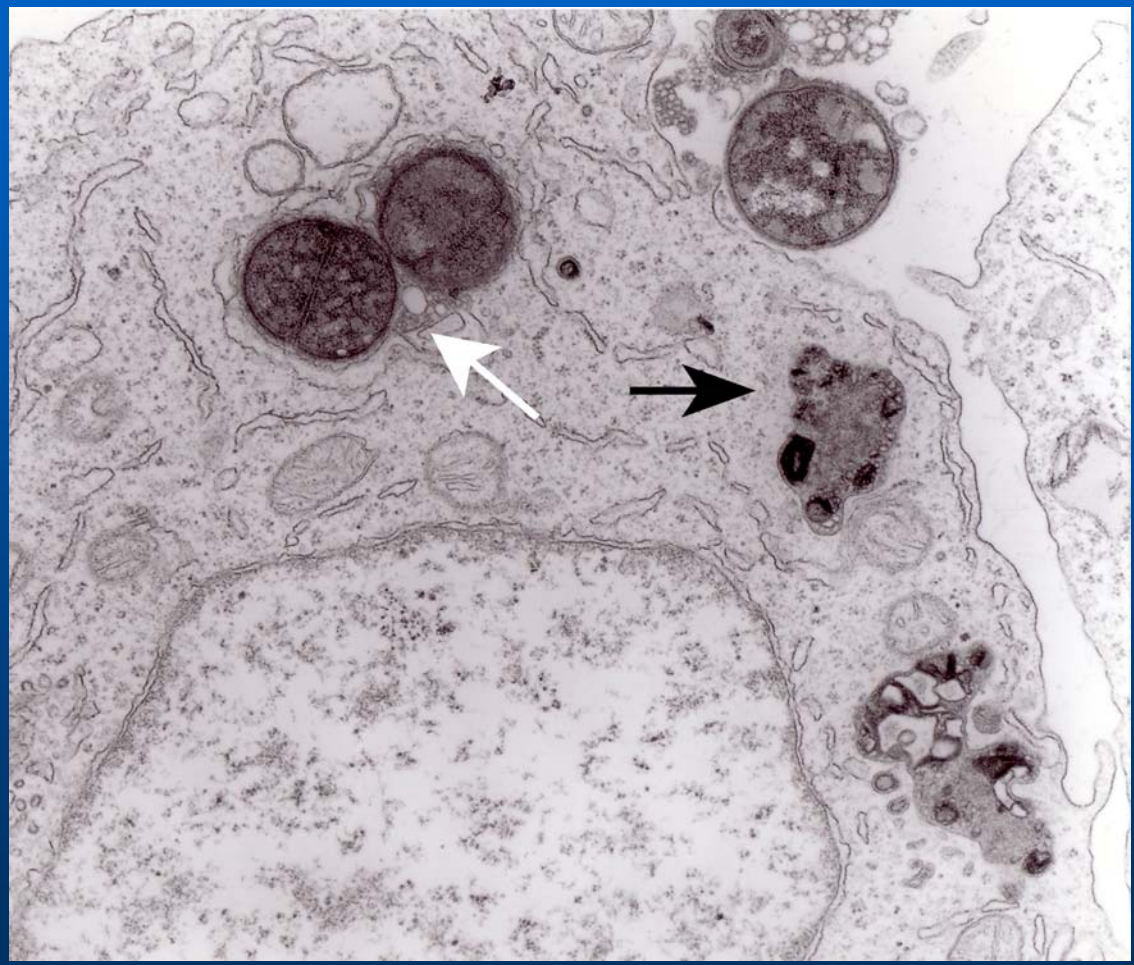
Van Bambeke *et al*. (2005) AAC – in press

Model of the interaction of oritavancin with eukaryotic cells



Can we dissociate activity from toxicity ?

cellular alterations co-exist with destroyed bacteria ….

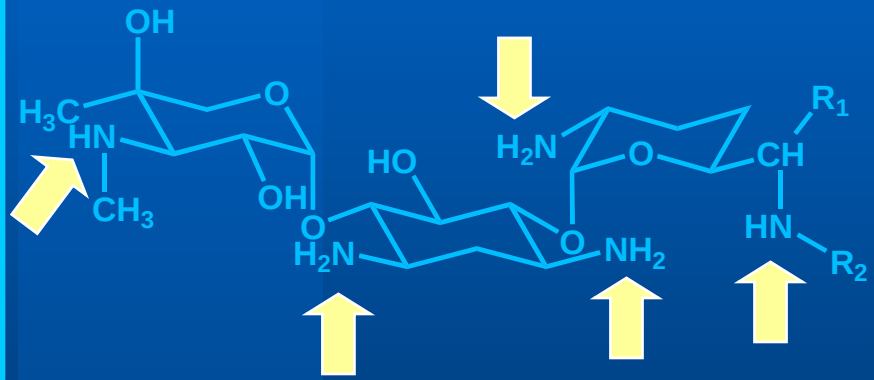


THP-1 macrophages; 25 mg/L; 24 h

Can we dissociate activity from toxicity ?

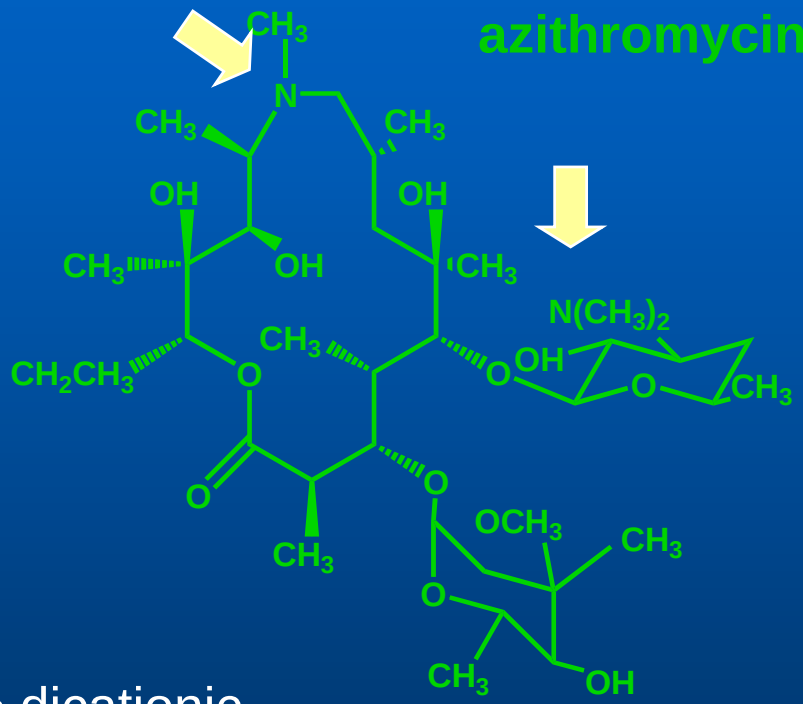
comparison with two other lysosomotropic cationic antibiotics

gentamicin



- polycationic, hydrophilic
- endocytosis
- phospholipidosis

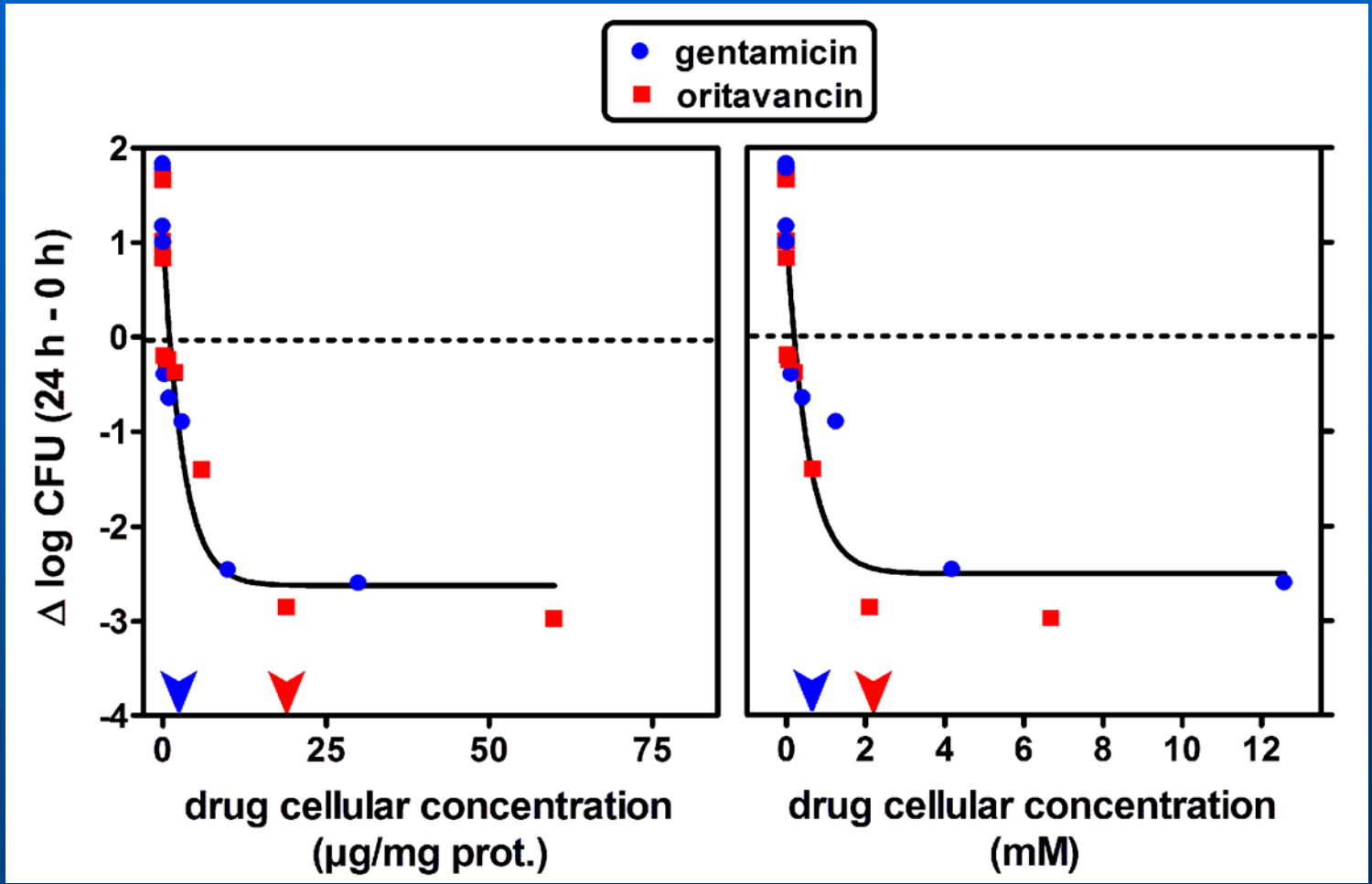
azithromycin



- dicationic
- diffusion/segregation
- accumulation of phospholipids and cholesterol

Lysosomotropic antibiotics and activity on *S. aureus*

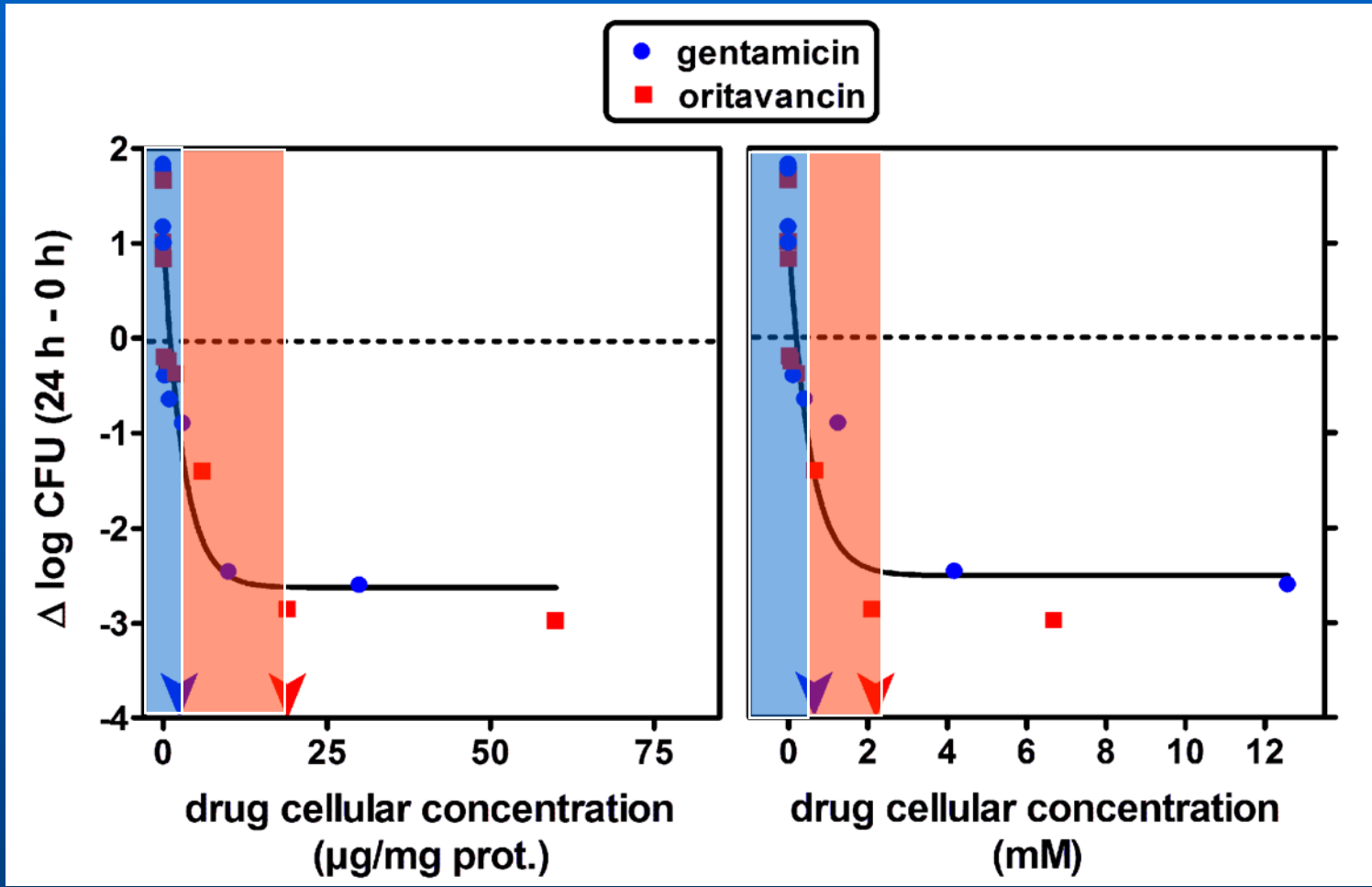
GEN and ORI are both conc.-dependent intracellularly



J774 macrophages

Lysosomotropic antibiotics and activity on *S. aureus*

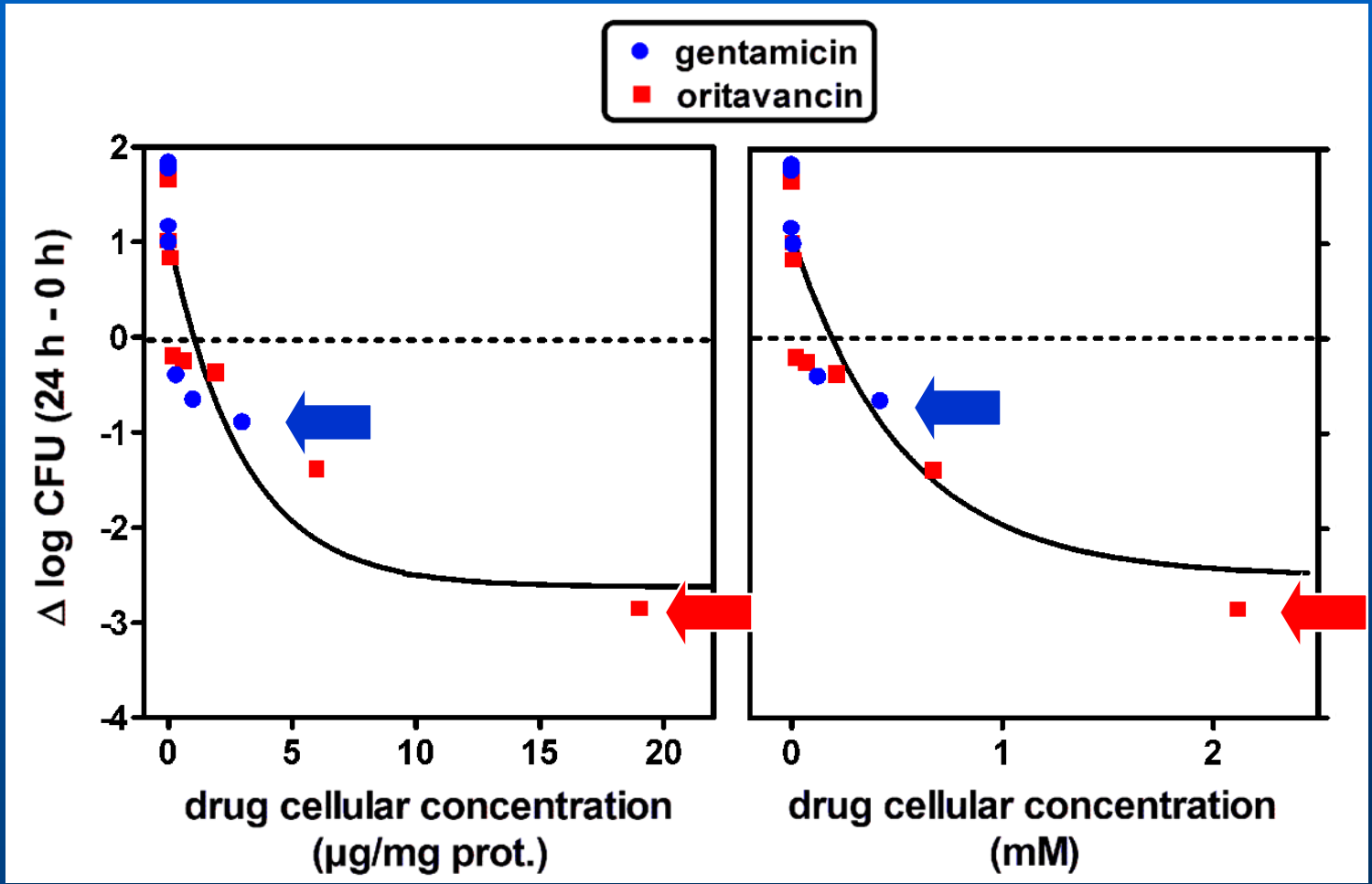
GEN and ORI are both conc.-dependent intracellularly



J774 macrophages

Lysosomotropic antibiotics and activity on *S. aureus*

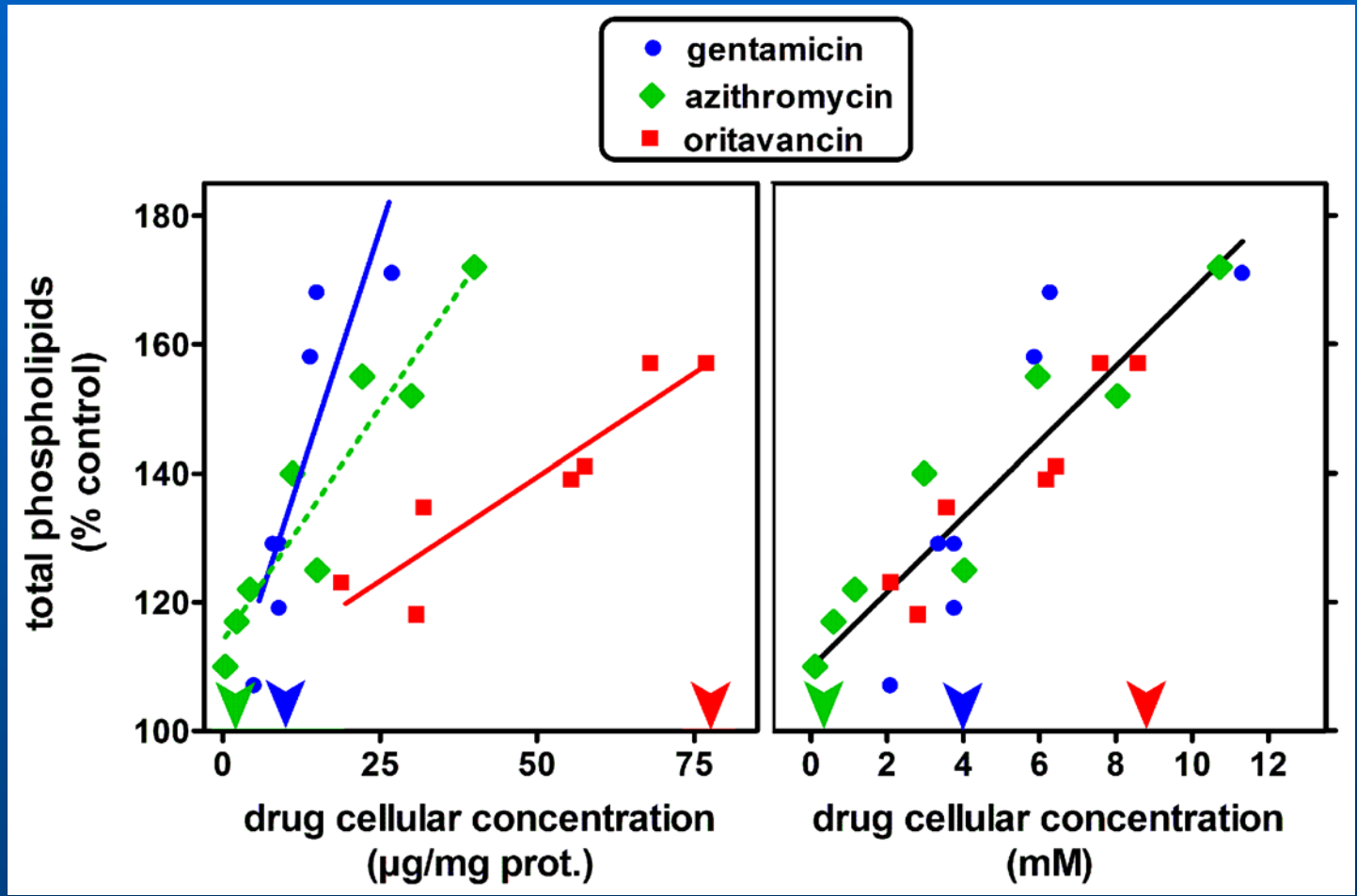
But GEN activity limited at clinically-relevant conditions



J774 macrophages

Lysosomotropic antibiotics and phospholipidosis

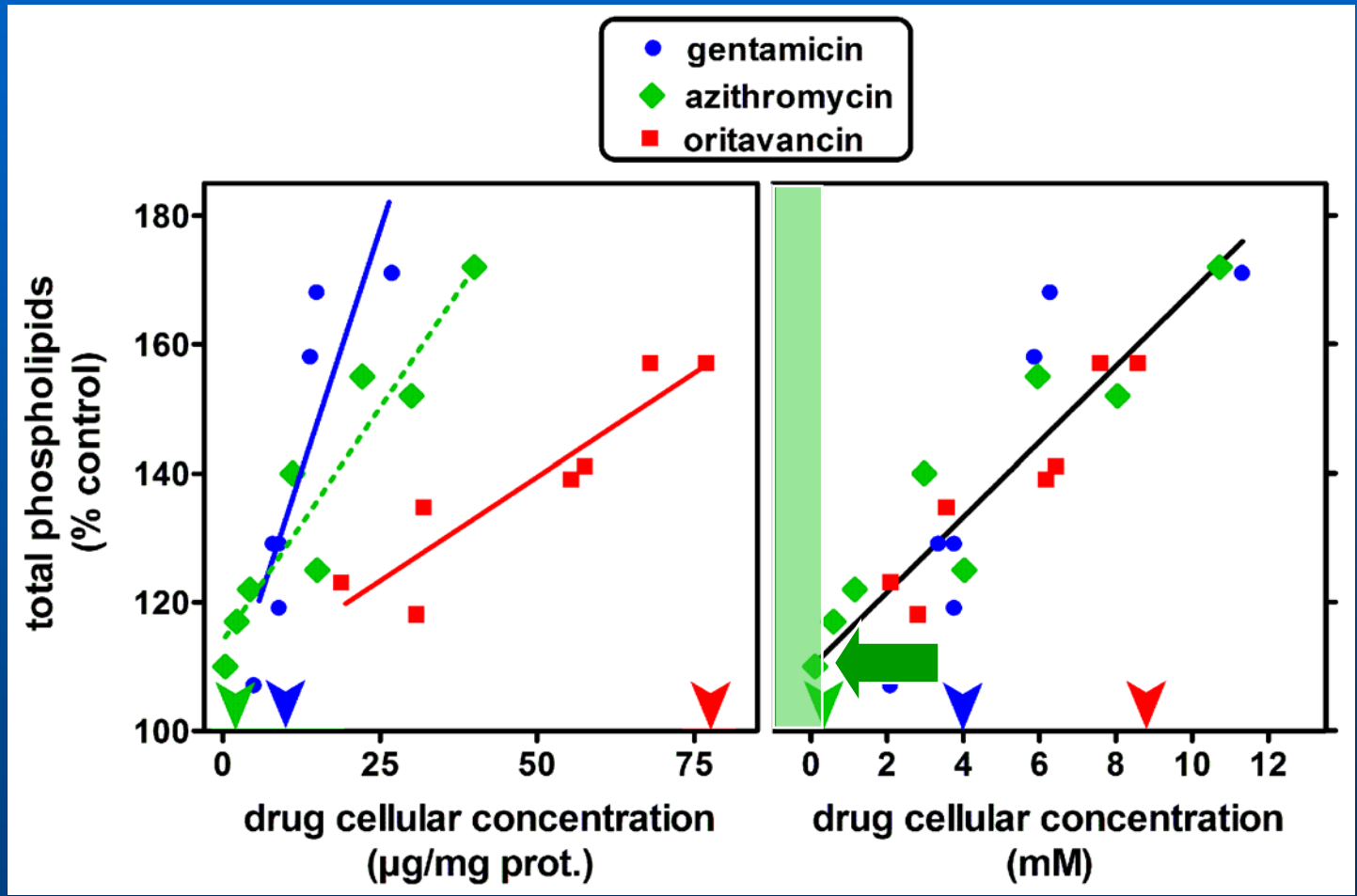
Phospholipidosis developing on a conc.-dependent manner



Rat embryo fibroblasts

Lysosomotropic antibiotics and phospholipidosis

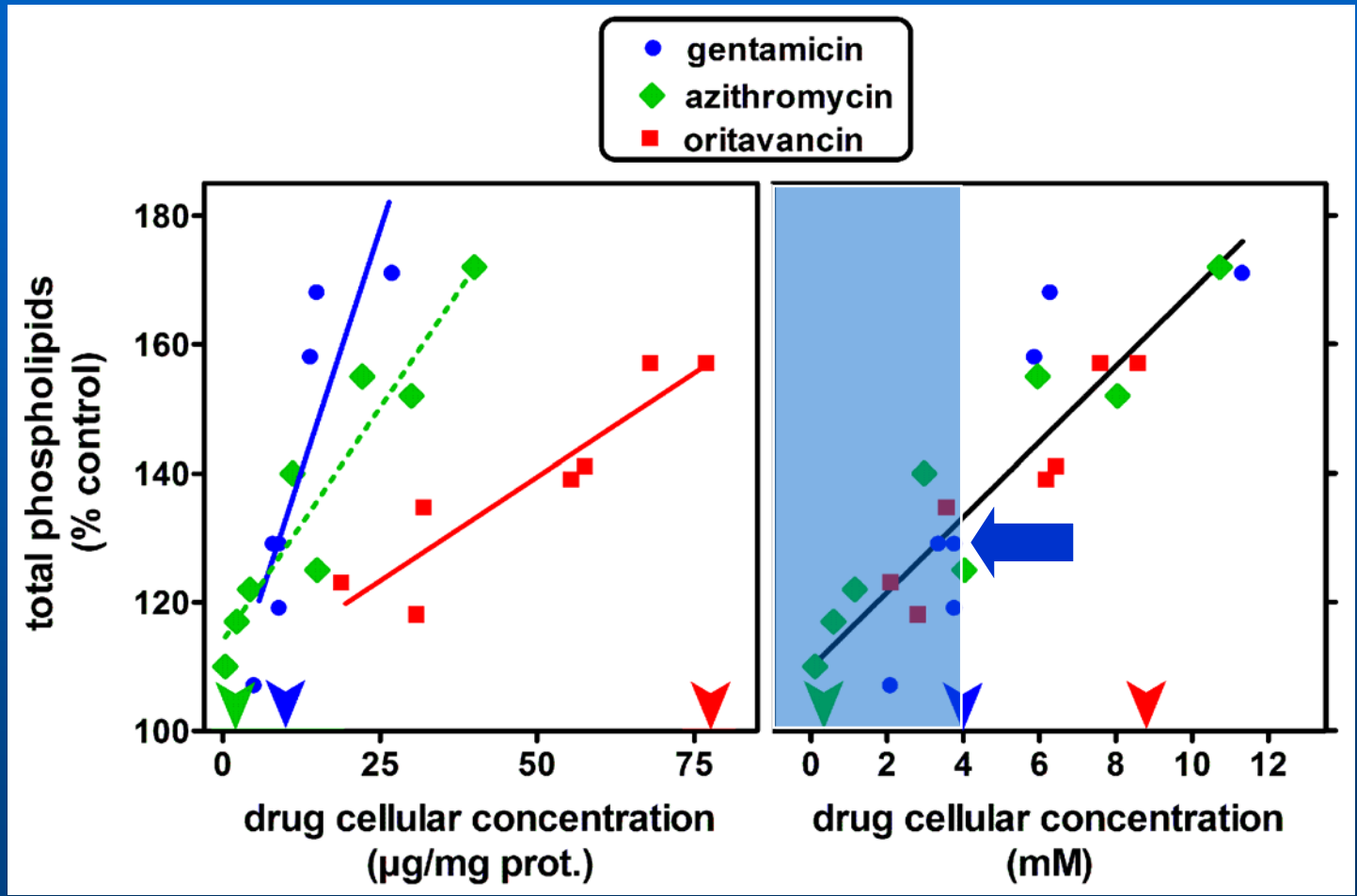
Toxic potential variable at clinically-relevant conditions



Rat embryo fibroblasts

Lysosomotropic antibiotics and phospholipidosis

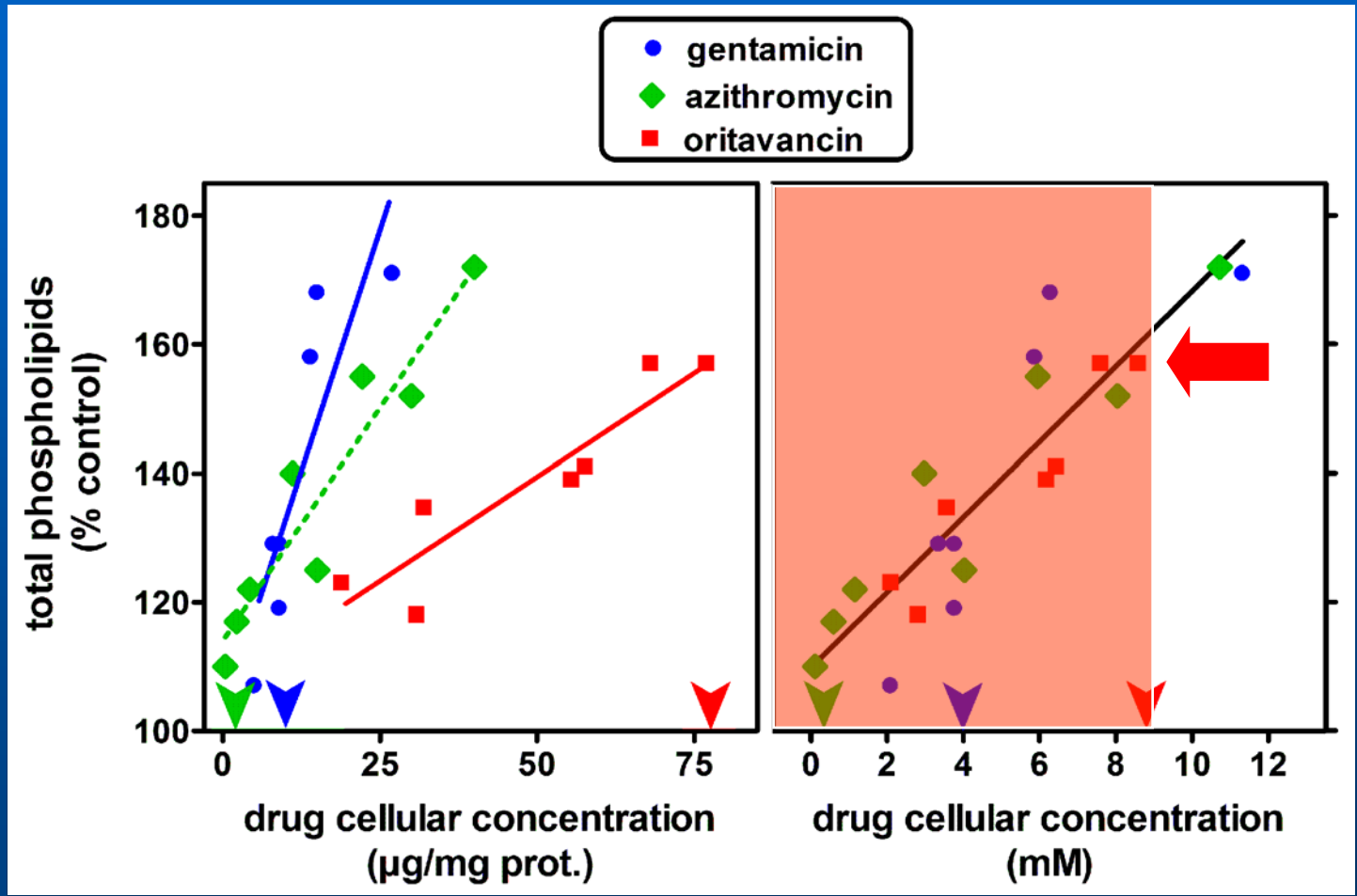
Toxic potential variable at clinically-relevant conditions



Rat embryo fibroblasts

Lysosomotropic antibiotics and phospholipidosis

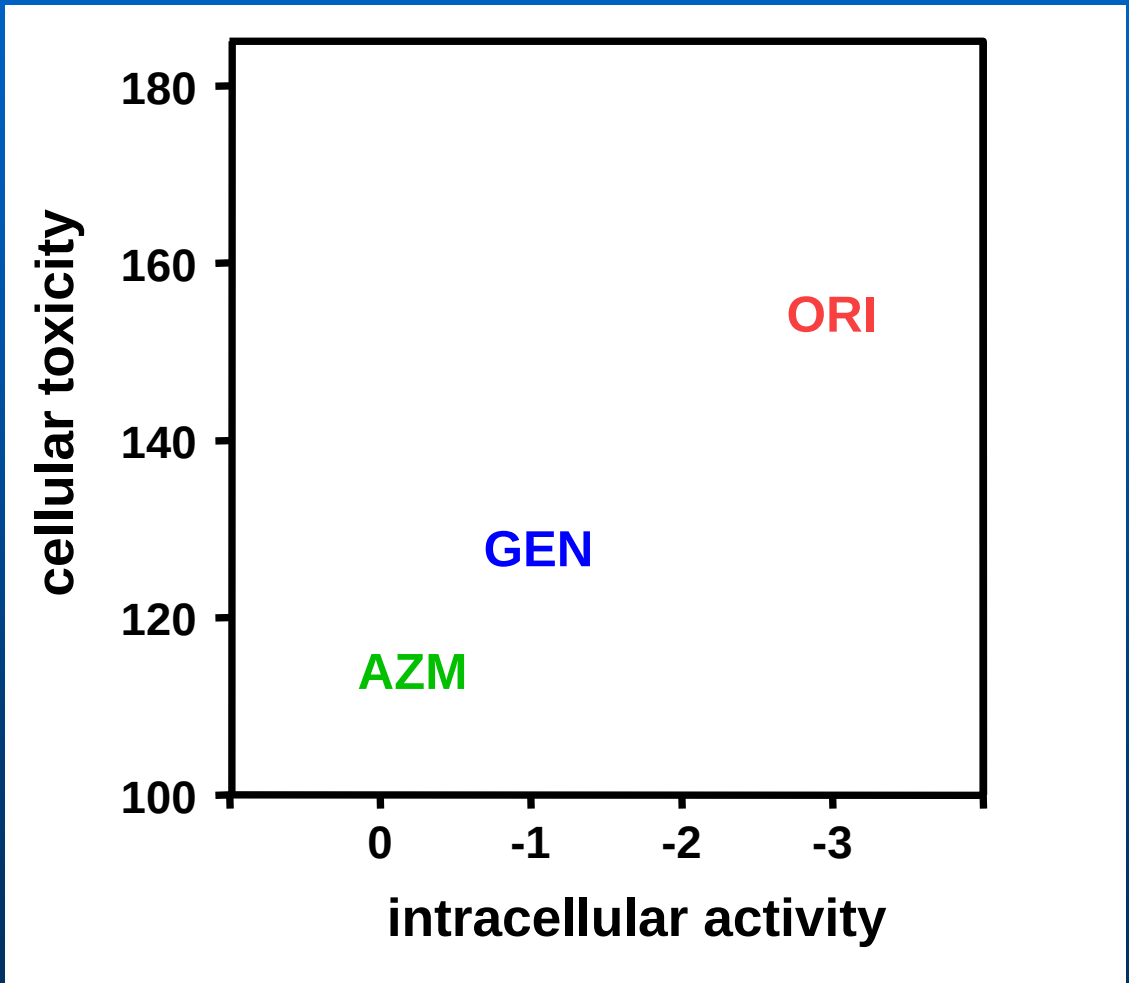
Toxic potential variable at clinically-relevant conditions



Rat embryo fibroblasts

Can we dissociate activity from toxicity ?

➔both processes are dependent on cellular concentration …



… and develop in parallel

Conclusion

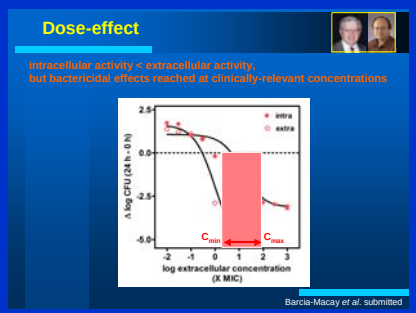
amphiphilic glycopeptides, a new type of « magic bullets»

pharmacodynamics:
bactericidal, conc-dep. activity

pharmacokinetics:
lysosomotropic accumulation

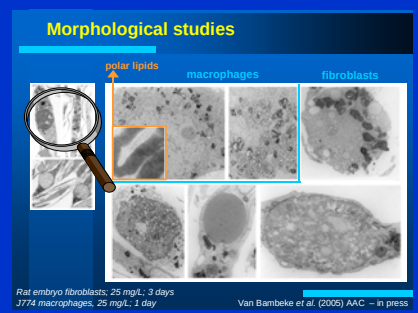
cellular pharmacodynamics:

conc. and time-dependent bactericidal activity towards extra AND intra *S. aureus*



cellular toxicity:

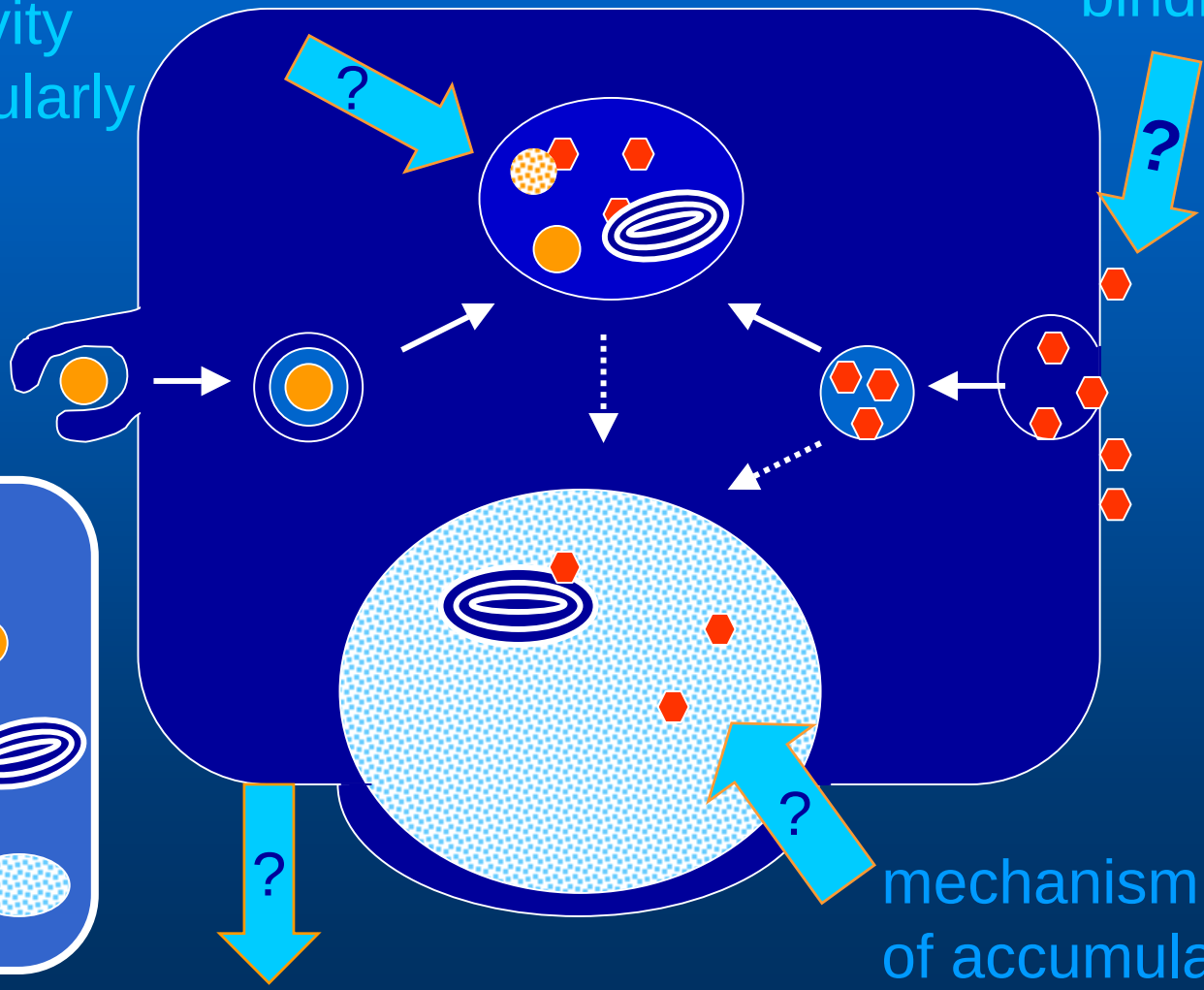
conc. and time-dependent cellular toxicity



Questions for future work

reasons for reduction in activity intracellularly

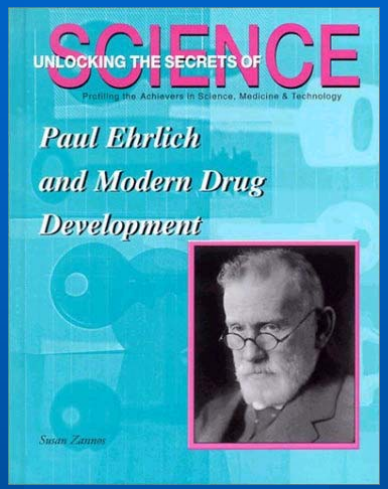
binding site



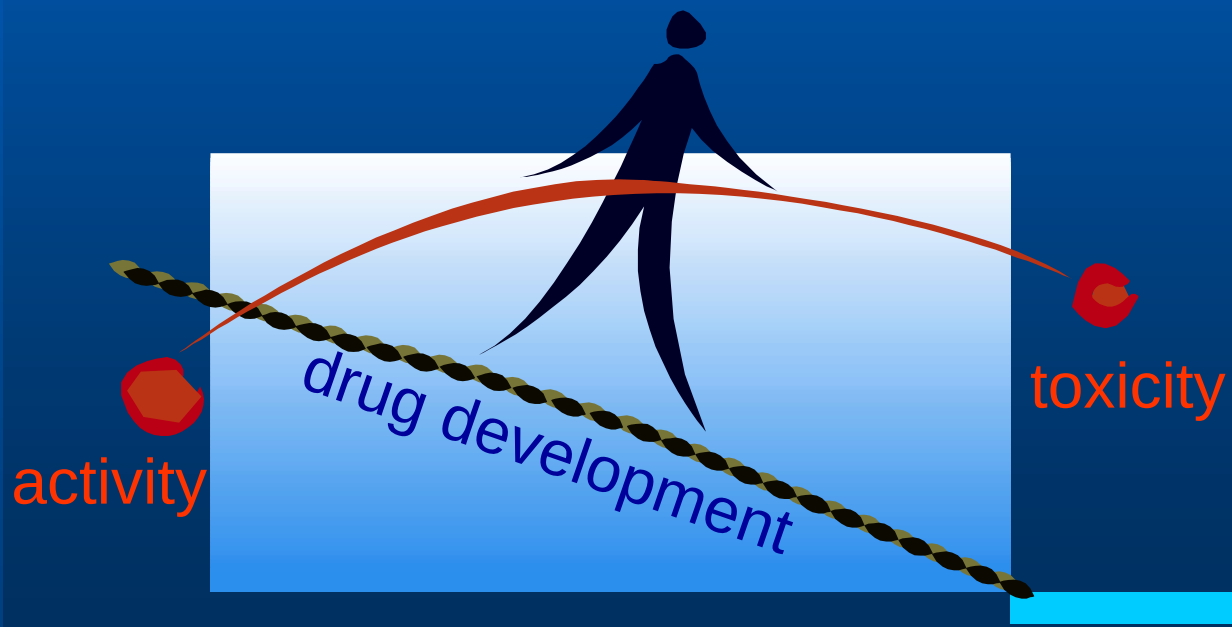
in vivo

mechanism of accumulation

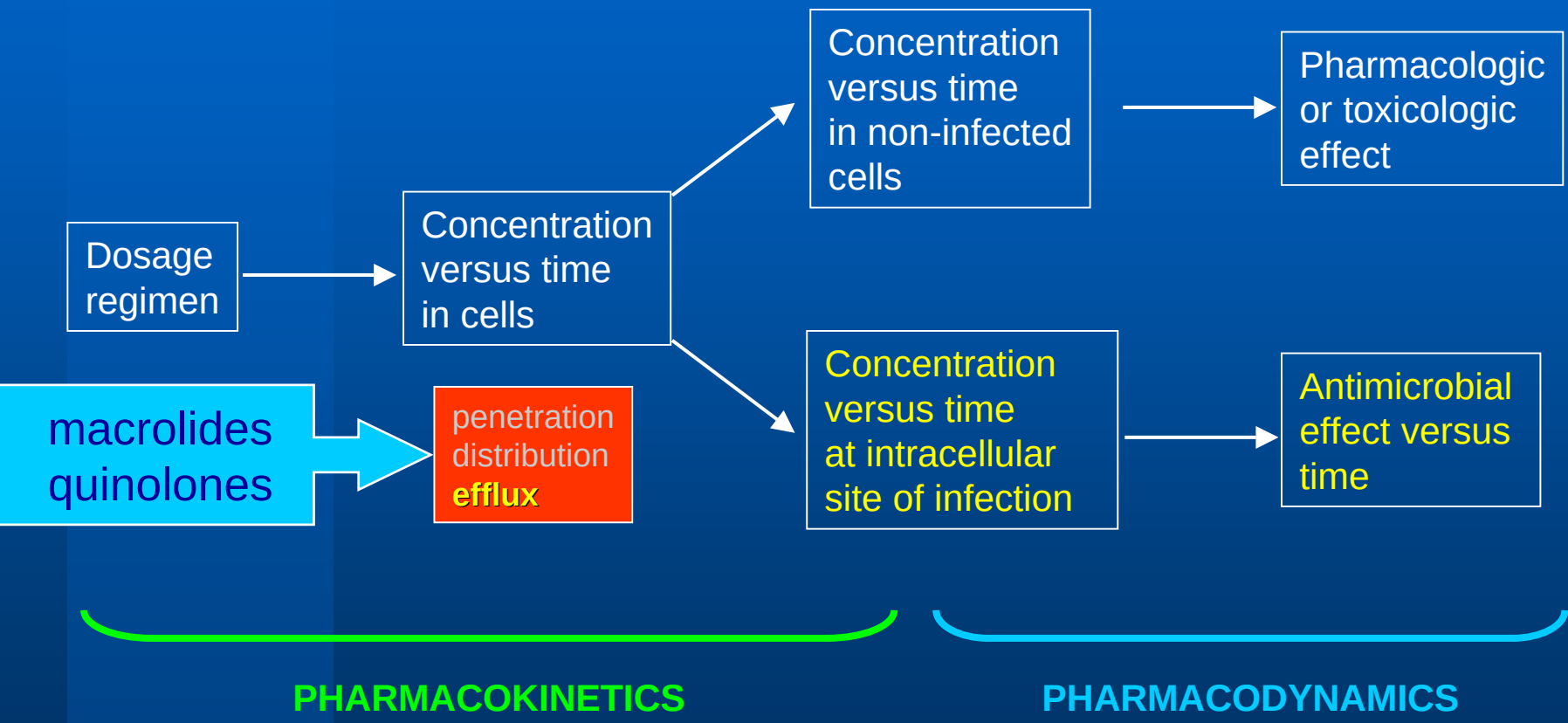
Take home message



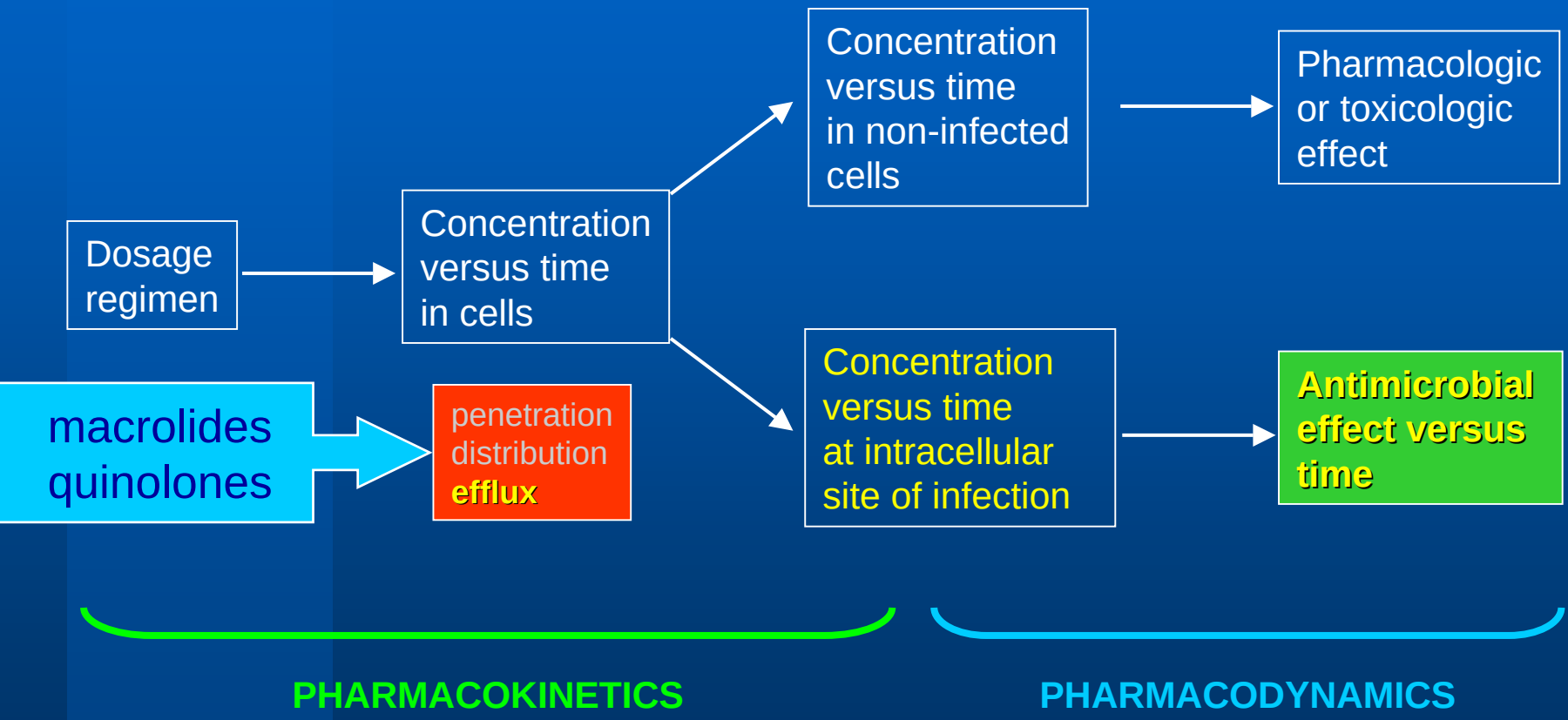
cellular accumulation,
the best and the worse of properties…



Intracellular “PK-PD”

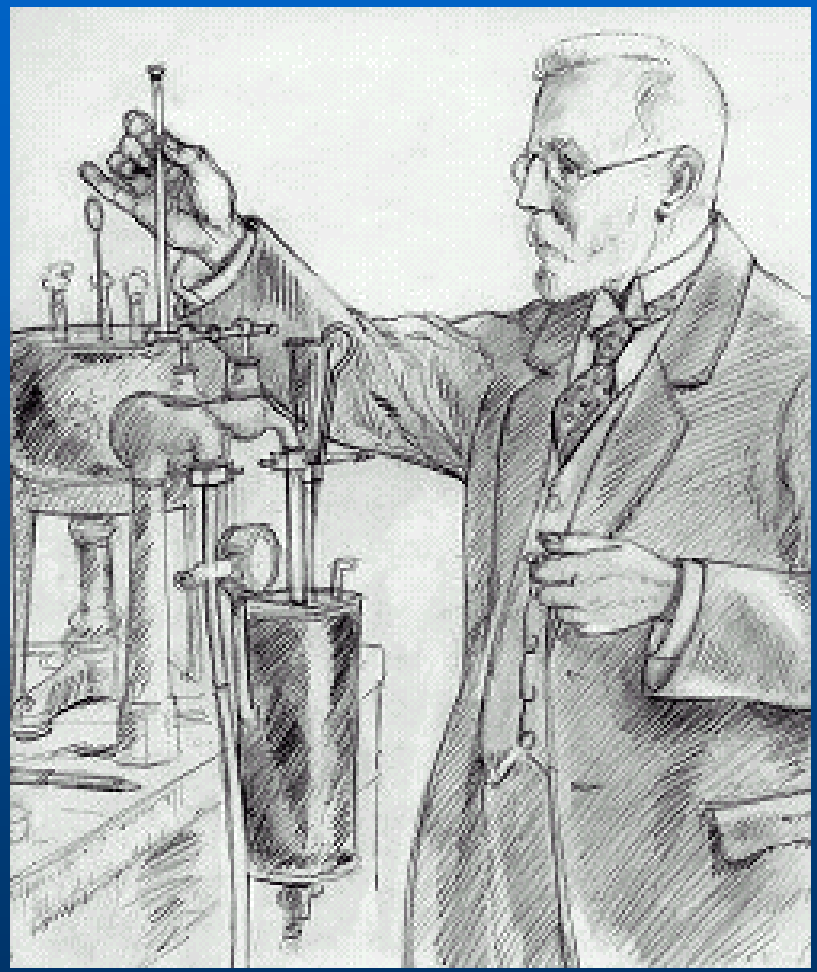


Intracellular “PK-PD”



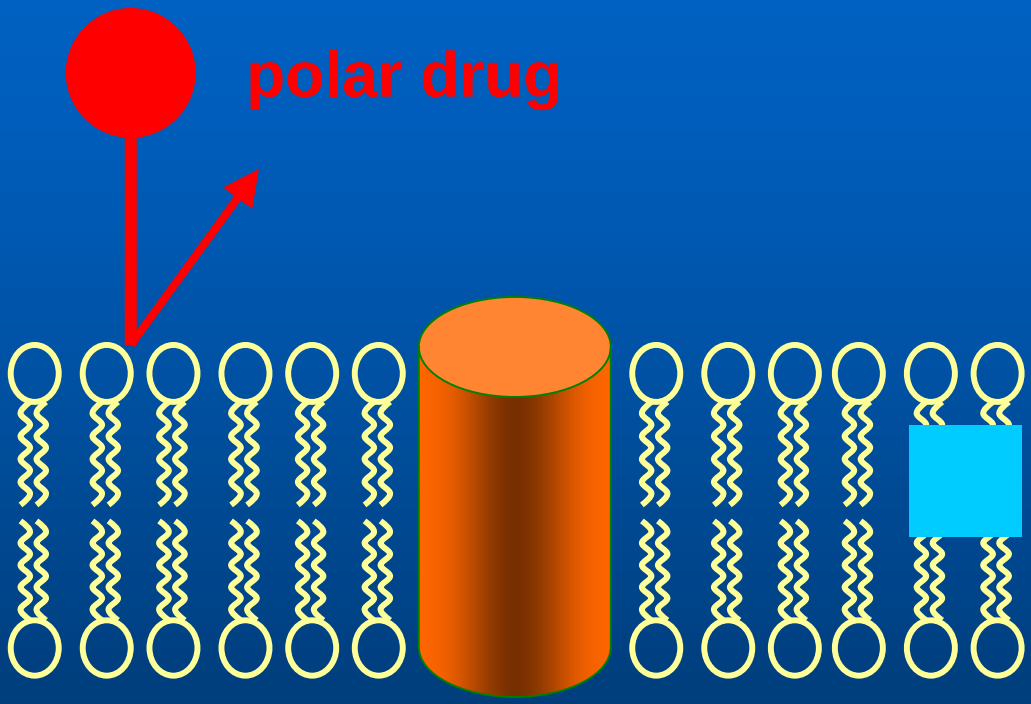
Efflux of magic bullets

from eukaryotic cells



macrolides
quinolones

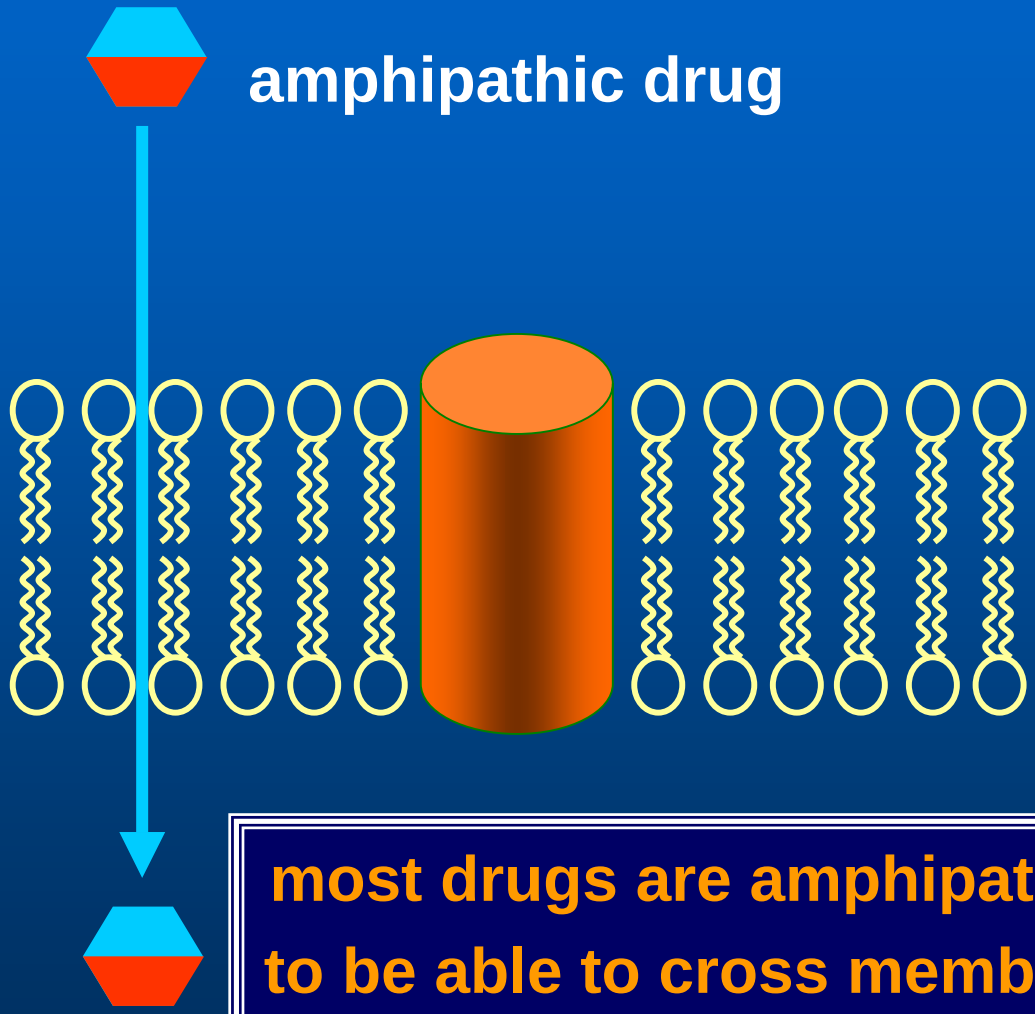
Why efflux transporters ?



physico-chemical properties are inadequate for reaching an intracellular target !

Van Bambeke *et al*. (2000) Biochem. Pharmacol. 60:457-70

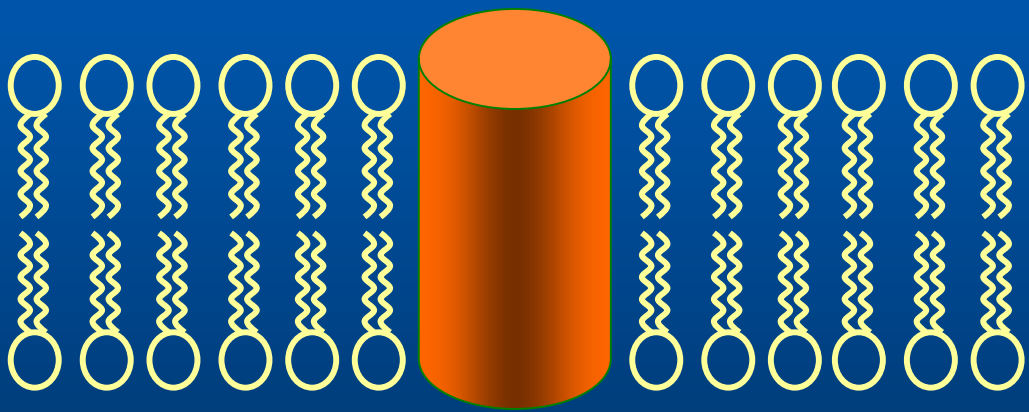
Why efflux transporters ?



most drugs are amphipathic by design, to be able to cross membrane barriers !

Van Bambeke *et al*. (2000) Biochem. Pharmacol. 60:457-70

Why efflux transporters ?



But a diffusible compound

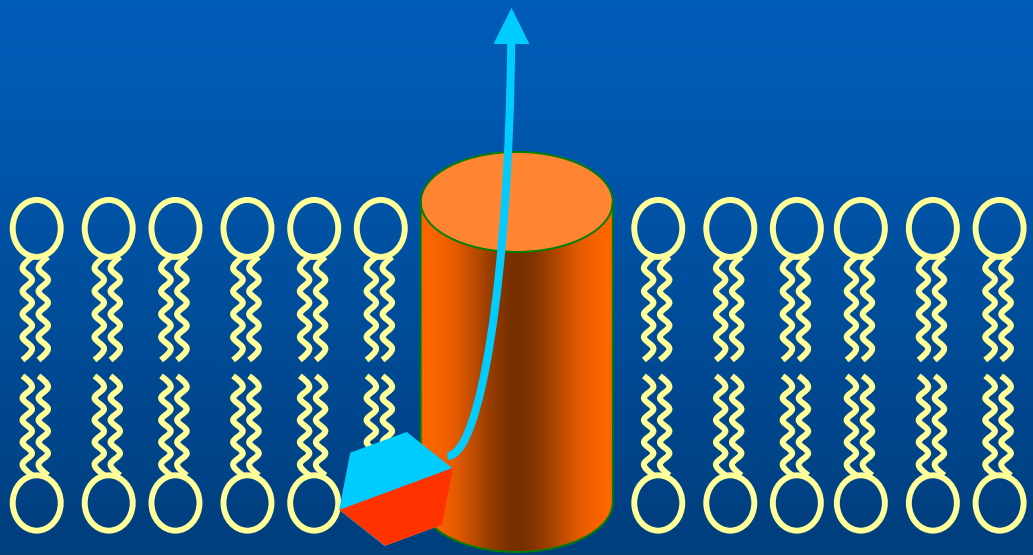
may have

potentially harmful effects !

Van Bambeke *et al*. (2000) Biochem. Pharmacol. 60:457-70

Why efflux transporters ?

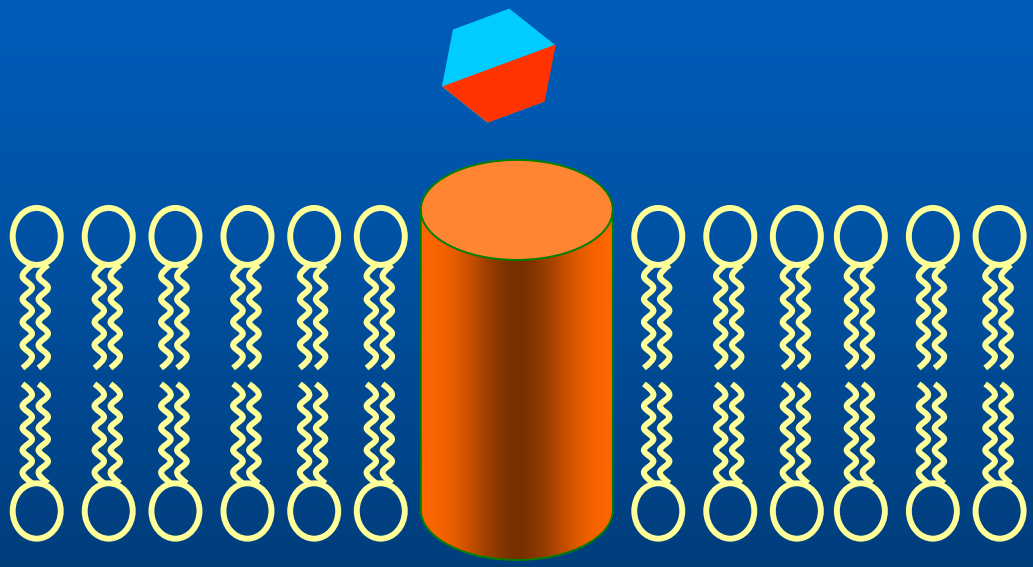
Extrusion by efflux pumps



Van Bambeke *et al*. (2000) Biochem. Pharmacol. 60:457-70

Why efflux transporters ?

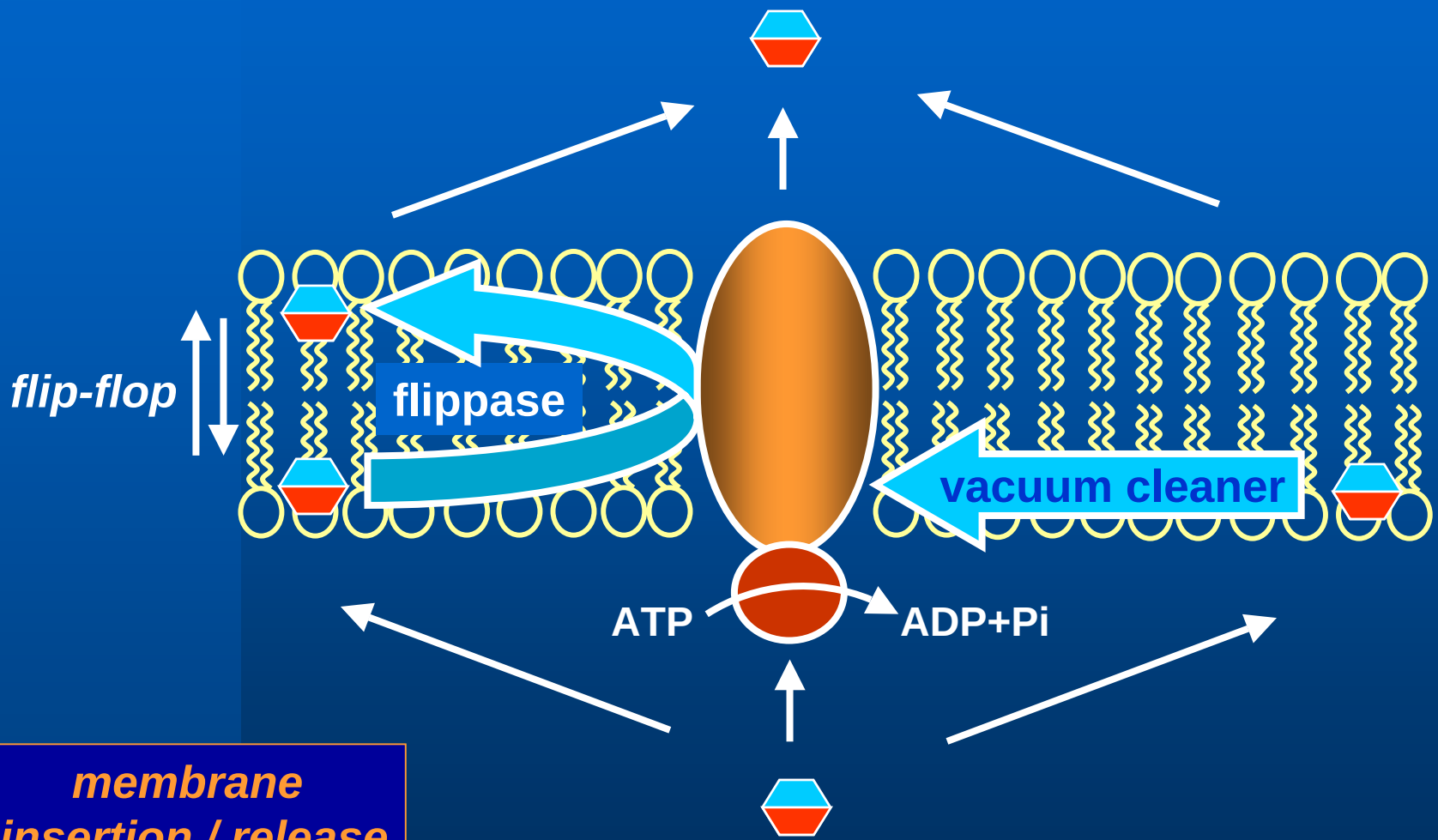
Extrusion by efflux pumps



general mean of protection
against cell invasion by diffusible molecules

Van Bambeke *et al*. (2000) Biochem. Pharmacol. 60:457-70

Mechanisms of active efflux



Van Bambeke *et al*. (2000) Biochem. Pharmacol. 60:457-70

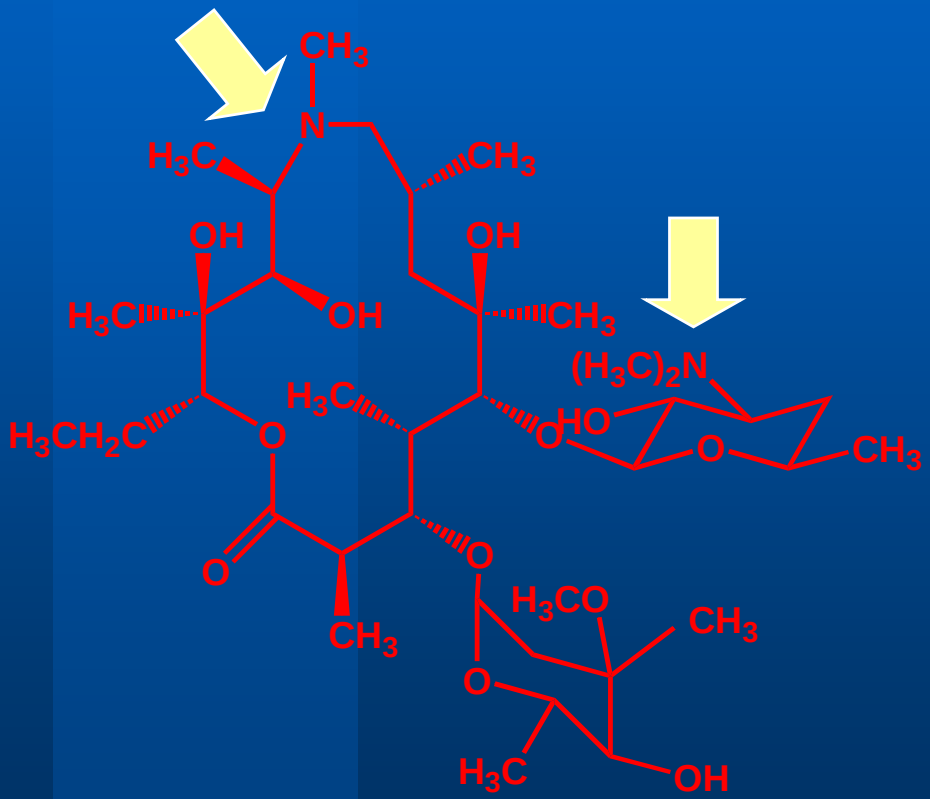
Antibiotics as substrates of efflux pumps

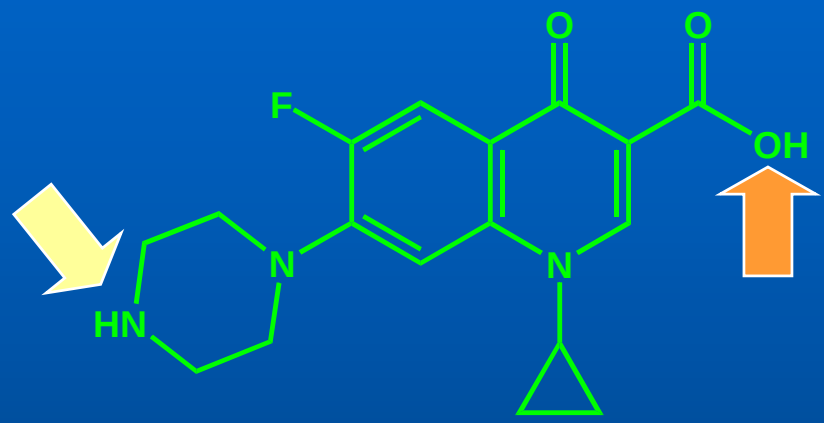
Antibiotic class	bacteria Gram (+)	bacteria Gram(-)	fungi	superior eucaryotes
β-lactams	●	●	●	●
fusidic acid		●		
macrolides	●	●	●	●
streptogramins	●			●
tetracyclines	●	●	●	●
aminoglycosides		●	●	
chloramphenicol	●	●	●	
rifamycins				●
sulfamides			●	
trimethoprim		●		
fluoroquinolones	●	●		●

Van Bambeke *et al*. (2000) Biochem. Pharmacol. 60:457-70

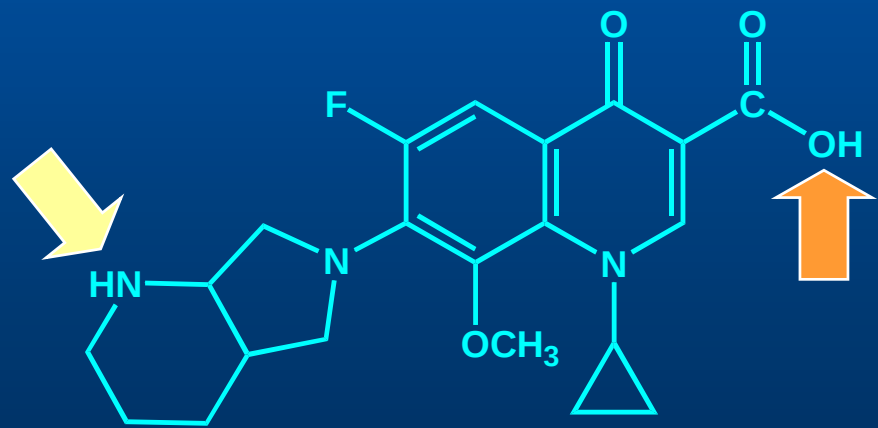
Antibiotics as substrates of efflux pumps

azithromycin





ciprofloxacin



moxifloxacin

Macrolides and quinolones as cell-associated antibiotics

Infection. 1995;23 Suppl 1:S10-4.

Clinical relevance of intracellular and extracellular concentrations of macrolides.

Carbon C.

C.H.U. Bichat-Claude Bernard, Paris, France.

The serum levels of the three macrolides--roxithromycin, clarithromycin and azithromycin--vary considerably. The prediction of the antibacterial effect against extracellular pathogens is based on circulating concentrations of free drug, peak and trough levels, the rate of killing, and the presence of a post-antibiotic effect. Intracellular activity depends on the distribution of the antibiotic and the localization of the bacteria, and is variable. Roxithromycin uptake is greater than that of erythromycin. The intracellular half-life may be long for some compounds (azithromycin > roxithromycin). The intracellular distribution is bimodal, both in the lysosomes and the cytoplasm, but the mechanisms of uptake have not yet been established. At low pH, accumulation is low and macrolides are less active in an acidic medium. Intracellular concentrations cannot readily be predicted on the basis of extracellular levels. Different models have shown that the greater the intracellular concentration, the better the clinical effect. In addition, the transport of macrolides by cells into the infected focus may play an important role in the therapeutic outcome. These factors influence the clinical indications for macrolides, their dosing regimens and breakpoints. In future, macrolides will be developed that are more selective for intracellular infections, while others, which will achieve significant serum levels, will be useful for a broader range of diseases. However, new compounds should be evaluated in different models of infection before clinical studies are instituted. The analysis of failures remains the most important approach in defining concentration/effect relationships.

Infection. 1991;19 Suppl 7:S365-71.

Quinolones in the treatment of lower respiratory tract infections caused by intracellular pathogens.

Chidiac C, Mouton Y.

Department of Infectious Diseases, University of Lille II, Central Hospital, Tourcoing, France.

Intracellular pathogens are inhibited to varying degrees, depending upon the strain of the organism and the quinolone tested. Quinolones achieve levels in the lower respiratory tract that equal or exceed serum concentrations, and they also achieve good intracellular concentrations. Experimental models of intracellular infection have demonstrated the efficacy of ciprofloxacin, difloxacin, fleroxacin, ofloxacin and pefloxacin. Animal models of experimental legionellosis have confirmed in vivo their efficacy in this field. Thus, quinolones appear to be a safe and efficacious alternative treatment in lower respiratory tract infection (LRTI) due to intracellular pathogens. Considering the in vitro and experimental studies, quinolones should play an important role in the treatment of LRTI caused by intracellular pathogens, and prospective controlled studies are strongly recommended.

Aim of the study

amphiphilic antibiotics

- accumulating in eucaryotic cells
- considered as useful for treating intracellular infections
- known substrates of efflux pumps in bacteria

efflux from macrophages ?

macrolides | **quinolones**

- phenotypic characterization of the active efflux
- consequences for intracellular activity

Aim of the study

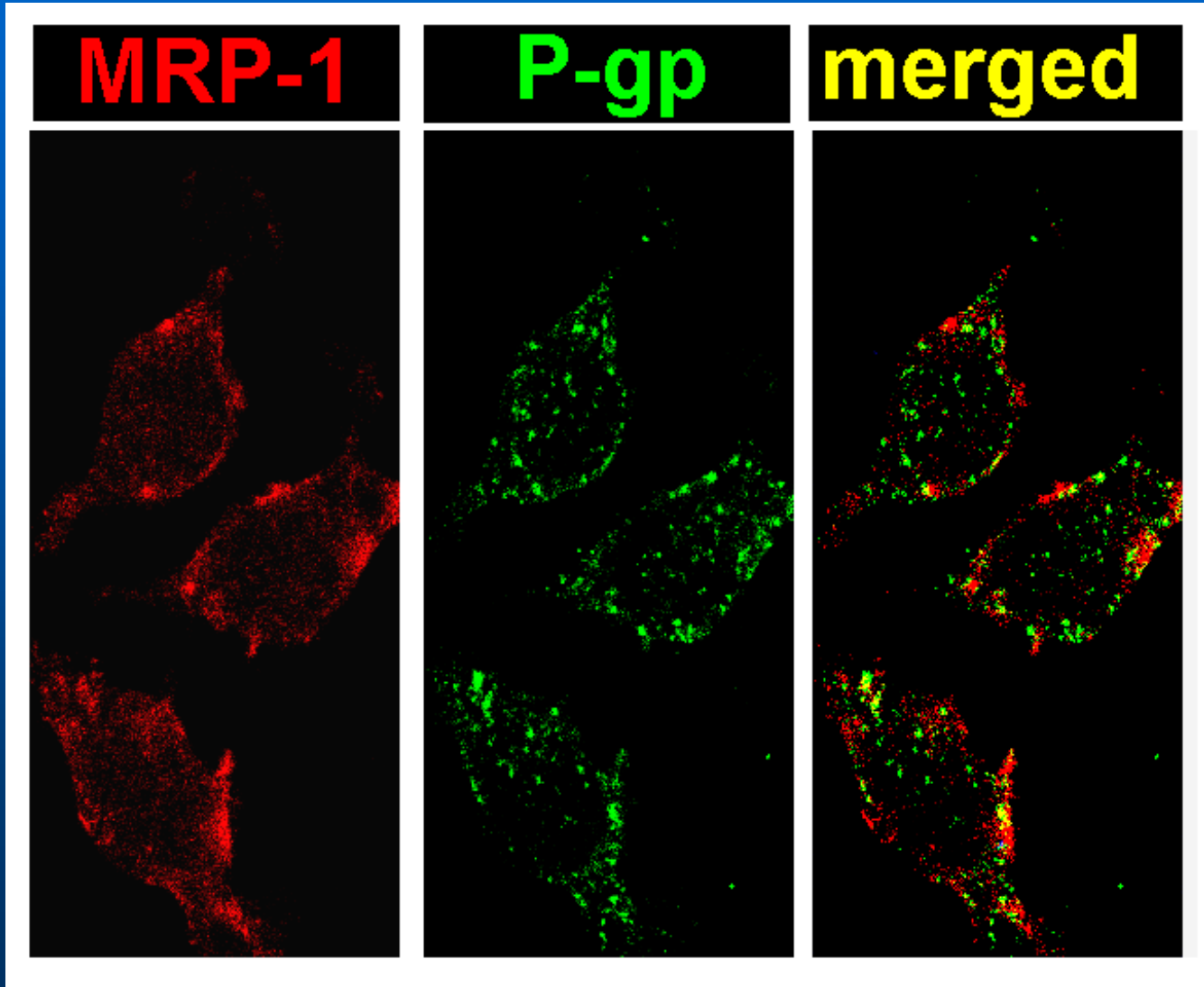
amphiphilic antibiotics
- accumulating in eucaryotic cells
- considered as useful for treating intracellular infections
- known substrates of efflux pumps in bacteria

efflux from macrophages ?

macrolides | **quinolones**

- **phenotypic characterization of the active efflux**
- consequences for intracellular activity

Efflux pumps expressed in J774 macrophages



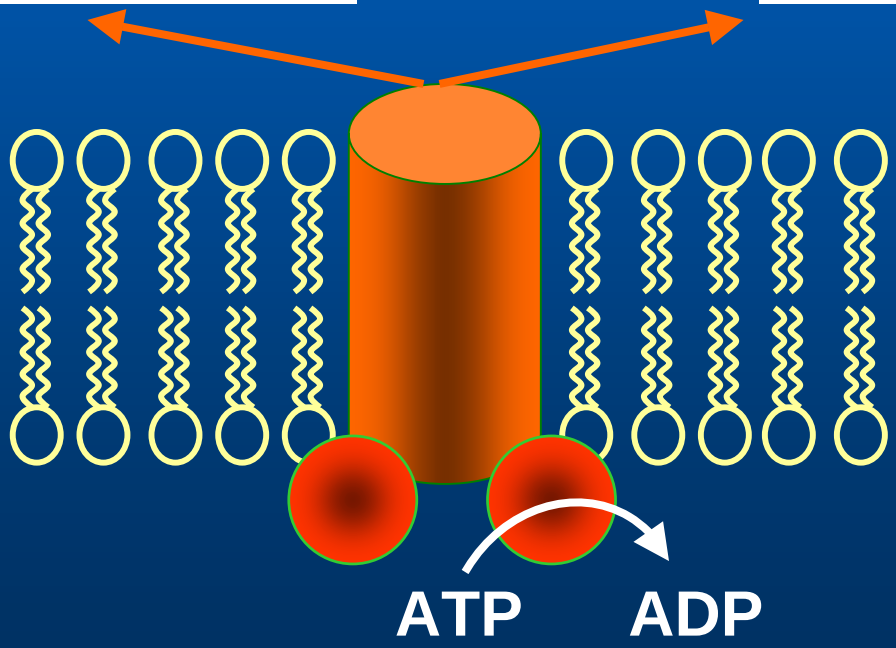
ABC multidrug transporters

cationic amphiphiles

anionic amphiphiles

MDR-1 (P-glycoprotein)

MRP1-10



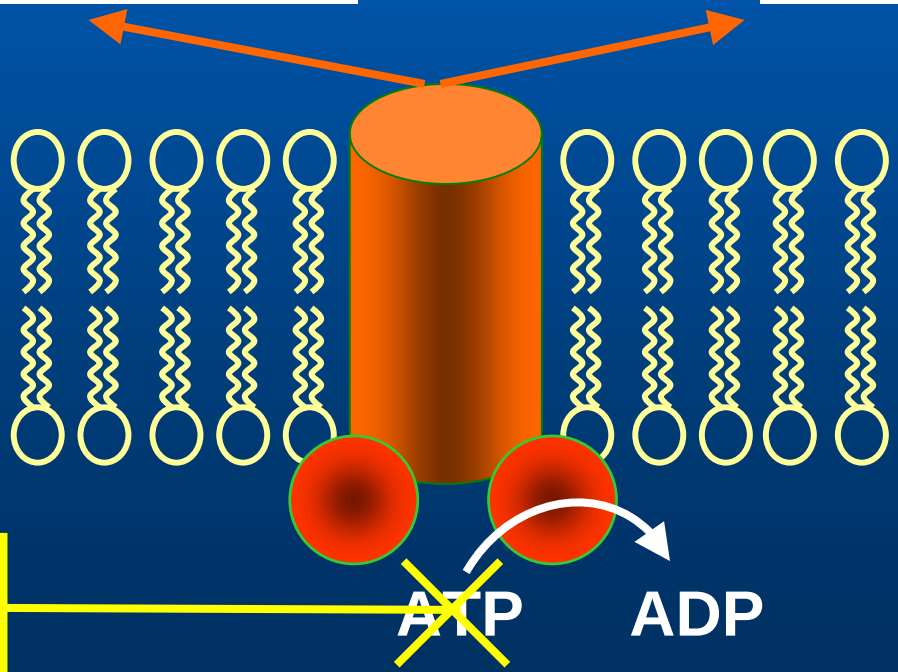
How to inhibit ABC transporters ?

cationic amphiphiles

anionic amphiphiles

MDR-1 (P-glycoprotein)

MRP1-10



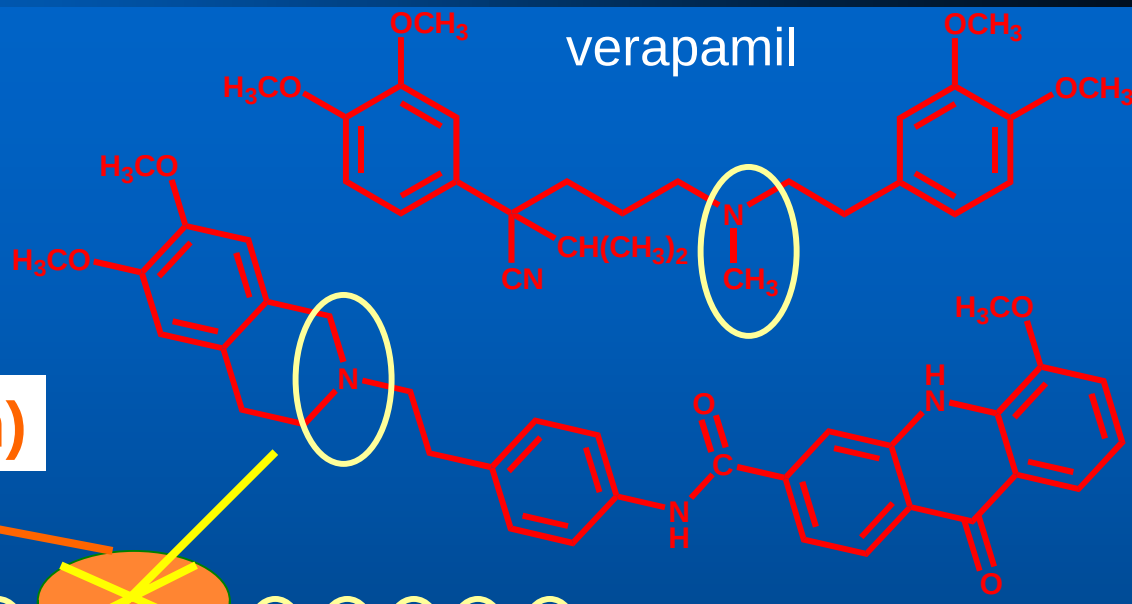
deoxyglucose
NaN_3

How to inhibit ABC transporters ?

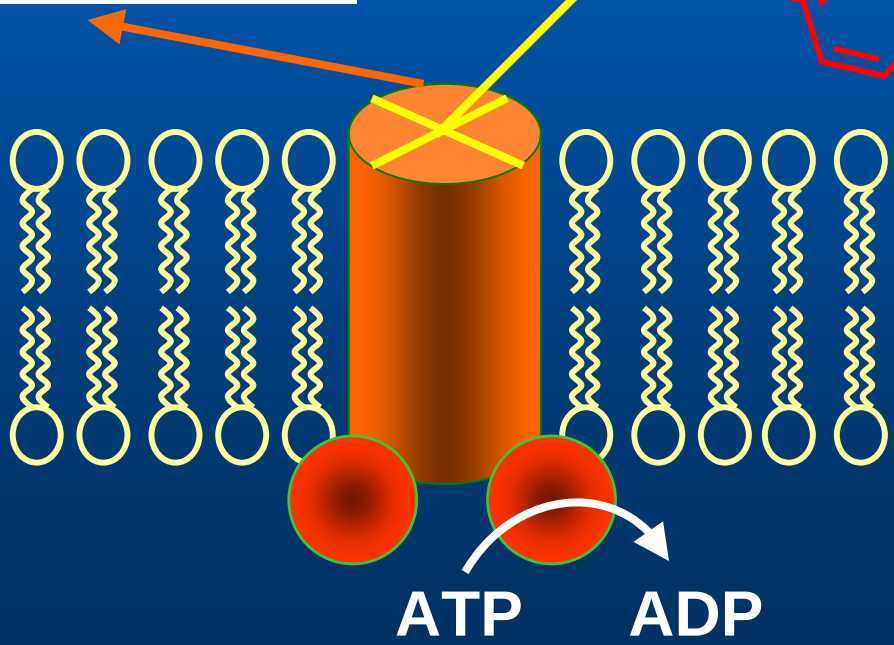
cationic amphiphiles

MDR-1 (P-glycoprotein)

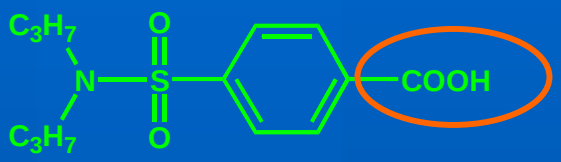
verapamil



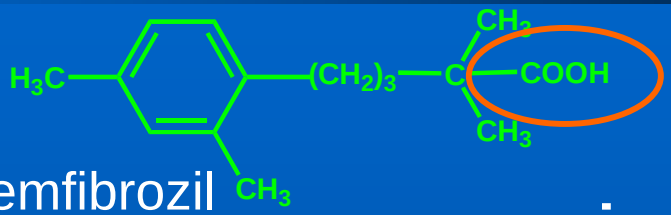
GF120918



How to inhibit ABC transporters ?

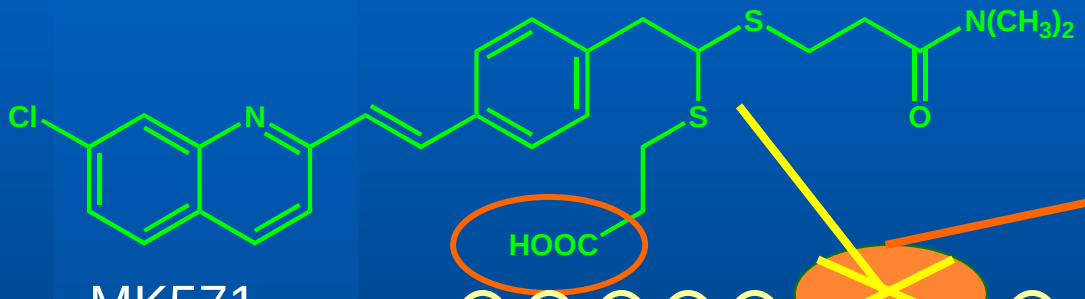


probenecid



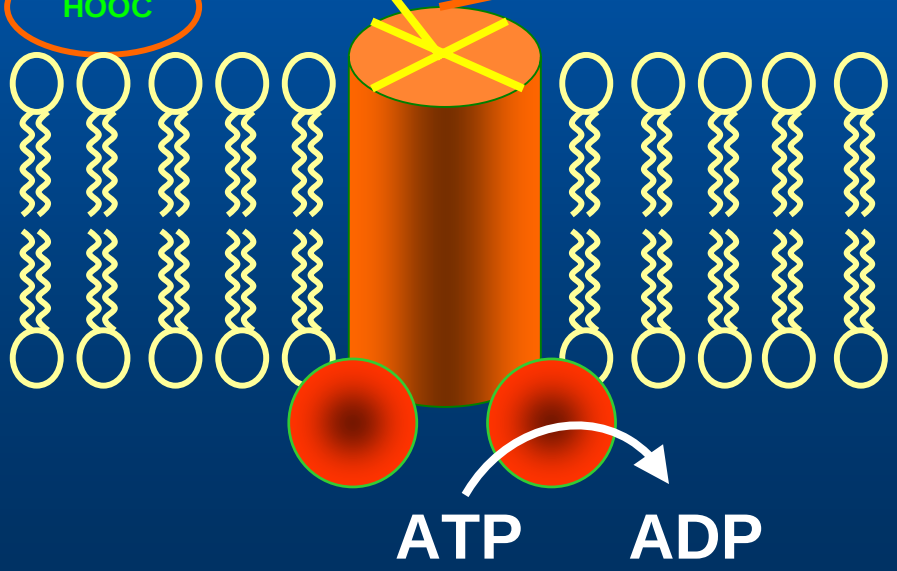
gemfibrozil

anionic amphiphiles



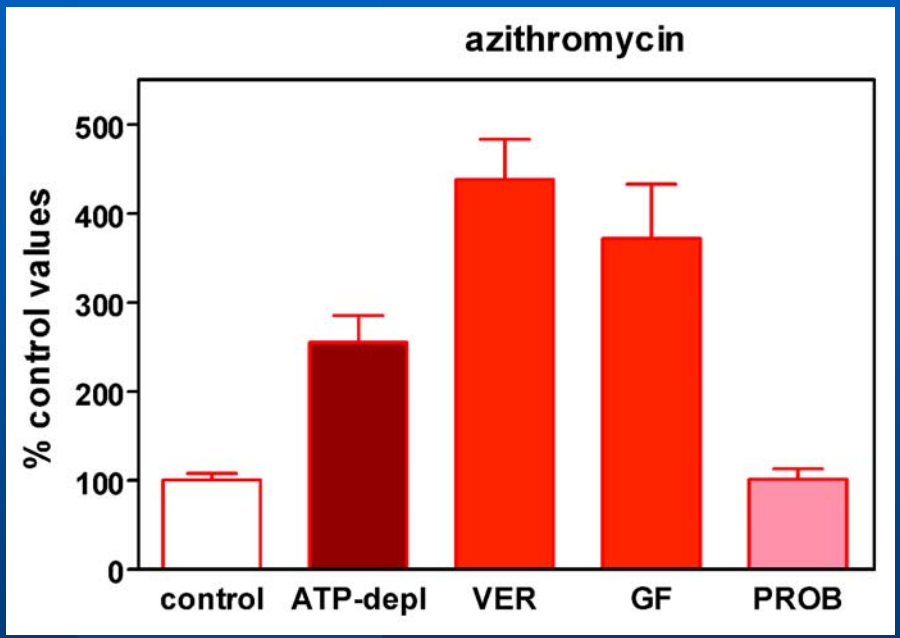
MK571

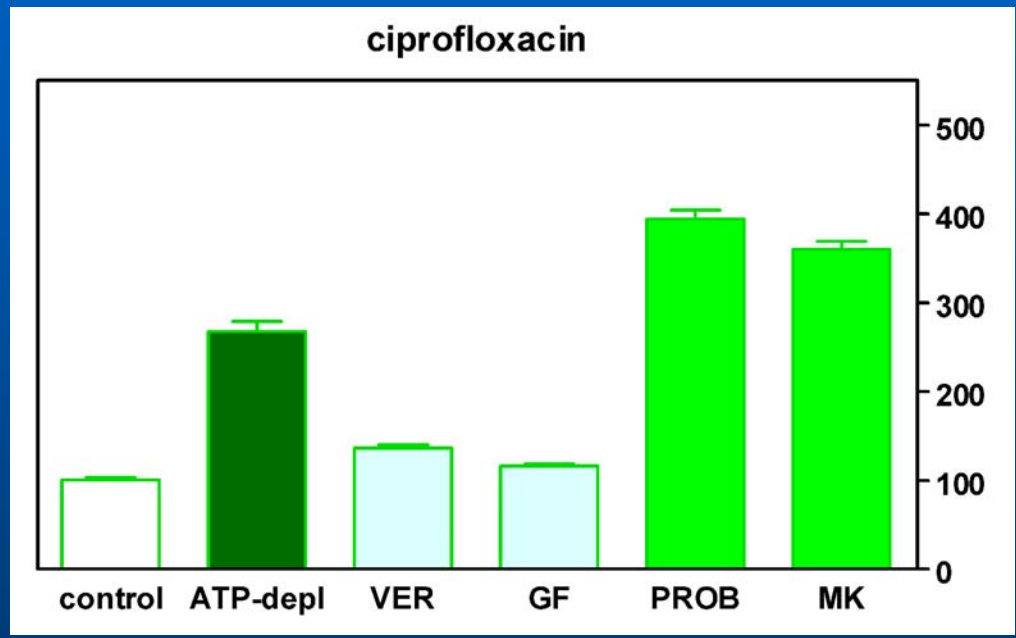
MRP1-10



Differential recognition by MDR pumps

Influence of ATP-depletion and pump inhibitors on accumulation at equilibrium





azithromycin & P-glycoprotein

ciprofloxacin & MRP

extracell. conc. 5 mg/L; AZM 3 h; CIP 2 h

Michot *et al*. AAC (2004) 48:2673-82

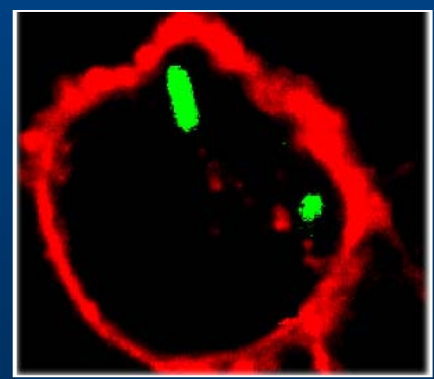
Aim of the study

amphiphilic antibiotics
- accumulating in eucaryotic cells
- considered as useful for treating intracellular infections
- known substrates of efflux pumps in bacteria

efflux from macrophages ?

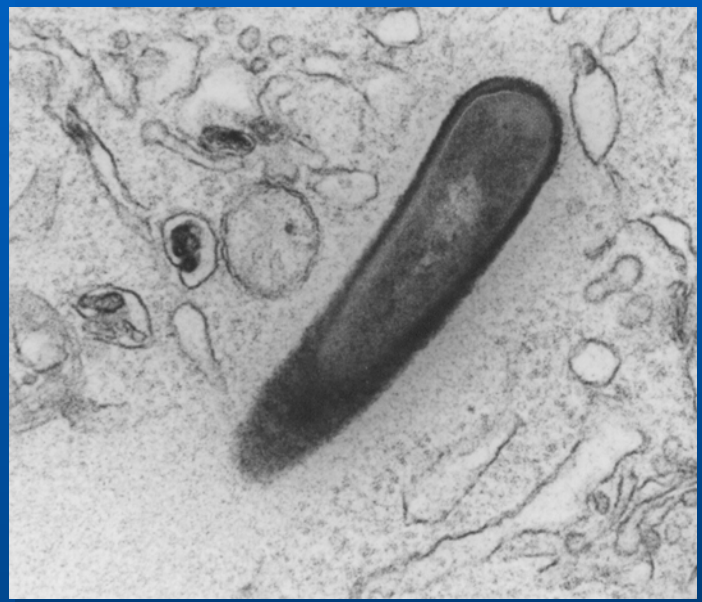
macrolides | **quinolones**

- phenotypic characterization of the active efflux
- **consequences for intracellular activity**



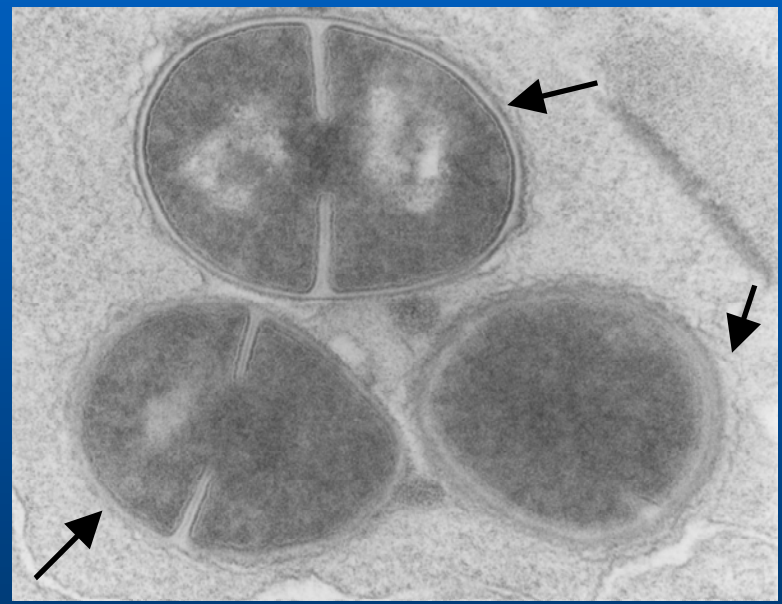
Models of intracellular infection

L. monocytogenes



cytosol

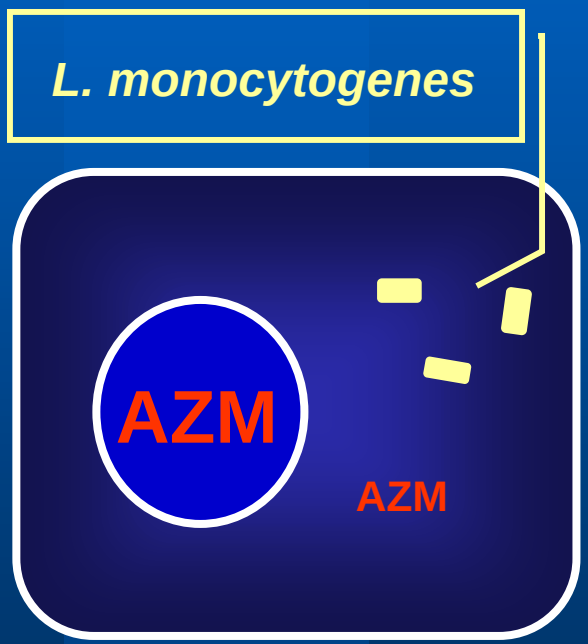
S. aureus

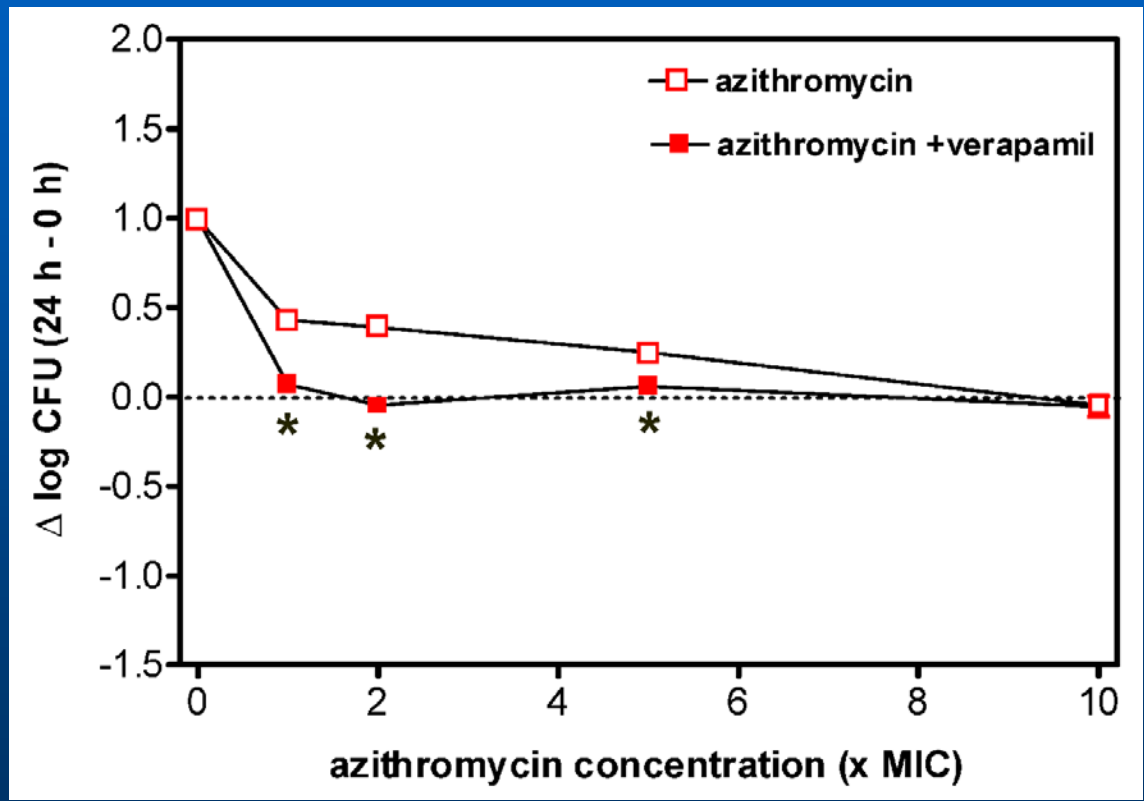


phagolysosomes

Influence of pump inhibitors on intracellular activity

azithromycin and *L. monocytogenes*



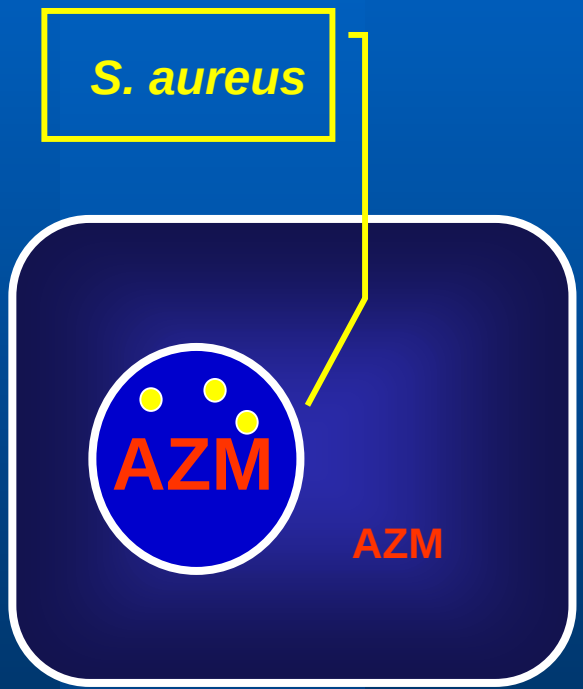


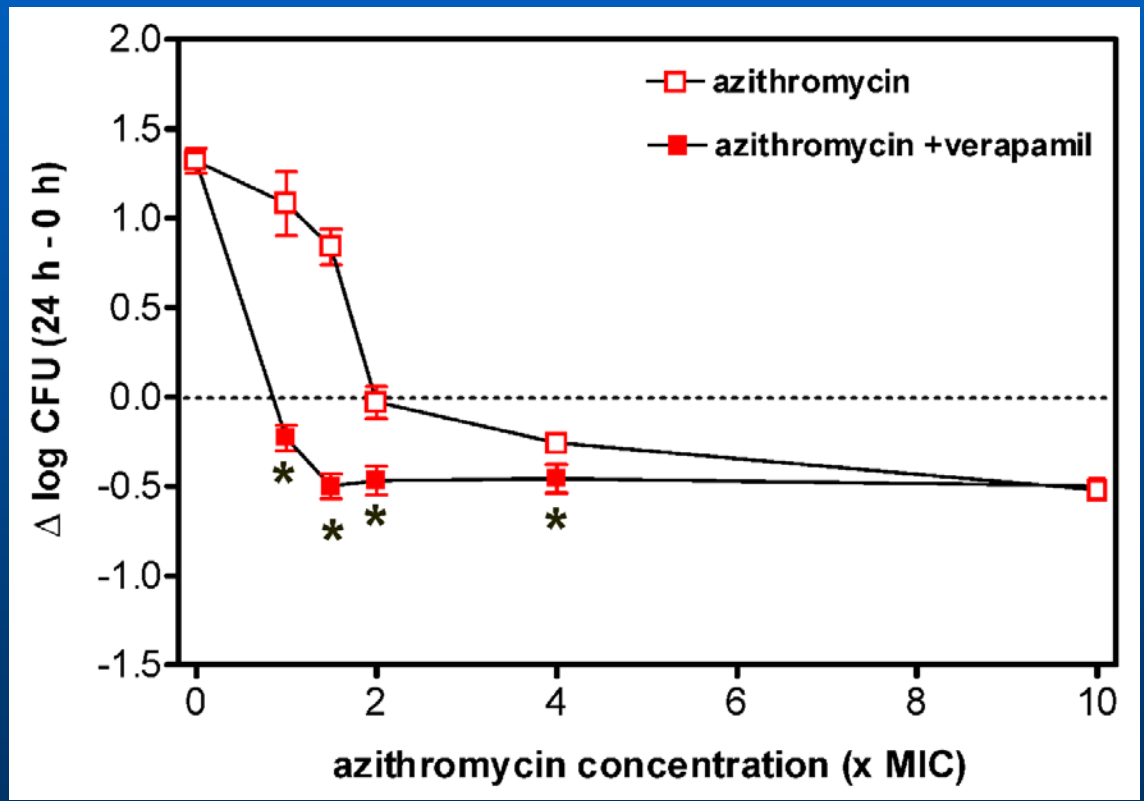
verapamil 20 µM; 24 h

Seral *et al* (2003) JAC 51:1167-73

Influence of pump inhibitors on intracellular activity

azithromycin and *S. aureus*



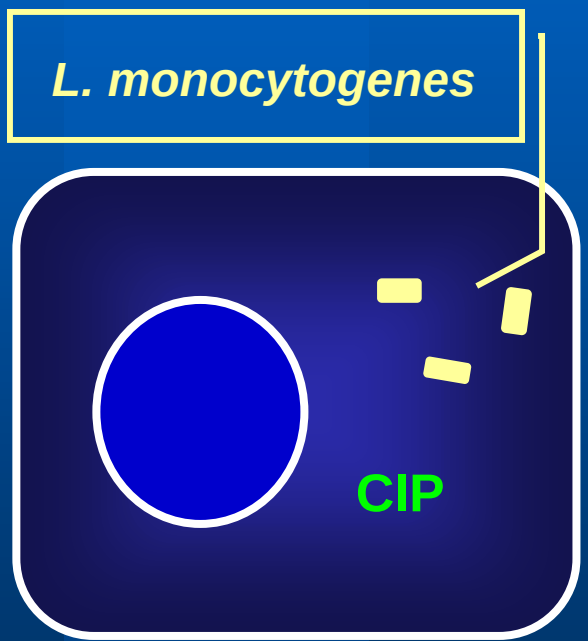


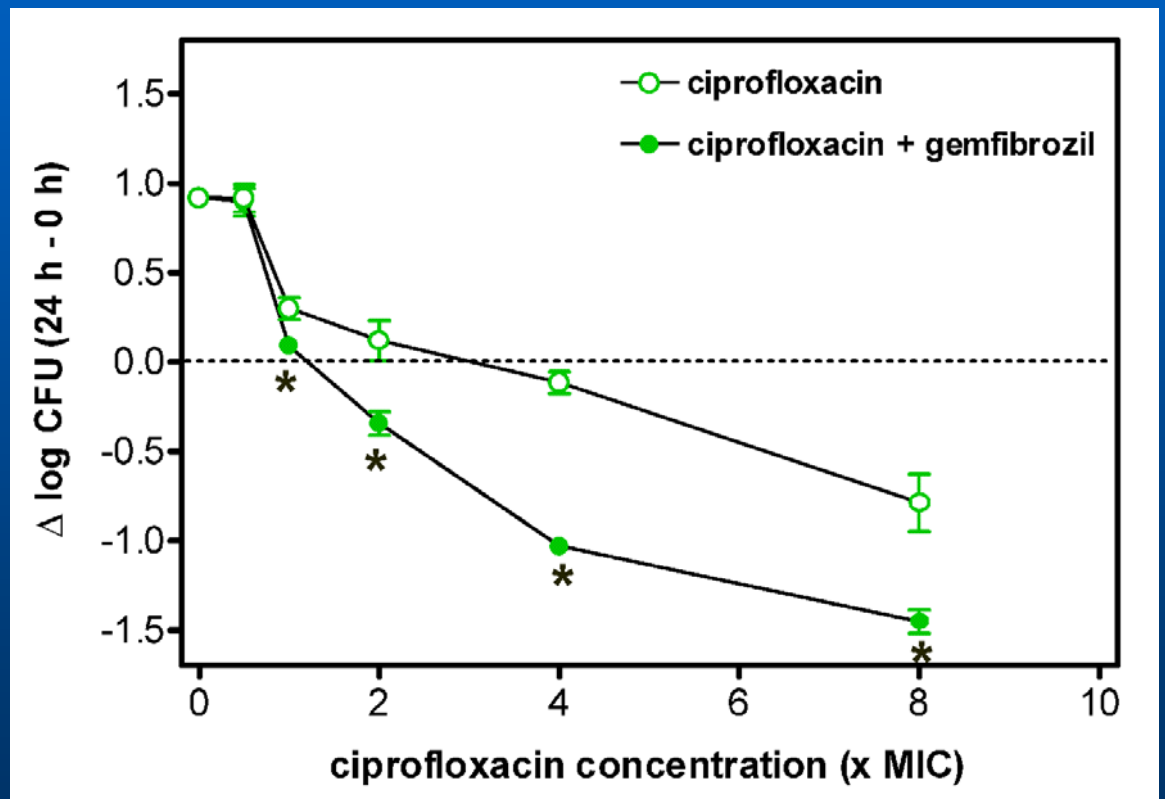
verapamil 20 µM; 24 h

Seral *et al* (2003) JAC 51:1167-73

Influence of pump inhibitors on intracellular activity

ciprofloxacin and *L. monocytogenes*



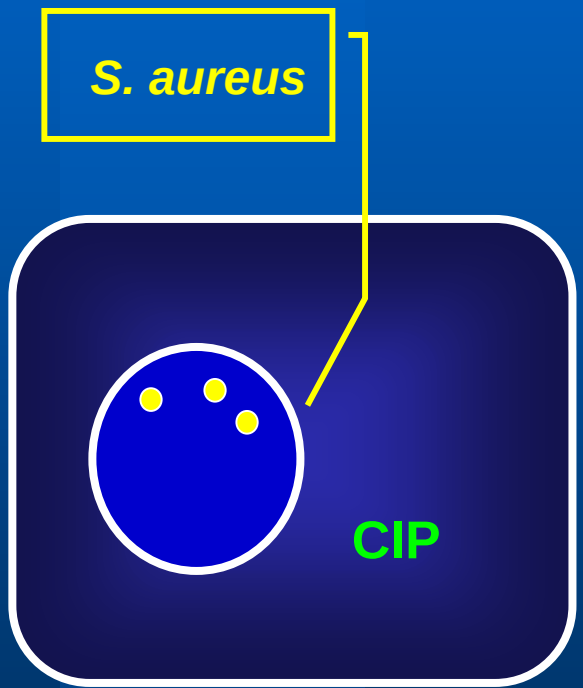


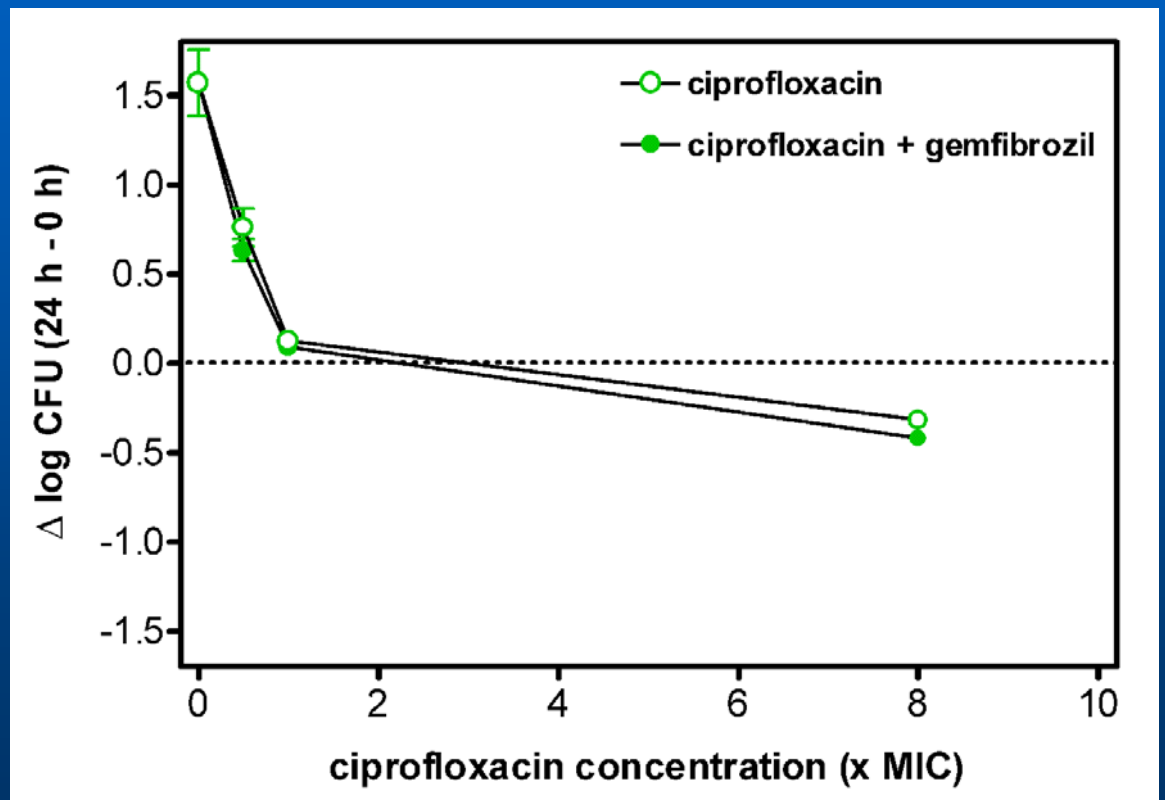
gemfibrozil 250 µM; 24 h

Seral *et al* (2003) JAC 51:1167-73

Influence of pump inhibitors on intracellular activity

ciprofloxacin and *S. aureus*



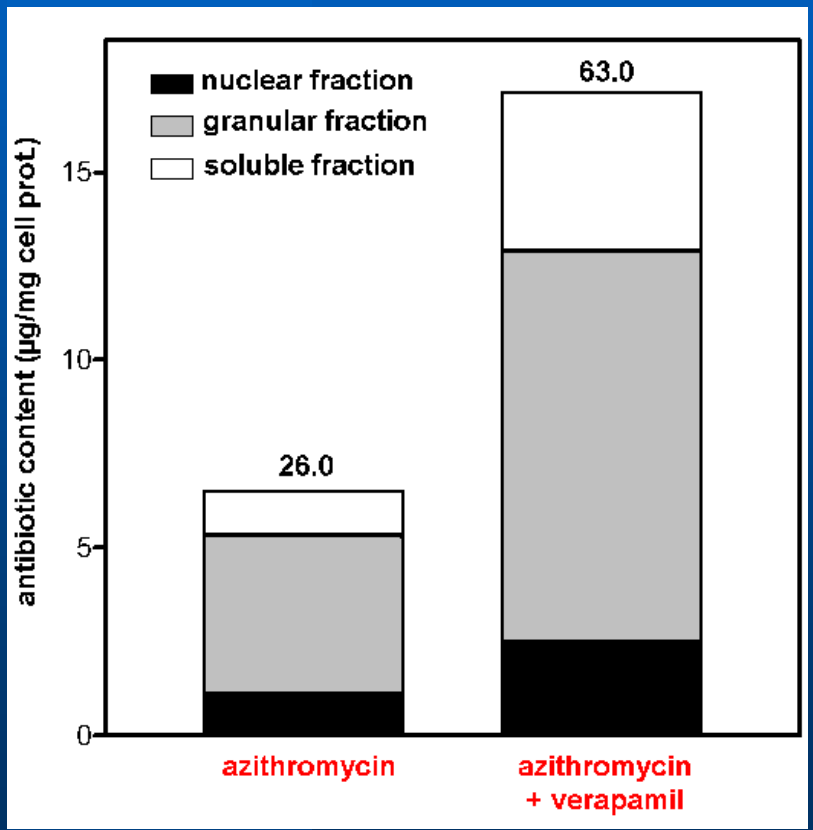


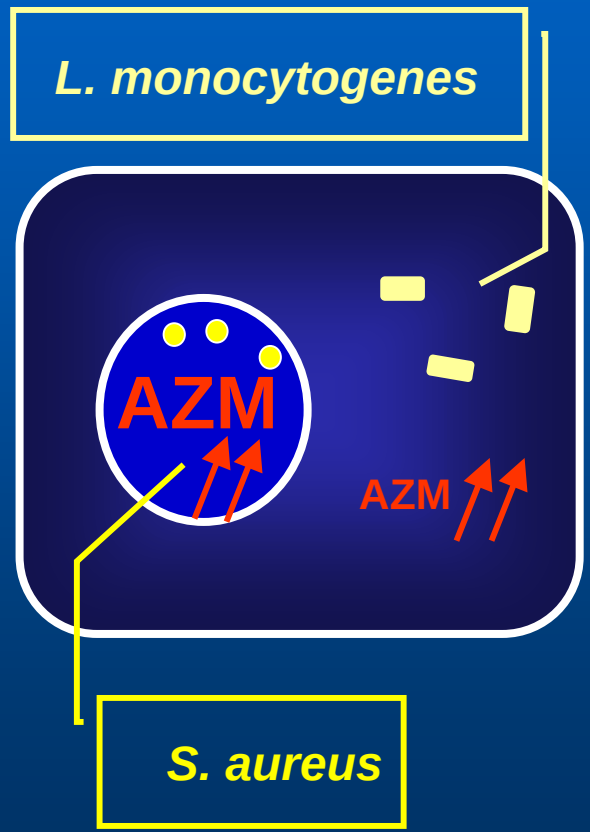
gemfibrozil 250 µM; 24 h

Seral *et al* (2003) JAC 51:1167-73

Influence of pump inhibitors on antibiotic distribution

verapamil enhances azithromycin concentration In cytosol and vacuoles

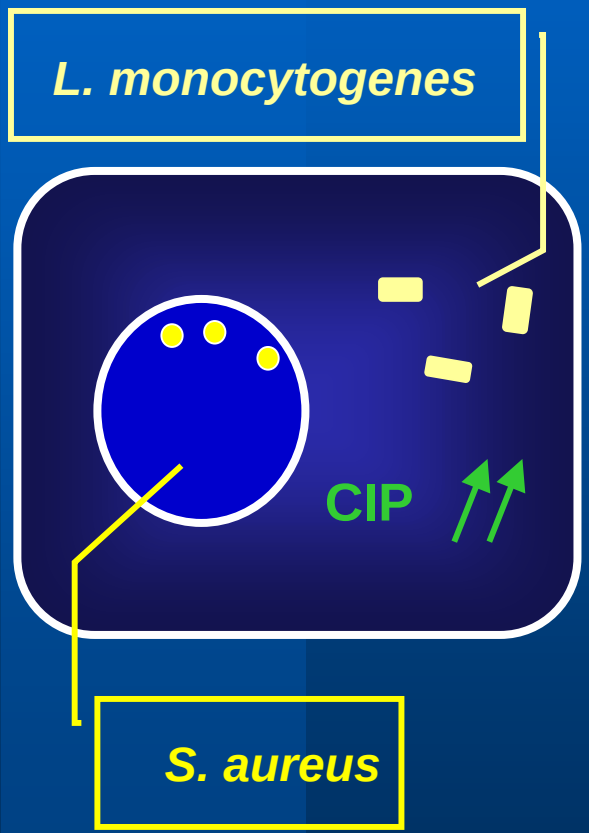


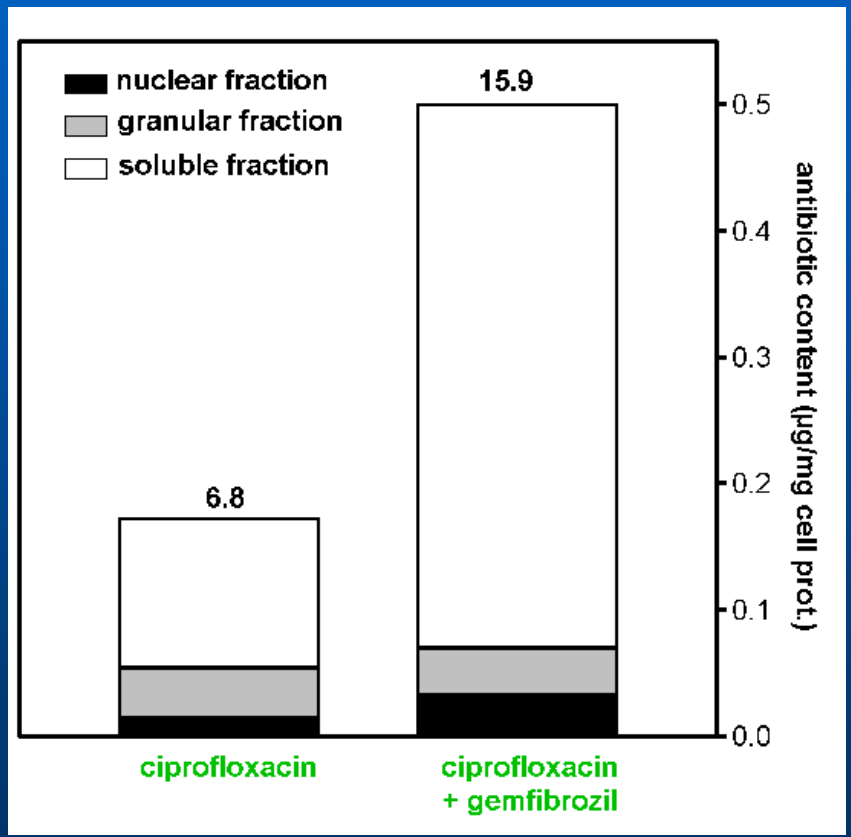


Seral *et al* (2003) JAC 51:1167-73

Influence of pump inhibitors on antibiotic distribution

gemfibrozil enhances ciprofloxacin cytosolic content

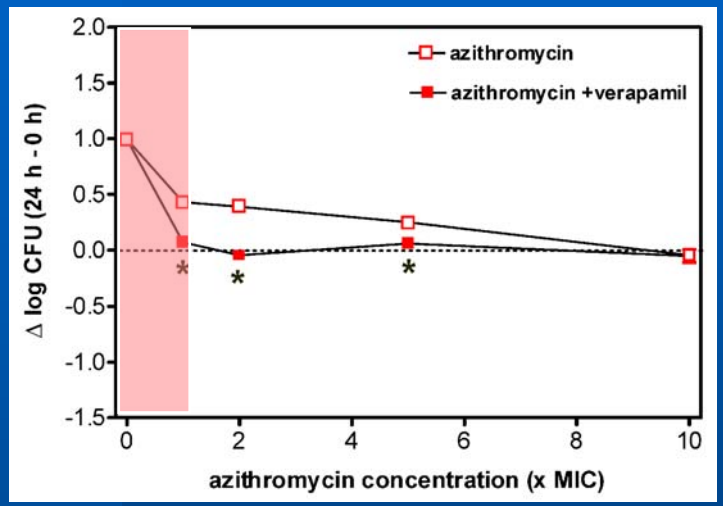


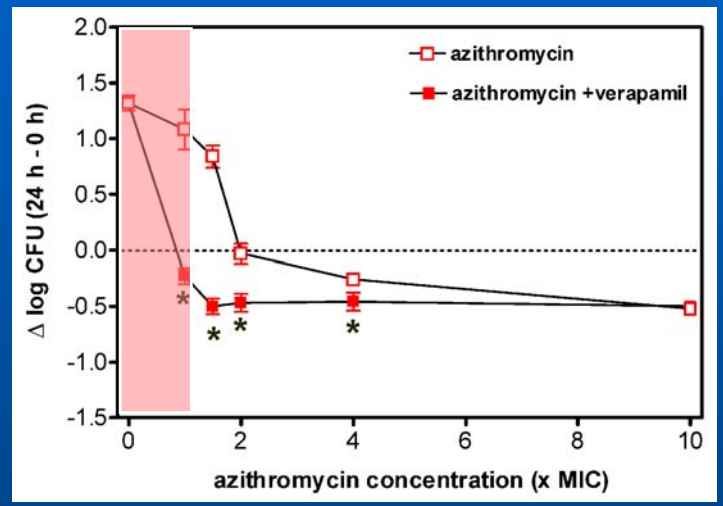


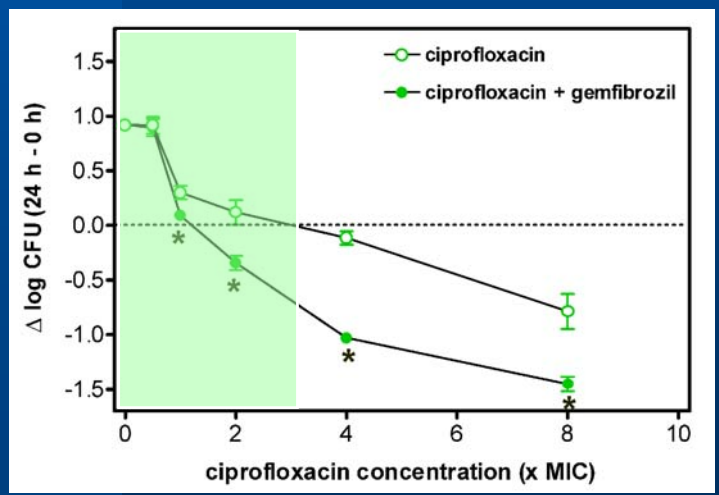
Seral *et al* (2003) JAC 51:1167-73

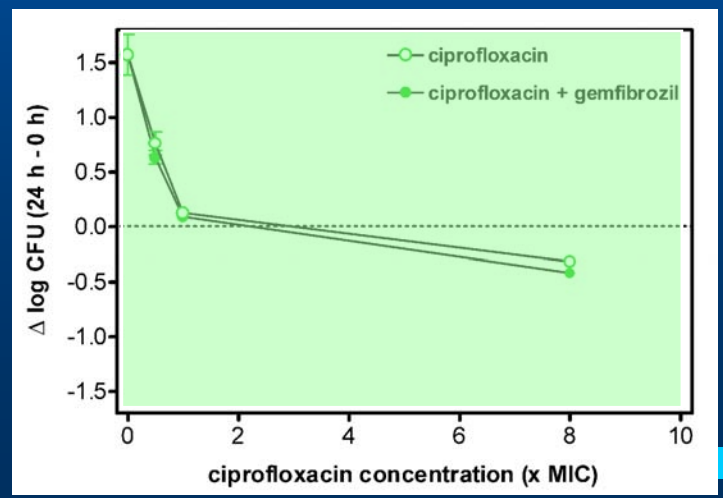
Are these effects clinically-relevant ?

constitutive efflux makes AZM and CIP activity suboptimal in a clinically-meaningful range of concentrations





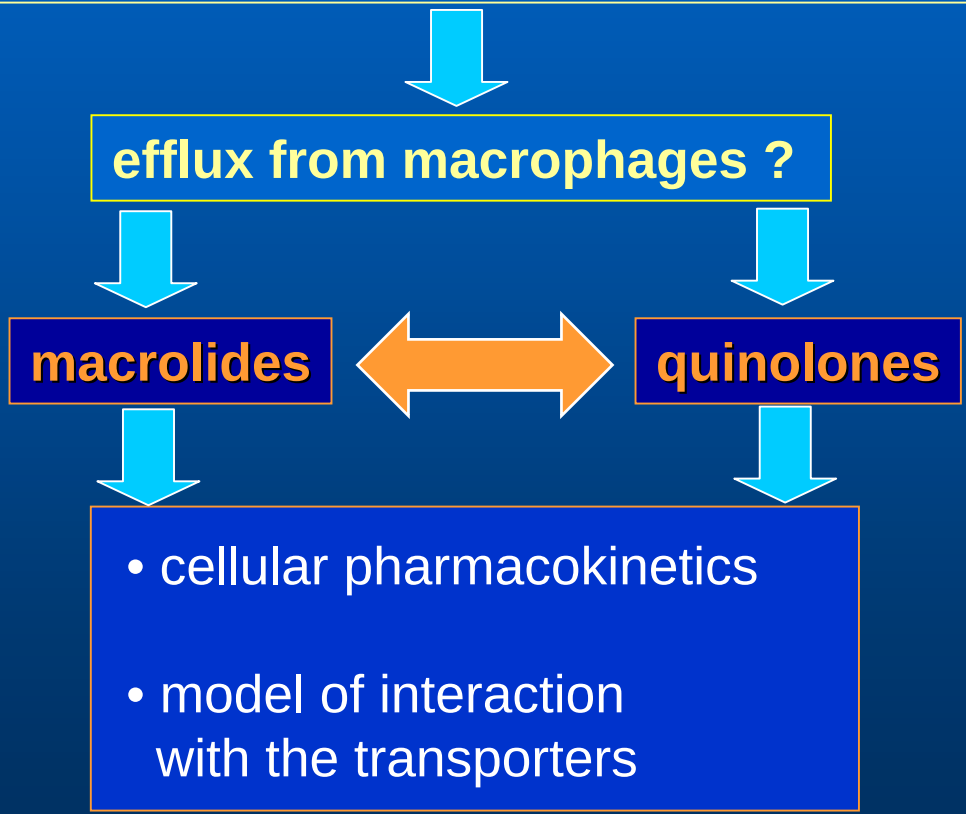




Aim of the study

amphiphilic antibiotics

- accumulating in eucaryotic cells
- considered as useful for treating intracellular infections
- known substrates of efflux pumps in bacteria



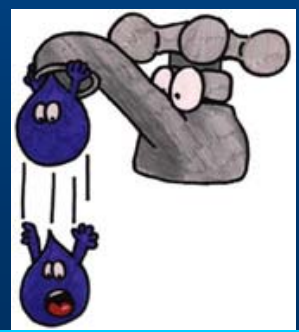
Aim of the study

amphiphilic antibiotics
- accumulating in eucaryotic cells
- considered as useful for treating intracellular infections
- known substrates of efflux pumps in bacteria

efflux from macrophages ?

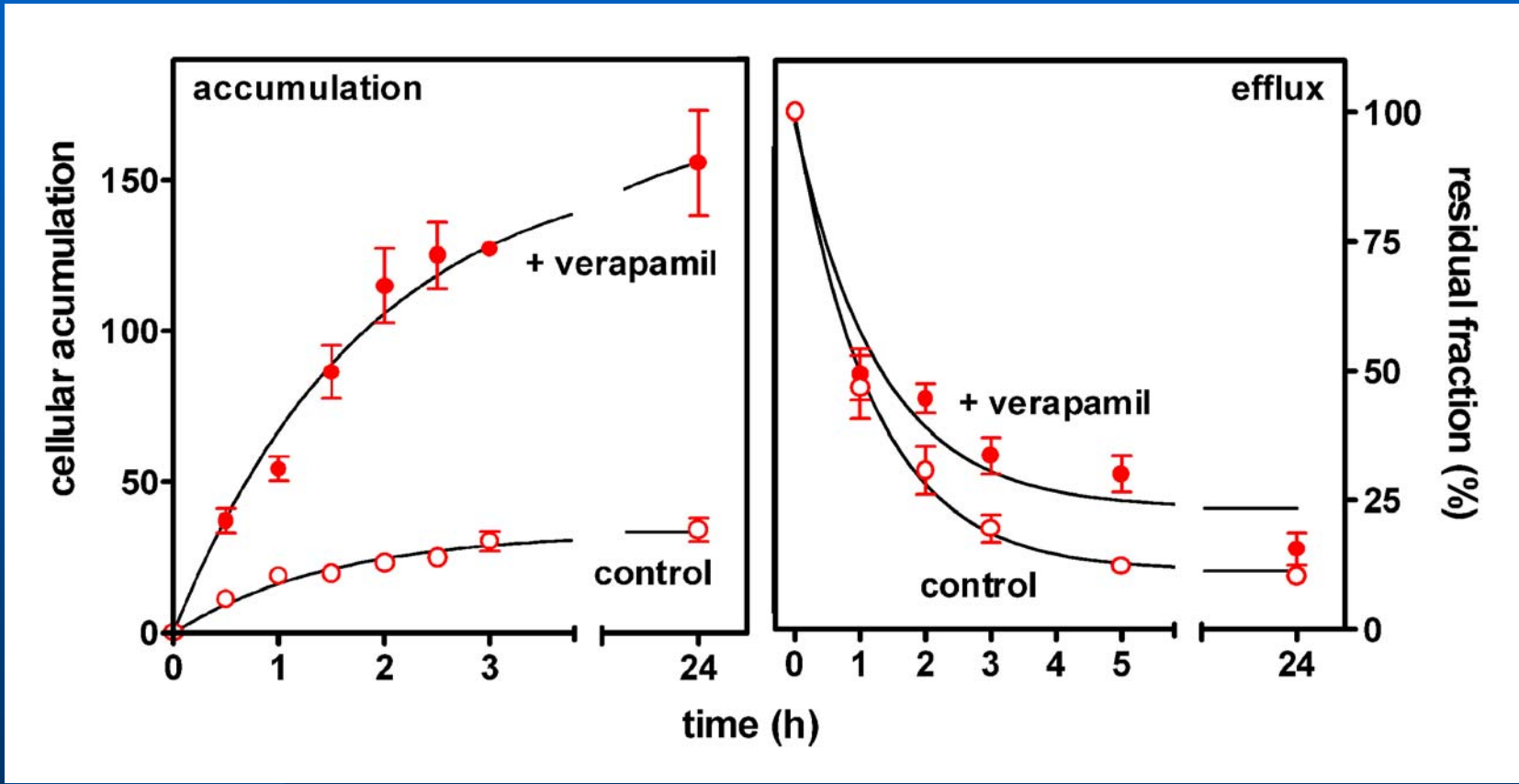
macrolides ⟷ **quinolones**

- **cellular pharmacokinetics**
- **model of interaction with the transporters**



Kinetics of accumulation and efflux for azithromycin

accumulation markedly increased; efflux marginally affected

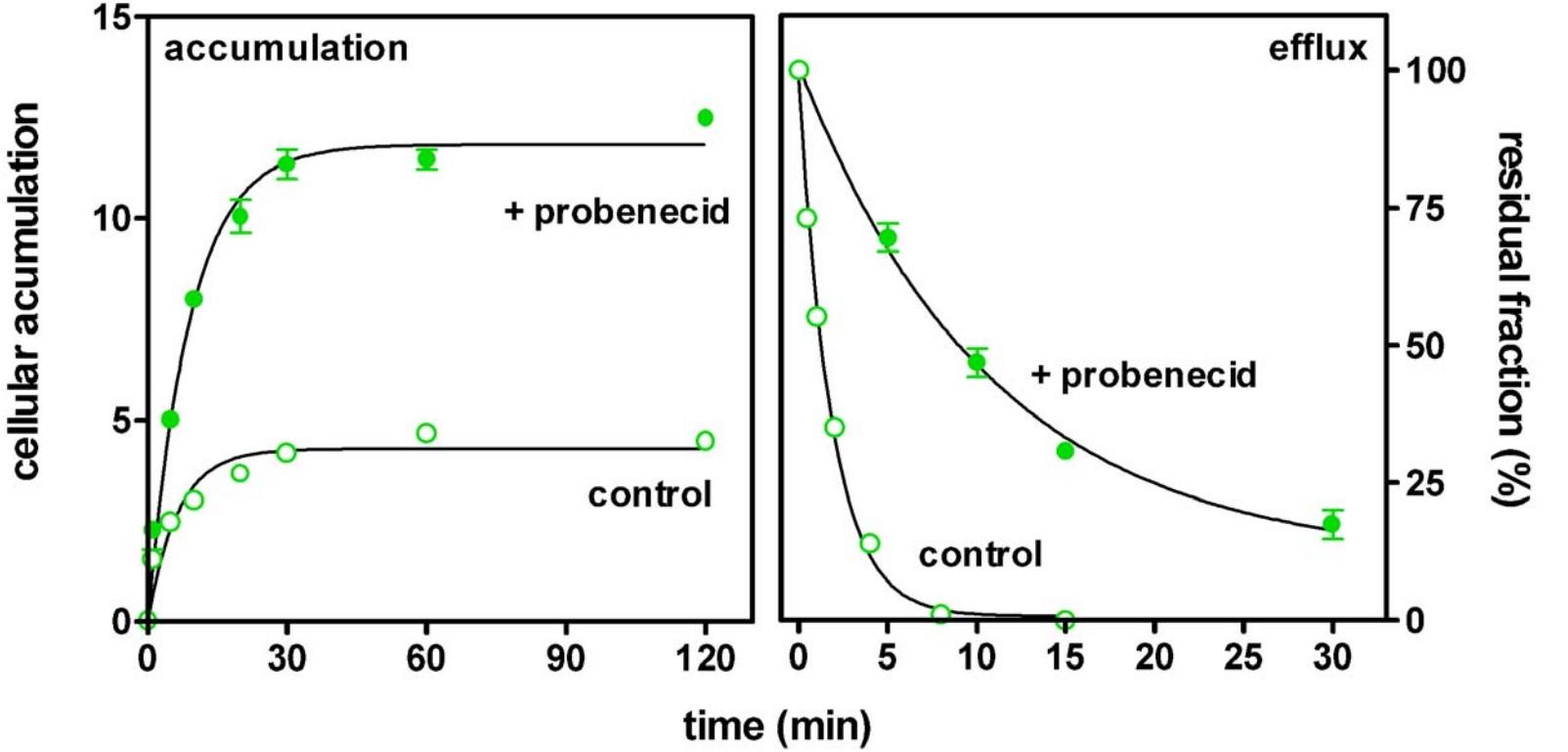


extracell. conc. 5 mg/L; verapamil 20 µM

Seral *et al* (2003) AAC 47:1047-51

Kinetics of accumulation and efflux for ciprofloxacin

both accumulation and efflux markedly affected

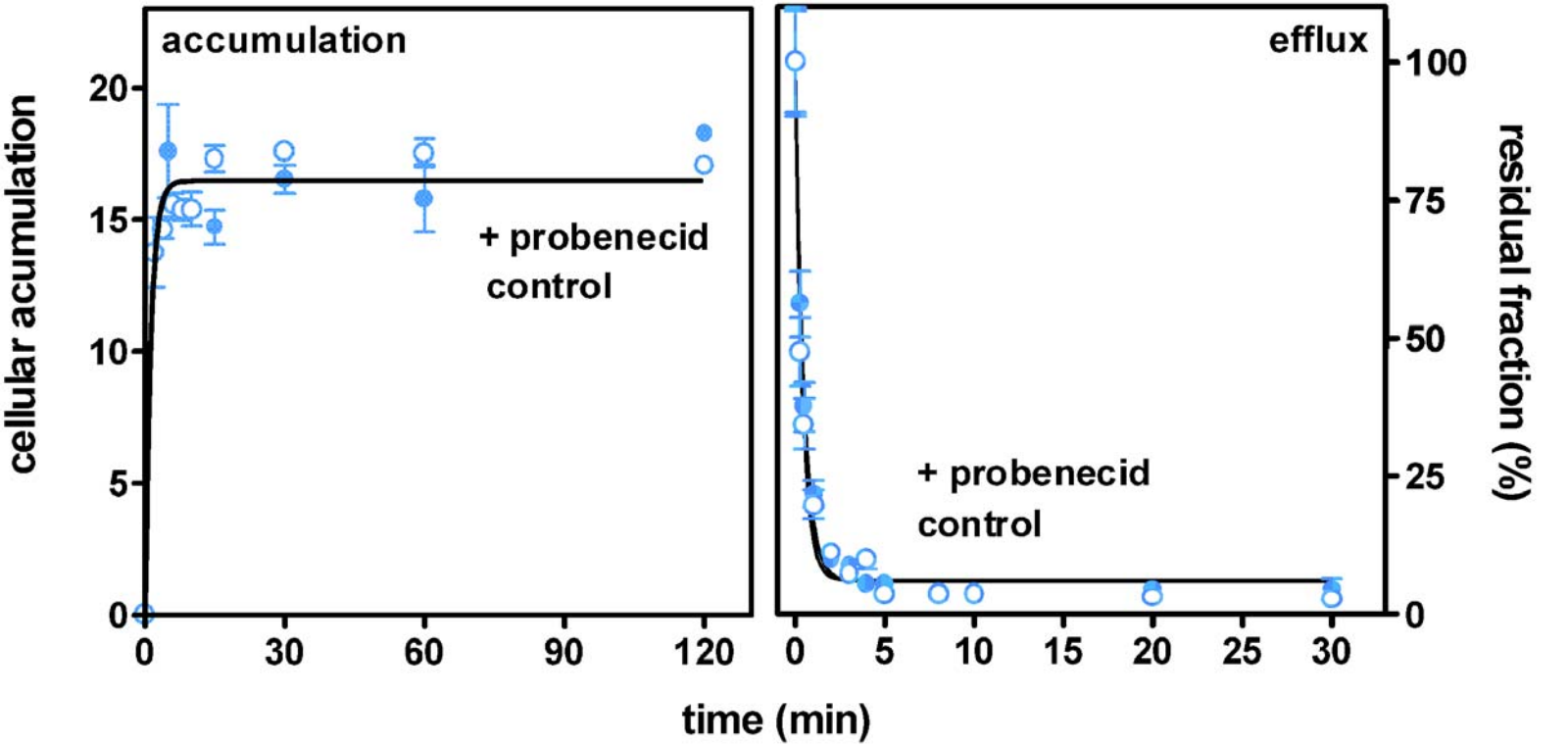


extracell. conc. 17 mg/L; probenecid 5 mM

Michot *et al*. AAC (2004) 48:2673-82

Kinetics of accumulation and efflux for moxifloxacin

neither accumulation nor efflux affected

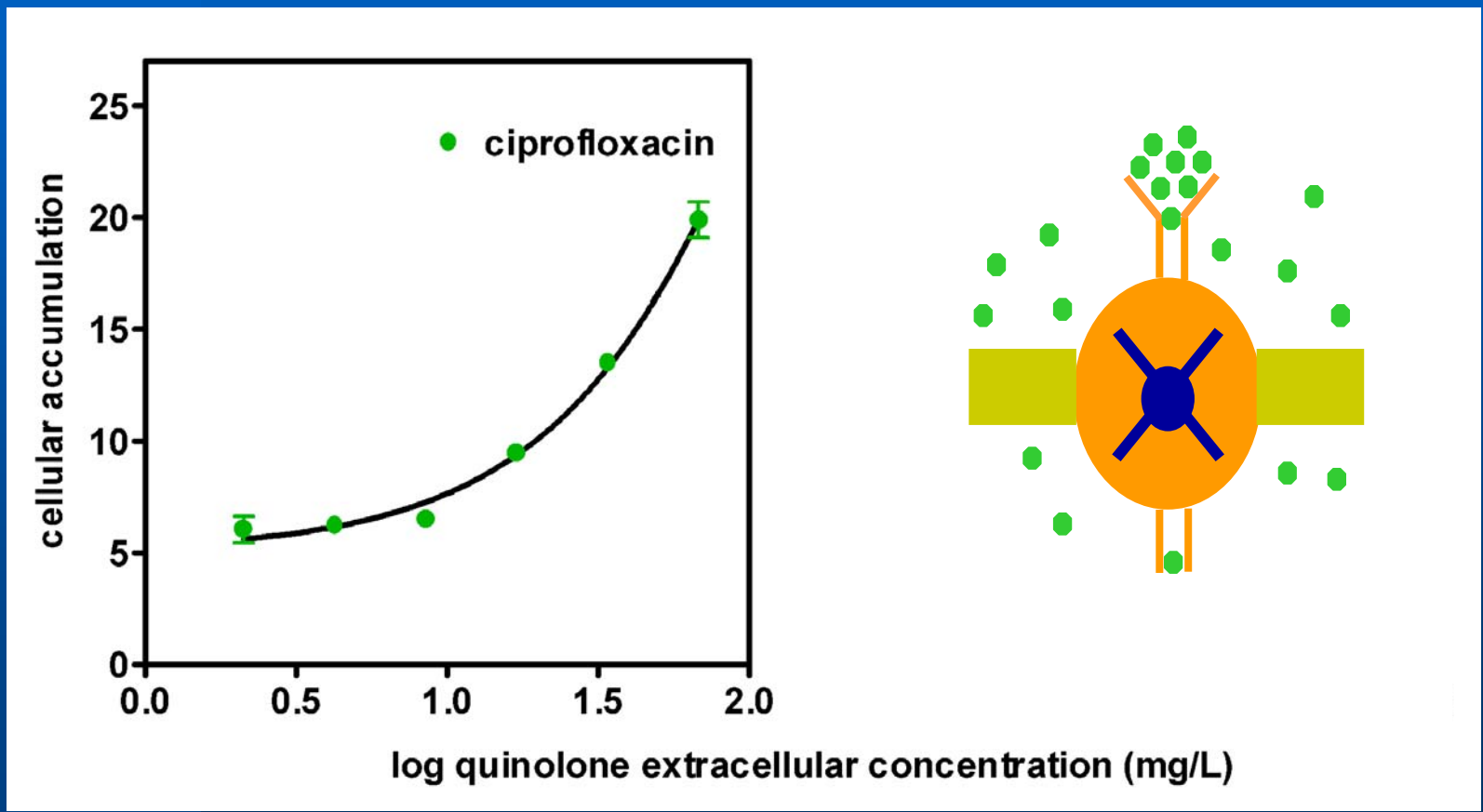


extracell. conc. 17 mg/L; probenecid 5 mM

Michot *et al*. AAC (2005) – in press

Quinolones as inhibitors of ciprofloxacin efflux

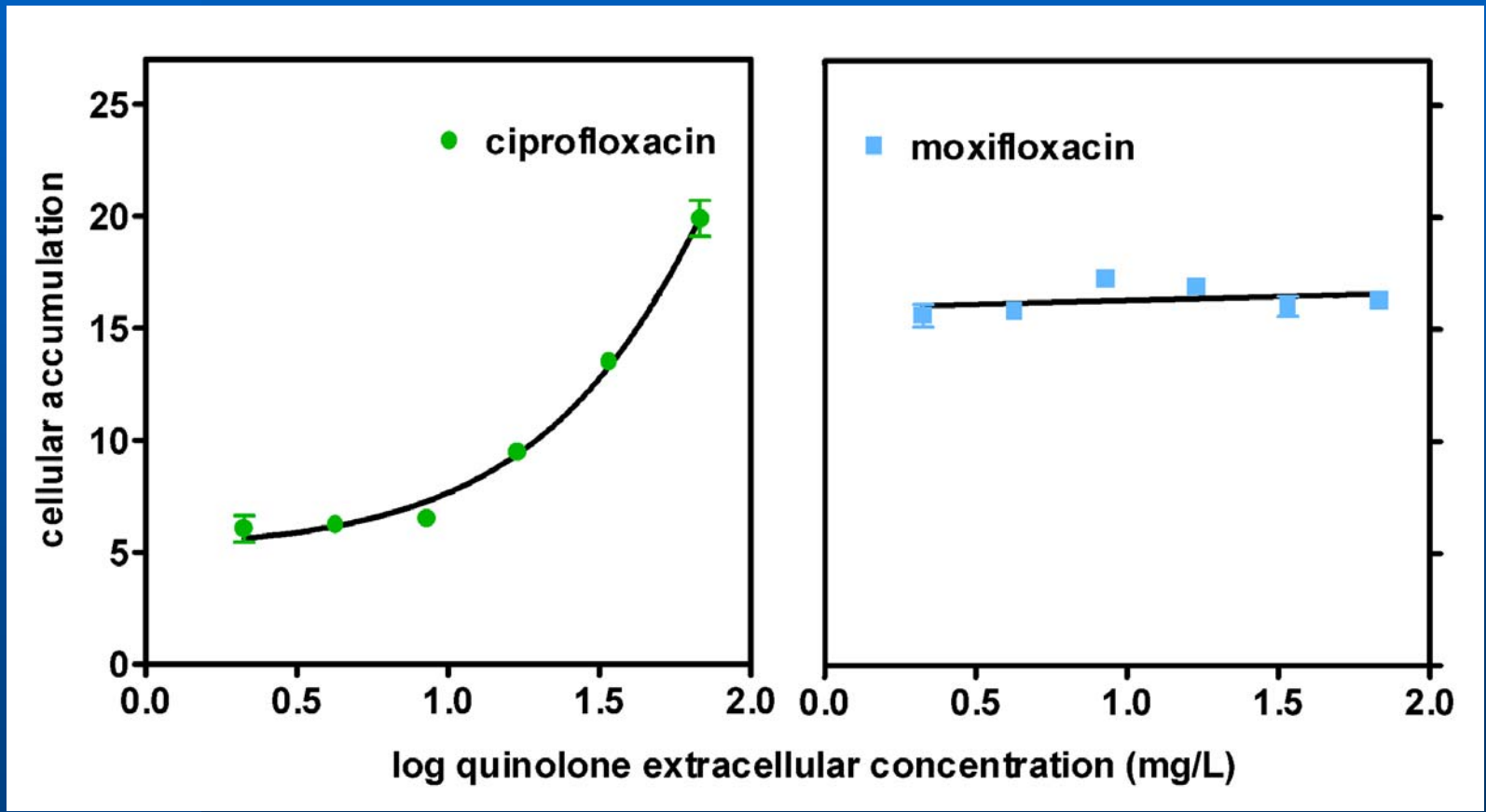
- ciprofloxacin efflux inhibited by ciprofloxacin



Michot *et al*. AAC (2005) – in press

Quinolones as inhibitors of ciprofloxacin efflux

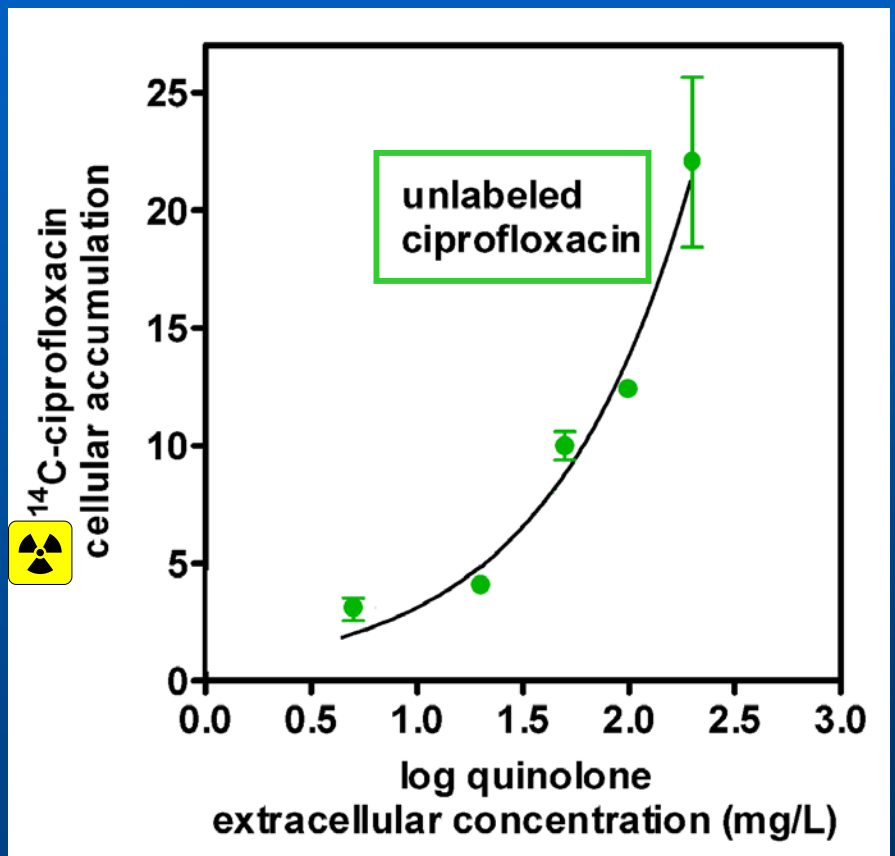
- ciprofloxacin efflux inhibited by ciprofloxacin
- moxifloxacin not affected

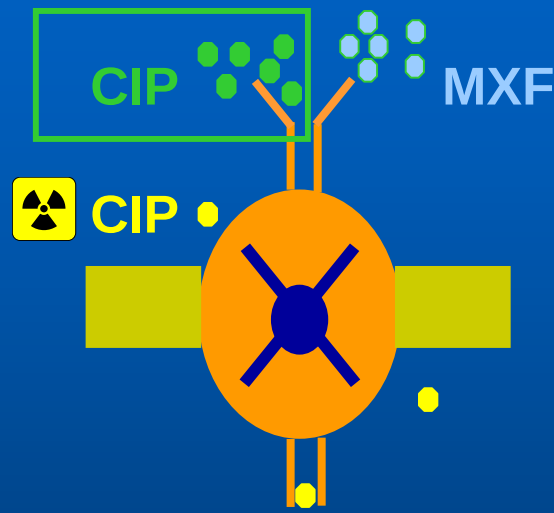


Michot *et al*. AAC (2005) – in press

Quinolones as inhibitors of ciprofloxacin efflux

- ciprofloxacin efflux inhibited by ciprofloxacin

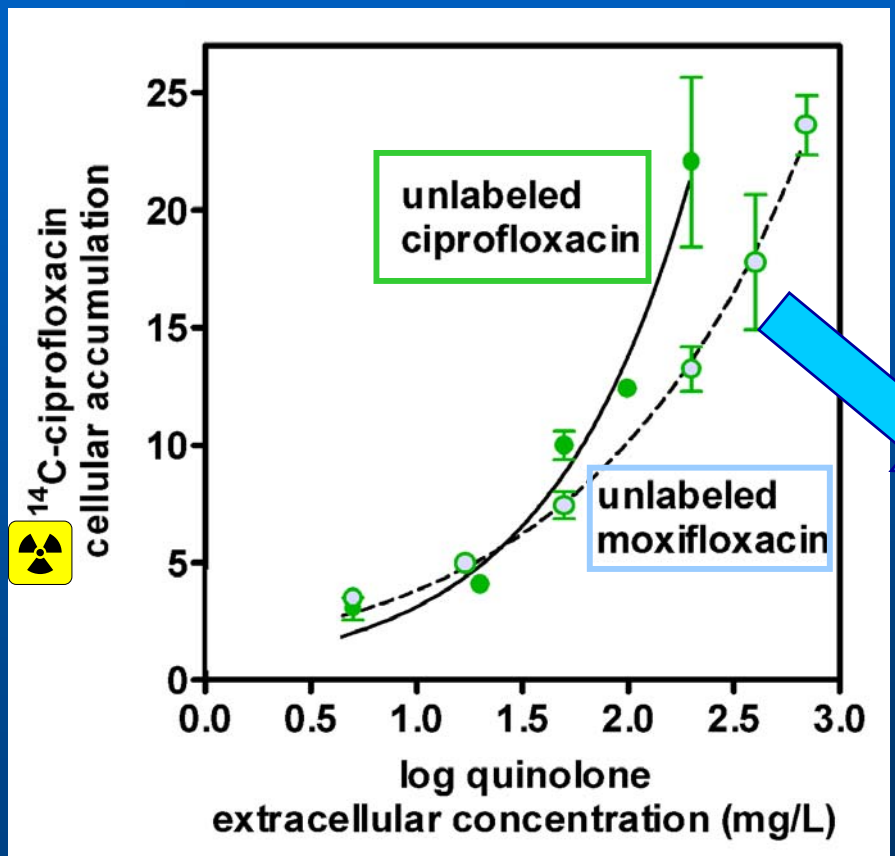


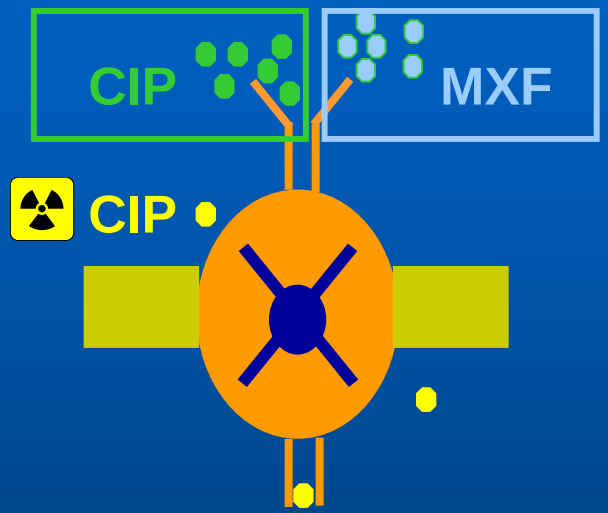


Michot *et al*. AAC (2005) – in press

Quinolones as inhibitors of ciprofloxacin efflux

- ciprofloxacin efflux inhibited by ciprofloxacin
  moxifloxacin





moxifloxacin also able to interact with the transporter !

Michot *et al*. AAC (2005) – in press

Comparison of kinetic parameters

drug	influx			efflux		
	flux (pmol/mg prot/min)	half-life (min)		flux (pmol/mg prot/min)	half-life (min)	
		control	inhibitor		control	inhibitor
AZM	1	44	71	1	49	53
CIP	5	8	8	6	1.2	7.2
MXF	68	0.2	0.2	66	0.6	0.6

Comparison of kinetic parameters

drug	influx			efflux		
	flux (pmol/mg prot/min)	half-life (min)		flux (pmol/mg prot/min)	half-life (min)	
		control	inhibitor		control	inhibitor
AZM	1	44	71	1	49	53
CIP	5	8	8	6	1.2	7.2
MXF	68	0.2	0.2	66	0.6	0.6

Comparison of kinetic parameters

drug	influx			efflux		
	flux (pmol/mg prot/min)	half-life (min)		flux (pmol/mg prot/min)	half-life (min)	
		control	inhibitor		control	inhibitor
AZM	1	44	71	1	49	53
CIP	5	8	8	6	1.2	7.2
MXF	68	0.2	0.2	66	0.6	0.6

Comparison of kinetic parameters

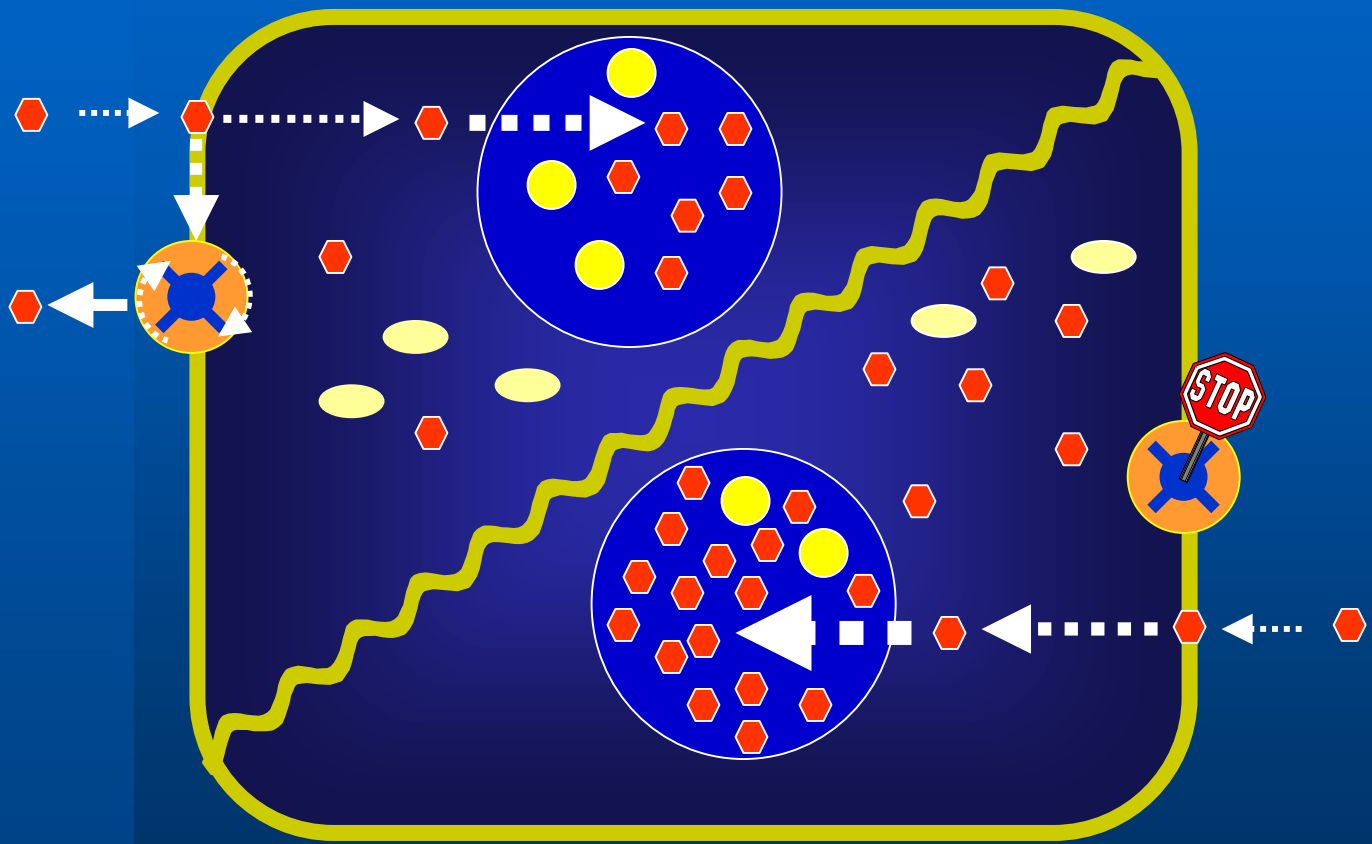
drug	influx			efflux		
	flux (pmol/mg prot/min)	half-life (min)		flux (pmol/mg prot/min)	half-life (min)	
		control	inhibitor		control	inhibitor
AZM	1	44	71	1	49	53
CIP	5	8	8	6	1.2	7.2
MXF	68	0.2	0.2	66	0.6	0.6

Comparison of kinetic parameters

drug	influx			efflux		
	flux (pmol/mg prot/min)	half-life (min)		flux (pmol/mg prot/min)	half-life (min)	
		control	inhibitor		control	inhibitor
AZM	1	44	71	1	49	53
CIP	5	8	8	6	1.2	7.2
MXF	68	0.2	0.2	66	0.6	0.6

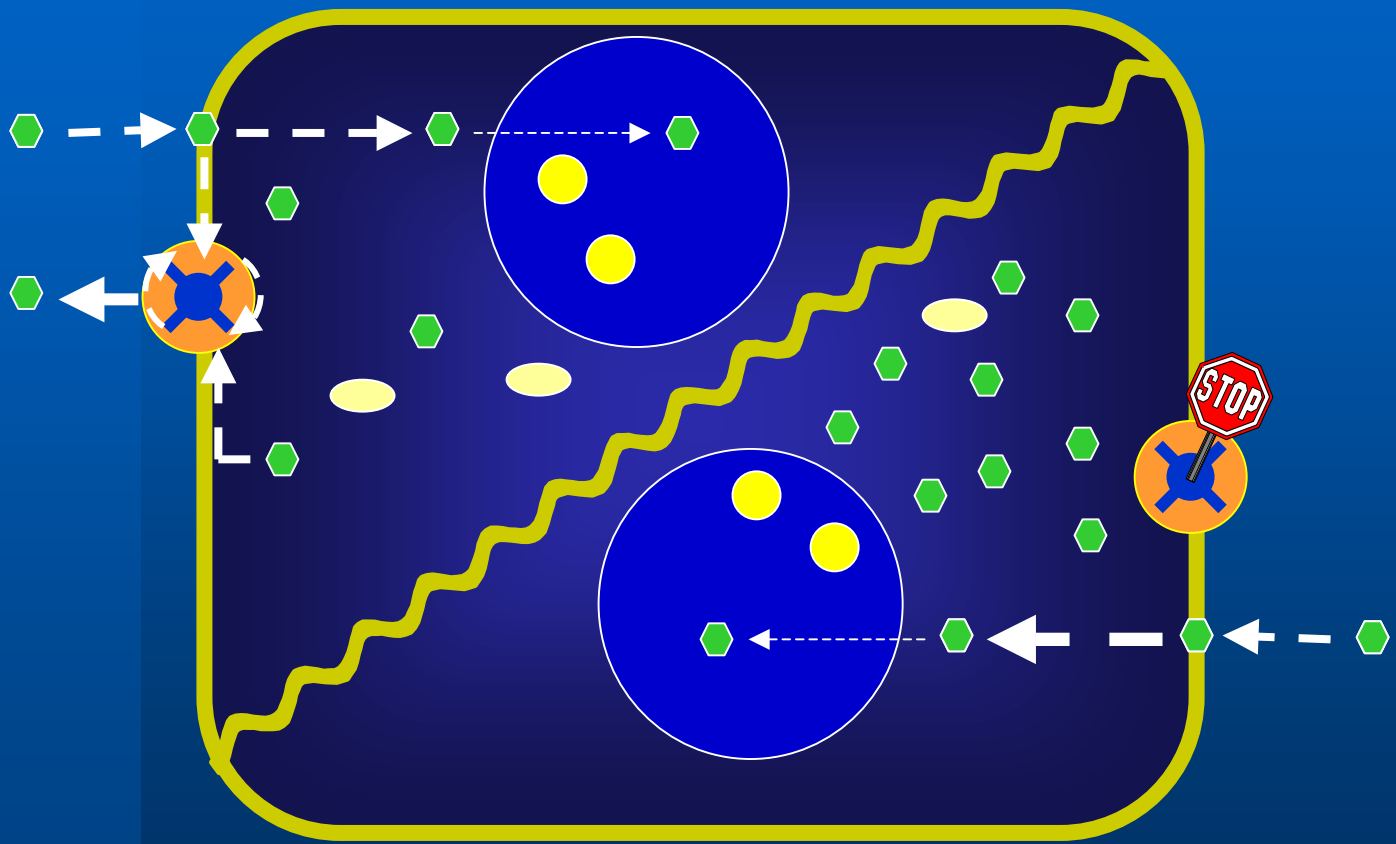
Azithromycin, ‘kick-back’ model

Gaj *et al*. (1998) Biochem. Pharmacol. 55:1199-211



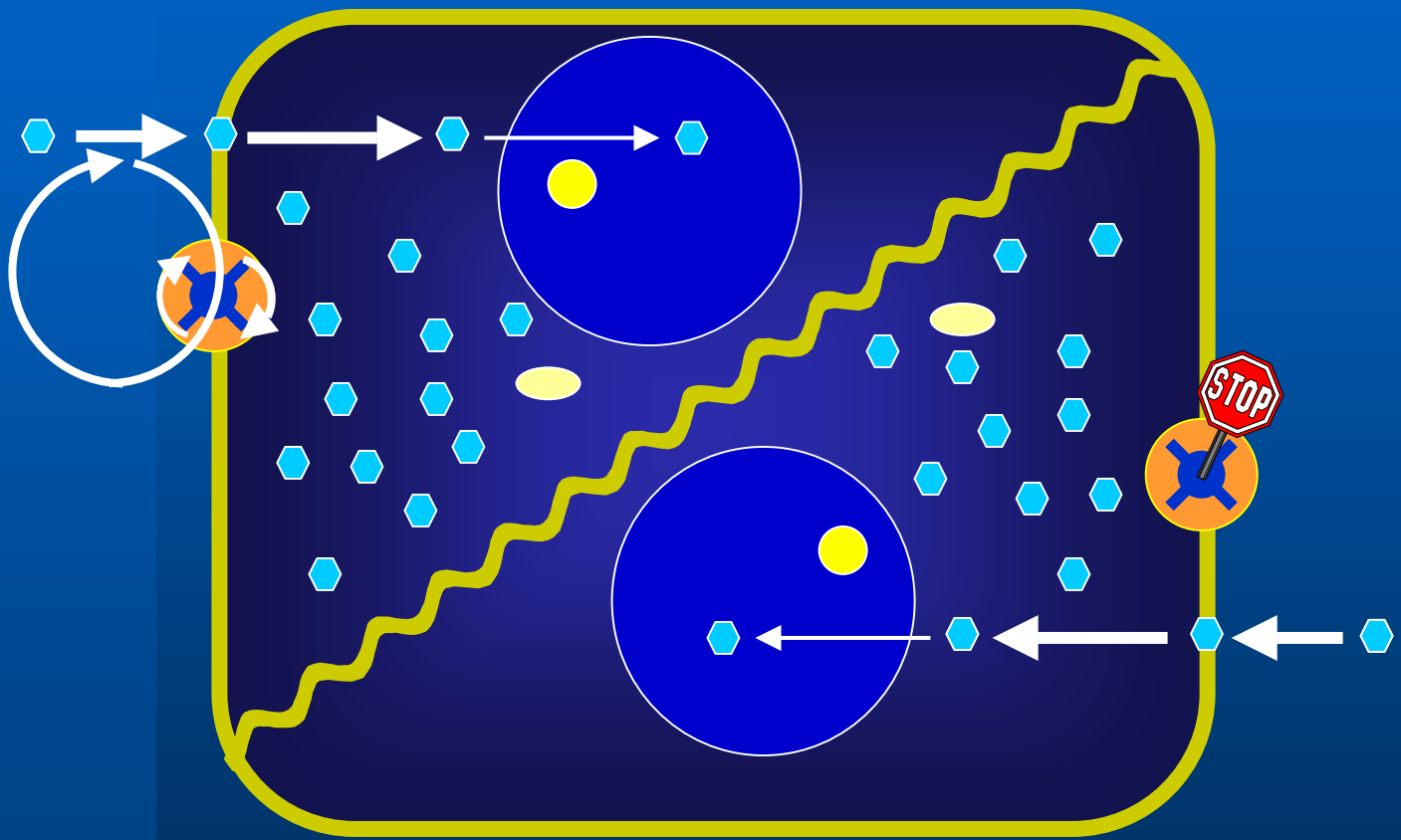
Ciprofloxacin, classical model

Kolaczkowski & Goffeau (1997) Pharmacol. Ther. 76:219-42



Moxifloxacin, ‘futile-cycle’ model

Eytan *et al*. (1996) JBC 271:12897-902



Conclusion

constitutive efflux of antibiotics in macrophages

pharmacokinetics:
suboptimal cellular accumulation

pharmacodynamics:
suboptimal intracell. activity

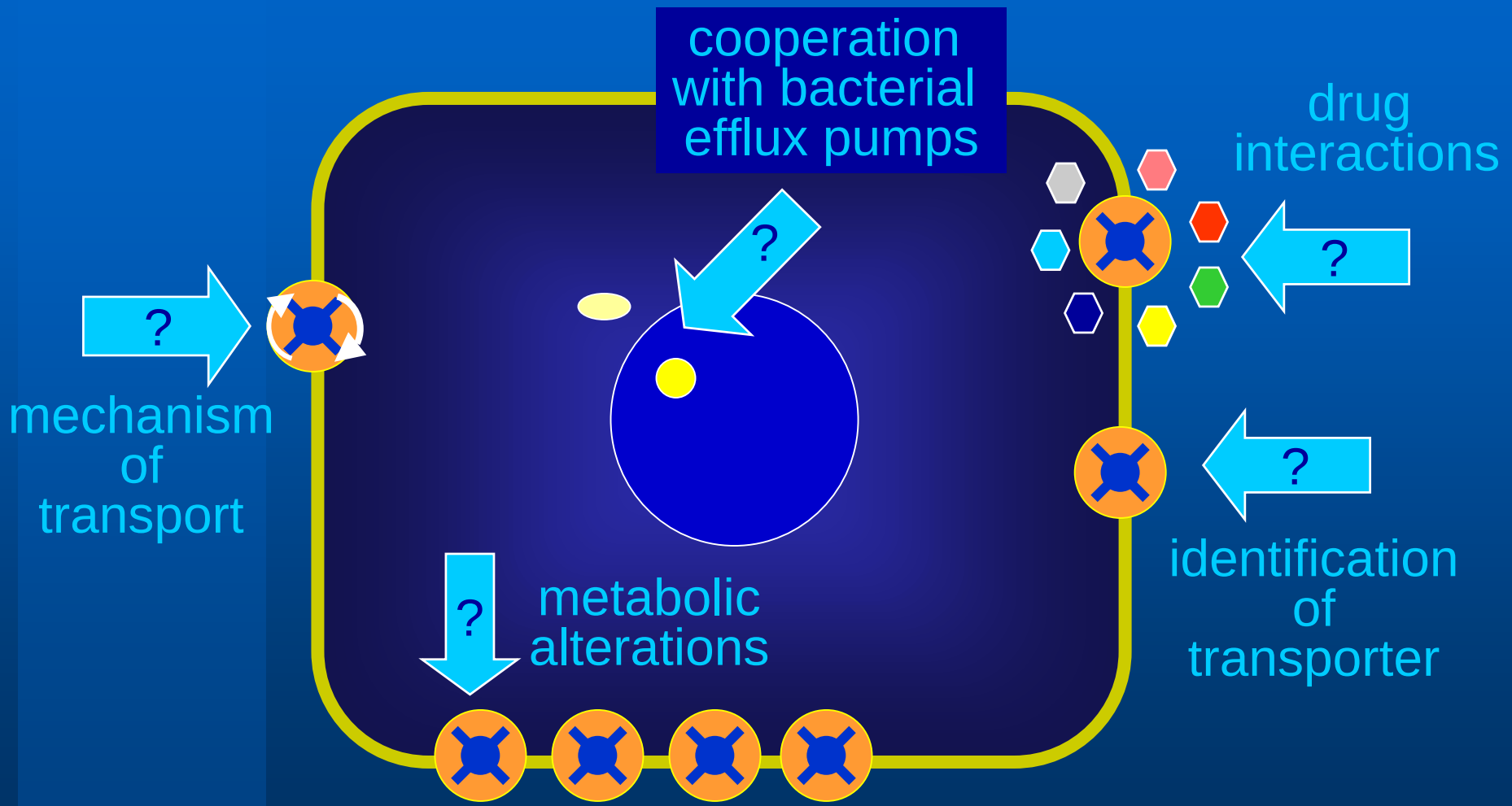
pharmacology:
differences in affinity within a AB class

pharmacokinetics:
wide spectrum transporters

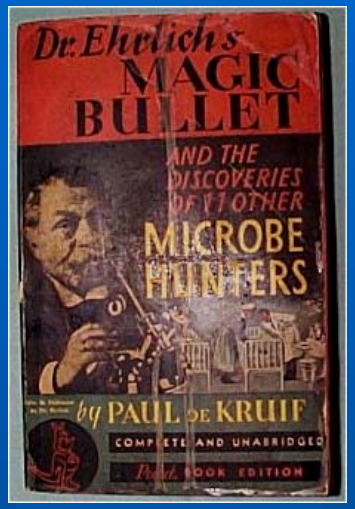
resistance ?

drug interactions ?

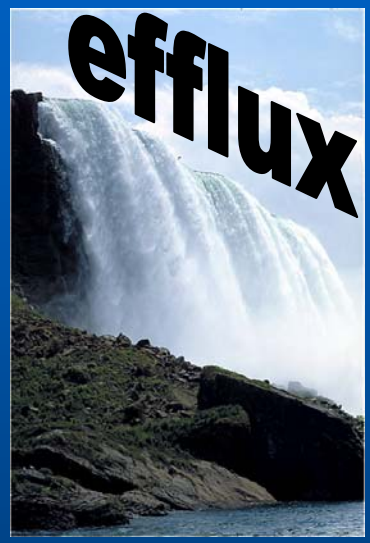
Questions for future research



Take home message



constitutive efflux is part of the game



→Take it into account

- **in the choice of your « magic bullets » …**
- **for their optimal targeting**

Thanks to ...

Thanks to ...

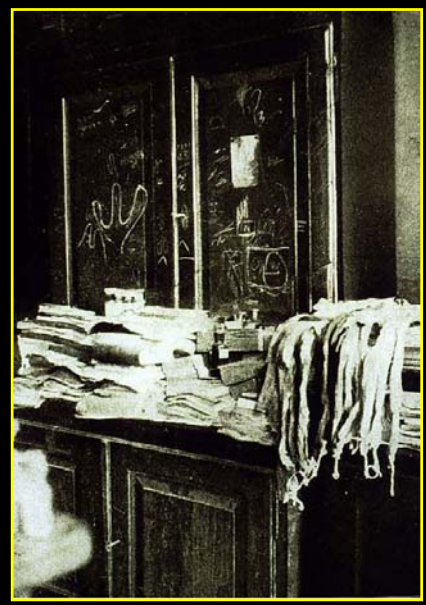
Evaluating magic bullets

- pharmacokinetics
  H. Chanteux, M. Heremans, J.M. Michot

- pharmacodynamics
  M. Barcia, N. Bles, S. Carryn, S. Lemaire, A. Olivier, C. Seral, S. Van de Velde

- toxicodynamics
  J.P. Montenez, H. Servais, D. Tyteca

- biophysics & molecular biology
  N. Caceres, N. Fa



Thanks to ...

New magic bullets

- chemistry
  E. Colacino, C. Dax, L. Efron, T. Happaerts, M. Renard

- pharmacology
  I. Tytgat, D. Van Ackeren

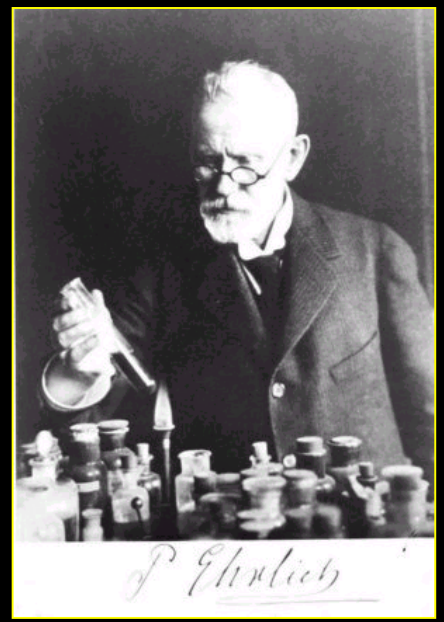
- modeling
  M. Prévost, M. Rooman, S. Vandevuer

Thanks to ...

Resistance to magic bullets

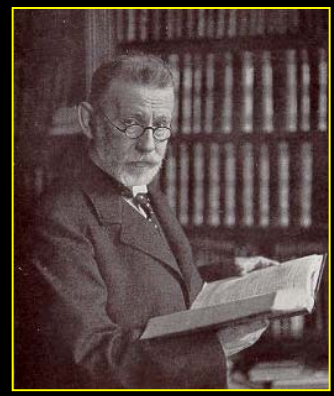
- efflux
  L. Avrain, N. Mesaros

- glycopeptides
  P. Courvalin and his team



Thanks to ...

Clinical use of magic bullets



- clinical pharmacy
  E. Ampe, V. Basma, A. Spinewine

Thanks to ...

Playing with magic bullets

- technical staff

  N. Aguilera, M.C. Cambier, O. Meert,
  F. Renoird, M. Vergauwen

- secretary

  M. Breugelmans

Thanks to ...

Ehrlich's colleagues

Thanks to ...

Inspiring research on magic bullets

Thanks to ...

Evaluating research on magic bullets

P. Courvalin, A. Dalhoff, M. Delmée,
H. Derendorf,Y. Glupczynski,
E. Sonveaux, F. Zech

Thanks to ...

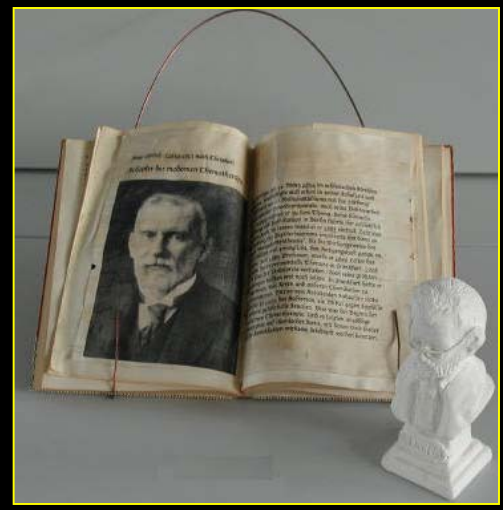
Paying for research on magic bullets

FNRS

UCL

Thanks to ...

Managing research on magic bullets



M.P. Mingeot-Leclercq

P.M. Tulkens


Thank you
for your attention