Importance of Global Harmonization of Antimicrobial Susceptibility Testing in Canada for Defining Antimicrobial Resistance

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Disclosures.

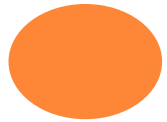
- Chair - Canadian Committee on Antimicrobial Susceptibility Testing (CANCAST)
- Quality Control studies for most new antimicrobial agents.
- Reviewer at CLSI and member of the CLSI Quality Control working group
- Deputy-Convener of ISO-TC212 (Technical Committee on Quality Laboratory Management) Working Group 4 (Microbiology)
- Member of CSA Z252 (TC212 Mirror Committee).
- Consultant for Thermo Fisher (Global) on antimicrobial susceptibility devices.

Objectives

- Recognize reasons for differences in antimicrobial agent breakpoints.
- Identify the need for global harmonization of antimicrobial agent breakpoints
- Summarize the development of a Canadian National Antibiotic Committee (CANCAST)
- Recognize the reasons for differences in antimicrobial agent breakpoints.
- Review the need for global harmonization of antimicrobial agent breakpoints

What's wrong with the status quo?

- Laboratories tend to do their own thing.
- Reporting antimicrobial susceptibility is variable
- Difficult compare rates of resistance for surveillance (is an "R" really an "R")
- Treatment may be based on either false resistance or susceptibility.
- Methodologies are the same but may in fact be different.
- Research into antimicrobial resistance becomes genetic rather than phenotypic.



Why differences in antimicrobial breakpoints.

- Historical
  - Early on, there was no resistance to newly developed agents.
  - So-called wild type strains (no known resistance determinants).
  - Breakpoints defined by clinical and microbiological failures in clinical trials
    - Buffer zones created – pressure from pharmaceutical clinical trials for higher breakpoints. (the “90 – 60” Rule!)
    - No clear public health need to create a lower breakpoint. No one was telling the Regulatory bodies when patients failed therapy.

Why differences in antimicrobial breakpoints.

- What happened!
  - The micro-organisms are a lot smarter than we mortals – they've been here a lot longer and will be long after we're gone.
  - Breakpoints that were created did not accurately detect the emergence of resistant strains.
  - Antimicrobial susceptibility testing methodologies were not standardized.
  - Early days
    - – any zone of inhibition was considered susceptible. Larger zones were just more susceptible.
    - - MIC testing was not widely used.
  - There was limited understanding of pharmacodynamics – the bug, the drug and the host!
  - Different groups established antimicrobial breakpoints based on their own methodologies and criteria.
  - The practical result: approximately 50% of CLSI breakpoints are different from EUCAST – almost all of those differences are higher breakpoints by CLSI.

Differences in International breakpoints

- Recent local clinical example.
  - Inpatient in a regional hospital with extensive cellulitis, fasciotomy, immobile, urinary catheter in place. Multiple antimicrobials used to treat the cellulitis
  - *Klebsiella pneumoniae* isolated from catheter urine at end of April . Treated with ciprofloxacin. Urine collected again mid-May. Same organism with same susceptibilities. Treated with nitrofurantoin.

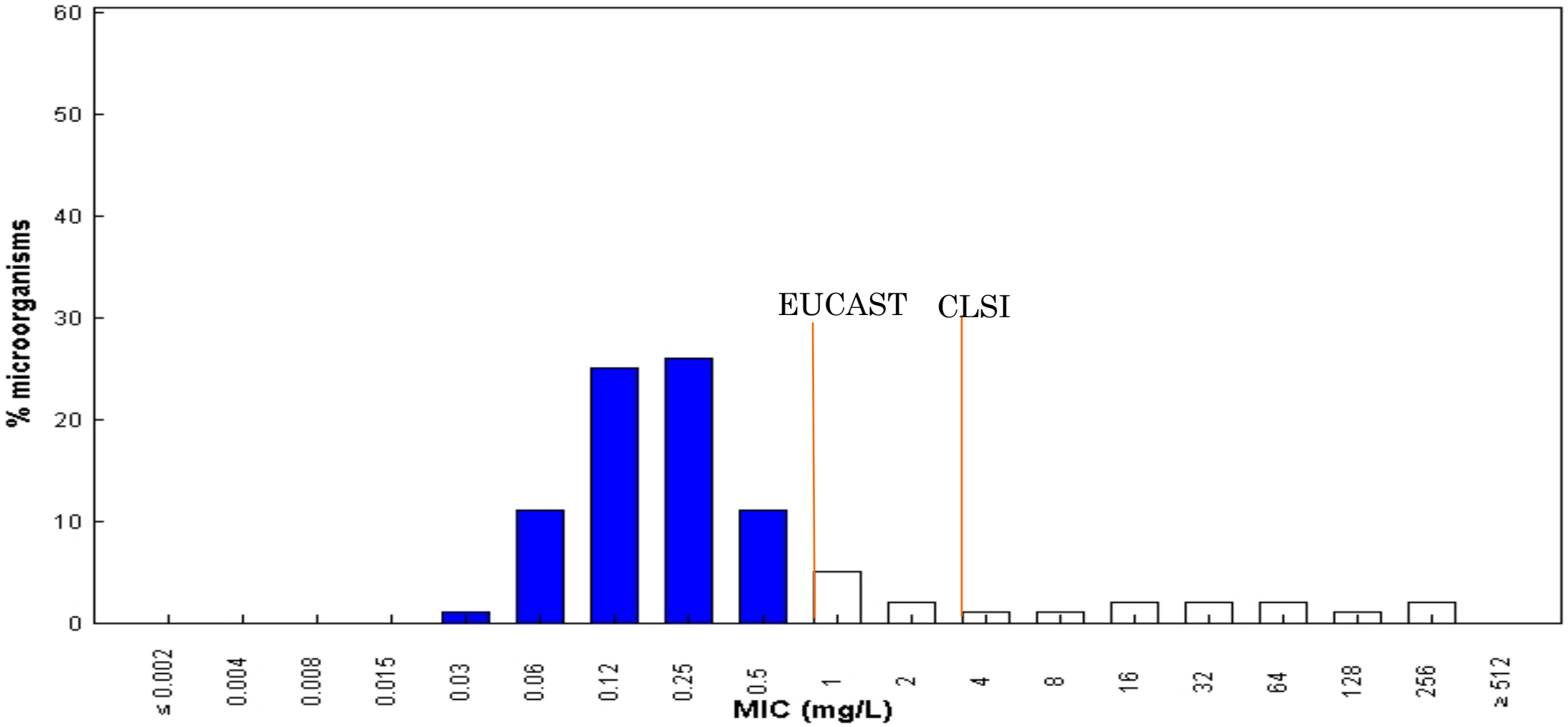
    Isolate resistant to first and second generation cephalosporins, and TMP-SMX: nitrofurantoin – Intermediate

    Reported as susceptible to carbapenems, and ciprofloxacin (MIC =1mg/L),

    ceftriaxone MIC 1 mg/L – Susceptible; ceftazidime 4 mg/L – Susceptible.

Ceftazidime / Klebsiella pneumoniae
International MIC Distribution - Reference Database 2016-05-23

MIC distributions include collated data from multiple sources, geographical areas and time periods and can never be used to infer rates of resistance



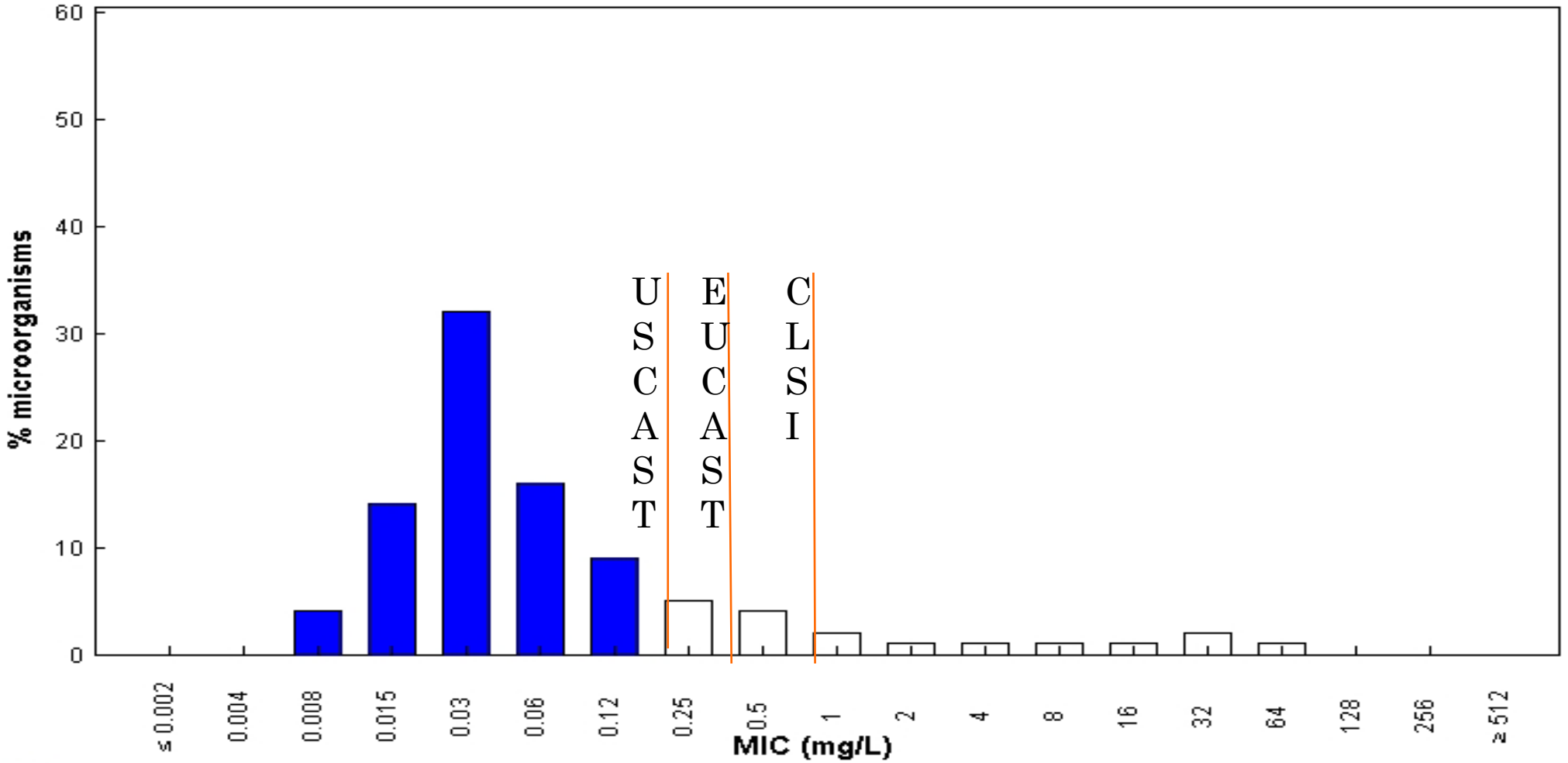
MIC
Epidemiological cut-off (ECOFF): 0.5 mg/L
Wildtype (WT) organisms: ≤ 0.5 mg/L

5326 observations (68 data sources)

EUCAST - 2016

Ciprofloxacin / Klebsiella pneumoniae
International MIC Distribution - Reference Database 2016-05-25

MIC distributions include collated data from multiple sources, geographical areas and time periods and can never be used to infer rates of resistance



MIC
Epidemiological cut-off (ECOFF): 0.125 mg/L
Wildtype (WT) organisms: ≤ 0.125 mg/L

5905 observations (71 data sources)

EUCAST 2016

Breakpoint differences (mg/L).

	CLSI		FDA		EUCAST		USCAST	
	S	R	S	R	S	R	S	R
Ceftriaxone	≤1	≥4	≤1	≥4	≤1	> 4	≤1	≥4
Ceftazidime	≤4	≥ 16	≤4	≥16	≤1	>2	≤1	≥4
Ciprofloxacin	≤ 1	≥ 4	≤ 1	≥ 4	≤ 0.5	>1	≤ 0.25	≥ 1

United States Committee on Antimicrobial Susceptibility Testing (USCAST)

MIC Breakpoint Tables Comparing the Interpretive Criteria of CLSI, EUCAST, USA-FDA and USCAST

Version 1.0, valid from 06-23-2015

Table 1. USCAST MIC breakpoints compared to three other antimicrobial agent breakpoint organizations when testing the fluoroquinolone class compounds (modified from the Quinolone Report, 2015).

Organism/Antimicrobial	MIC breakpoints in µg/mL by criteria organization (Susceptible/Resistant)			
	CLSI[a]	USA-FDA	EUCAST[b]	USCAST
Enterobacteriaceae				
Ciprofloxacin	≤1 / ≥4	≤1 / ≥4[c]	≤0.5 / >1	≤0.25 / ≥1
Levofloxacin	≤2 / ≥8	≤2 / ≥8[d]	≤1 / >1	≤0.5 / ≥2
Moxifloxacin	--	≤2 / ≥8[e]	≤0.5 / >1	≤0.25 / ≥0.5 (valid for *E. coli*, *Klebsiella* spp., *Enterobacter* spp., *Citrobacter* spp., and *M. morganii*)
Norfloxacin (UTI)	≤4 / ≥16	≤4 / ≥16[f]	≤0.5 / >1	-
Ofloxacin	≤2 / ≥8	≤2 / ≥8[g]	≤0.5 / >1	-
Nalidixic acid (UTI)	≤16 / ≥32	≤16 / ≥32[h]	Screen only (a surrogate)	≤16 / ≥32 (Salmonella susceptibility screen)[i]
Pseudomonas spp.				
Ciprofloxacin	≤1 / ≥4	≤1 / ≥4[c]	≤0.5 / >1	≤0.5 / ≥1 (high dose)
Levofloxacin	≤2 / ≥8	≤2 / ≥8[d]	≤1 / >2	≤0.5 / ≥1 (high dose)
Norfloxacin (UTI)	≤4 / ≥16	≤4 / ≥16[f]	-	-
Ofloxacin	≤2 / ≥8	≤2 / ≥8[g]	-	-
S. maltophilia				
Levofloxacin	≤2 / ≥8	-[d]	-	-

Breakpoint Changes Affect Resistance

Susceptibility breakpoints	Carbapenem and CLSI year[a]							
	Doripenem		Ertapenem		Imipenem		Meropenem	
	2010	2013	2010	2013	2010	2013	2010	2013
Susceptible	NC	≤1	≤2	≤0.5	≤4	≤1	≤4	≤1
Intermediate	NC	2	4	1	8	2	8	2
Resistant	NC	≥4	≥8	≥2	≥16	≥4	≥16	≥4

a. Criteria from CLSI reference (5-7)
b. NC=no criteria published

Rennie and Jones CJIDMM. 2014

Breakpoint Changes affect Resistance

Enteric group (no. tested)	% susceptible (2012/2010 criteria): Ertapenem	Imipenem	Meropenem	Doripenem
Enterobacteriaceae (19,382)	97.11/98.10	92.38/98.58	98.28/98.61	98.32/-[a]
E. coli (6,882)	99.56/99.83	99.83/99.99	99.85/99.93	99.90/-
Klebsiella spp. (5,467)	94.71/95.08	95.32/95.85	95.26/95.88	95.34/-
Enterobacter spp. (2,662)	92.90[b]/97.90	94.74[b]/98.99	98.65/99.21	98.69/-
Citrobacter spp. (746)	97.72/98.79	97.05/99.33	98.79/99.33	98.93/-
Serratia spp. (1,119)	98.03/98.84	92.94[b]/99.29	98.84/99.20	98.75/-
P. mirabilis (1,244)	99.92/100.0	64.47[b]/99.76	99.92/100.0	99.76/-
M. morganii (490)	100.0/100.0	19.59[b]/100.0	100.0/100.0	100.0/-

a. No earlier breakpoints were published by CLSI.
b. Significant ($p < 0.05$) decline in susceptibility rate, generally >4% decrease; results are underlined for each species.

Rennie and Jones CJIDMM. 2014

Tools for defining and optimizing antimicrobial breakpoints.

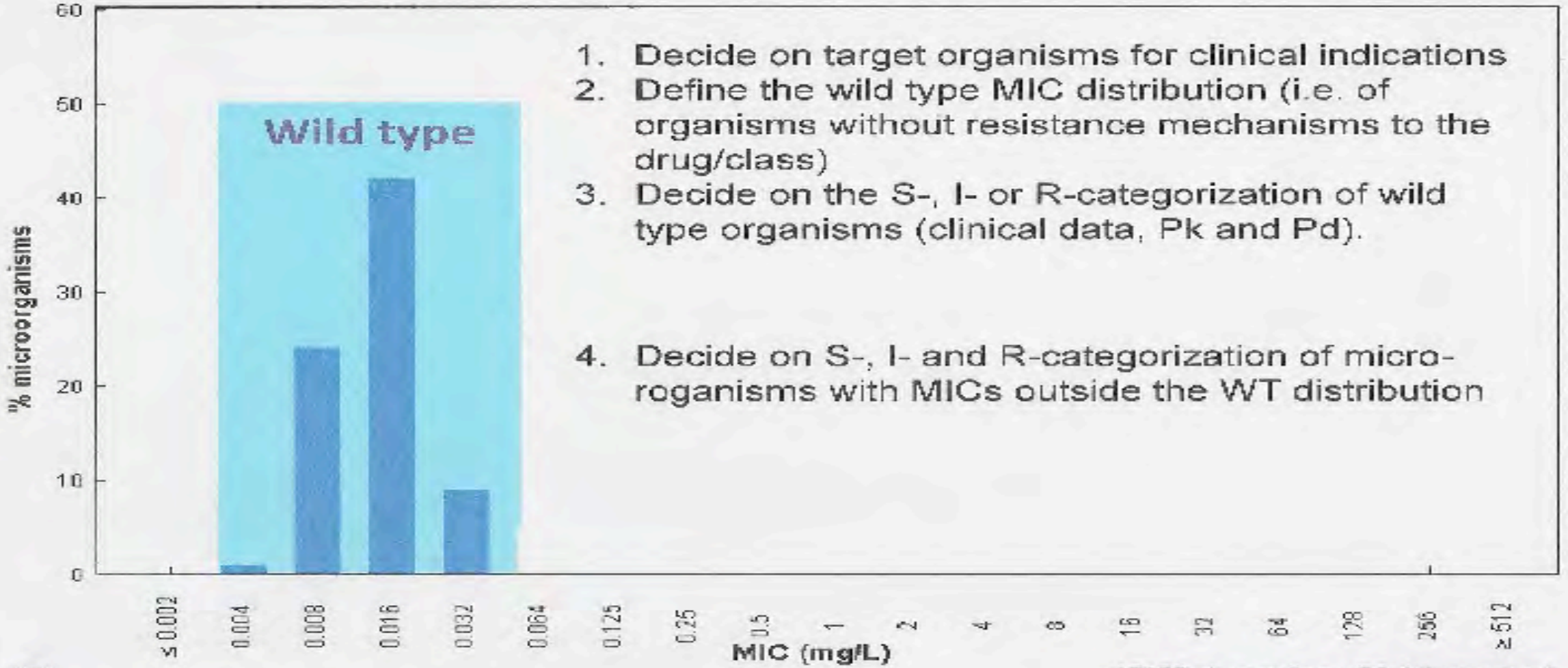
- Many tools are used to define antimicrobial breakpoints:
  - MIC distributions, ECOFFs, CBPs
  - Pharmacokinetic parameters
  - Dosing
  - Pharmacodynamic parameters
    - Target attainment, influence of neutrophils, protein binding etc.
- In the end, MICs are most often predictive of outcome.

Important PK/PD Questions.

- **Predictive Parameter** - What PK characteristic most efficiently improves or optimizes antimicrobial activity?
- **Magnitude of Parameter** - How much drug is needed?
- **Magnitude Variables** – What factors impact how much drug is needed?
- **Correlation in Humans** – Does this predict outcome in clinical disease?

Ciprofloxacin / Escherichia coli
EUCAST MIC Distribution - Reference Database

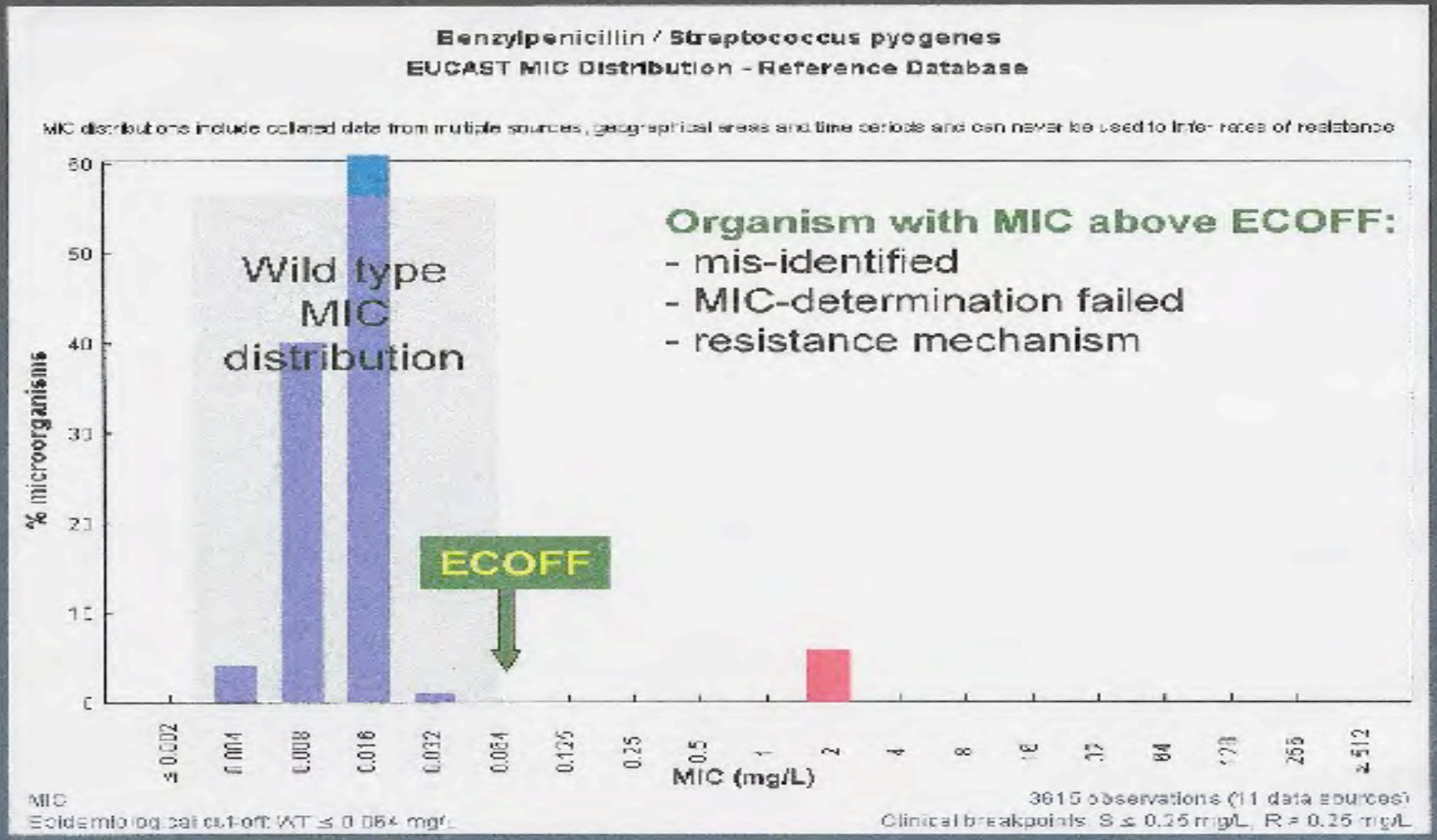
MIC distributions include collated data from multiple sources, geographical areas and time periods and can never be used to infer rates of resistance



1. Decide on target organisms for clinical indications
2. Define the wild type MIC distribution (i.e. of organisms without resistance mechanisms to the drug/class)
3. Decide on the S-, I- or R-categorization of wild type organisms (clinical data, Pk and Pd).
4. Decide on S-, I- and R-categorization of micro-roganisms with MICs outside the WT distribution

MIC
Epidemiological cut-off: WT ≤ 0.032 mg/L

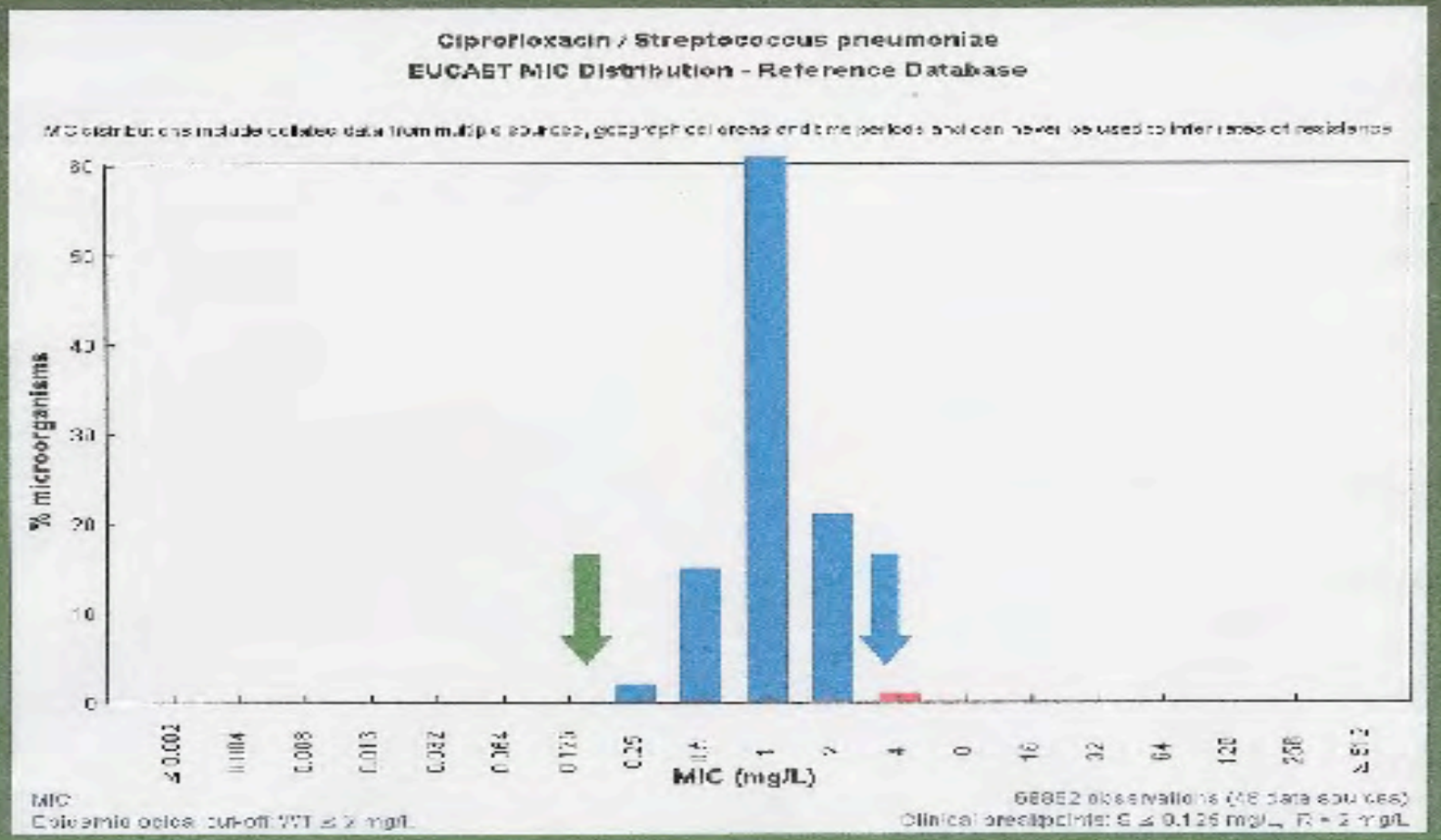
17977 observations (82 data sources)
Clinical breakpoints: S ≤ 0.5 mg/L, R > 1 mg/L



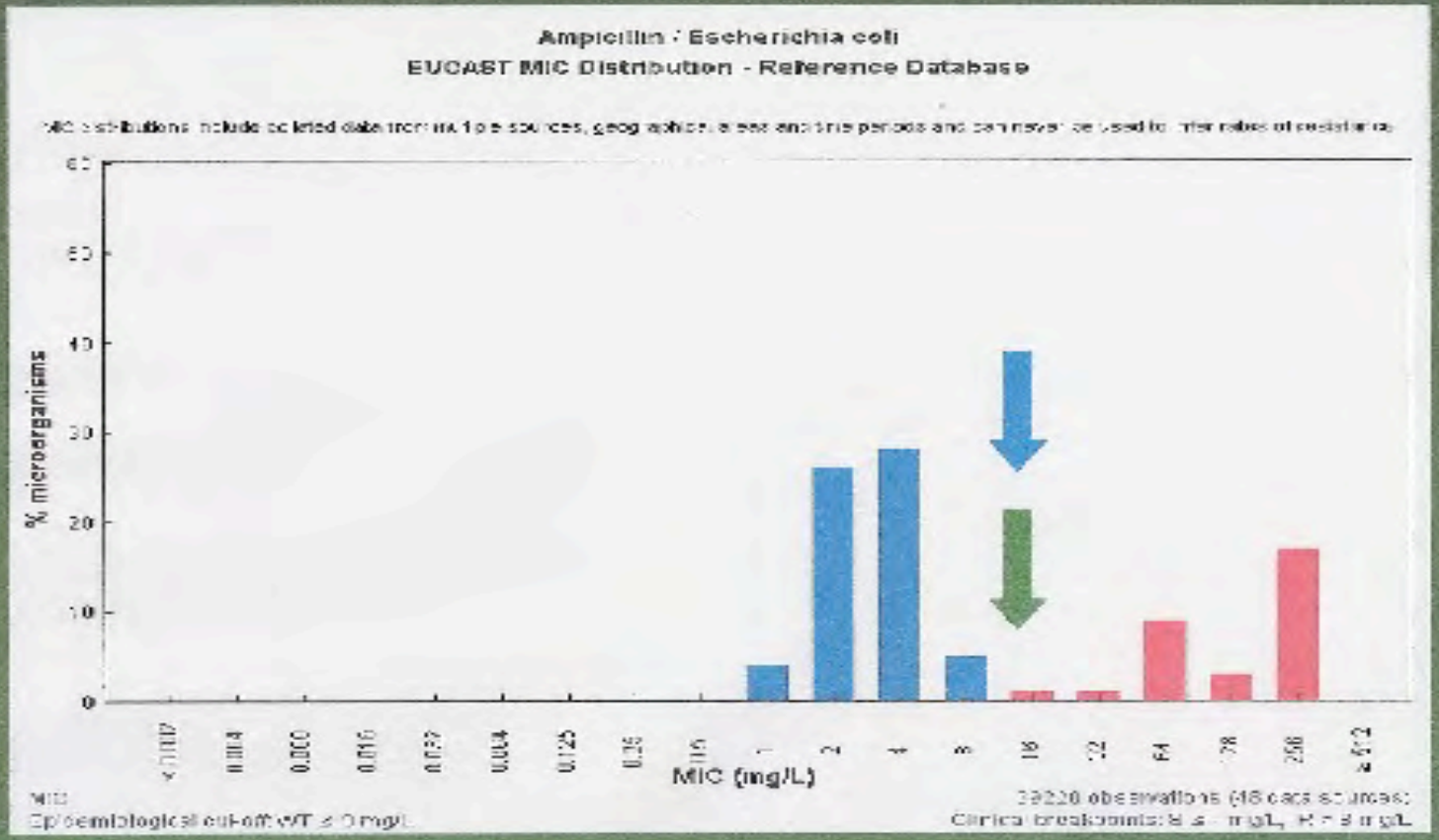
The ECOFF defines the upper end MIC of organisms without resistance!

CBP **lower** than ECOFF

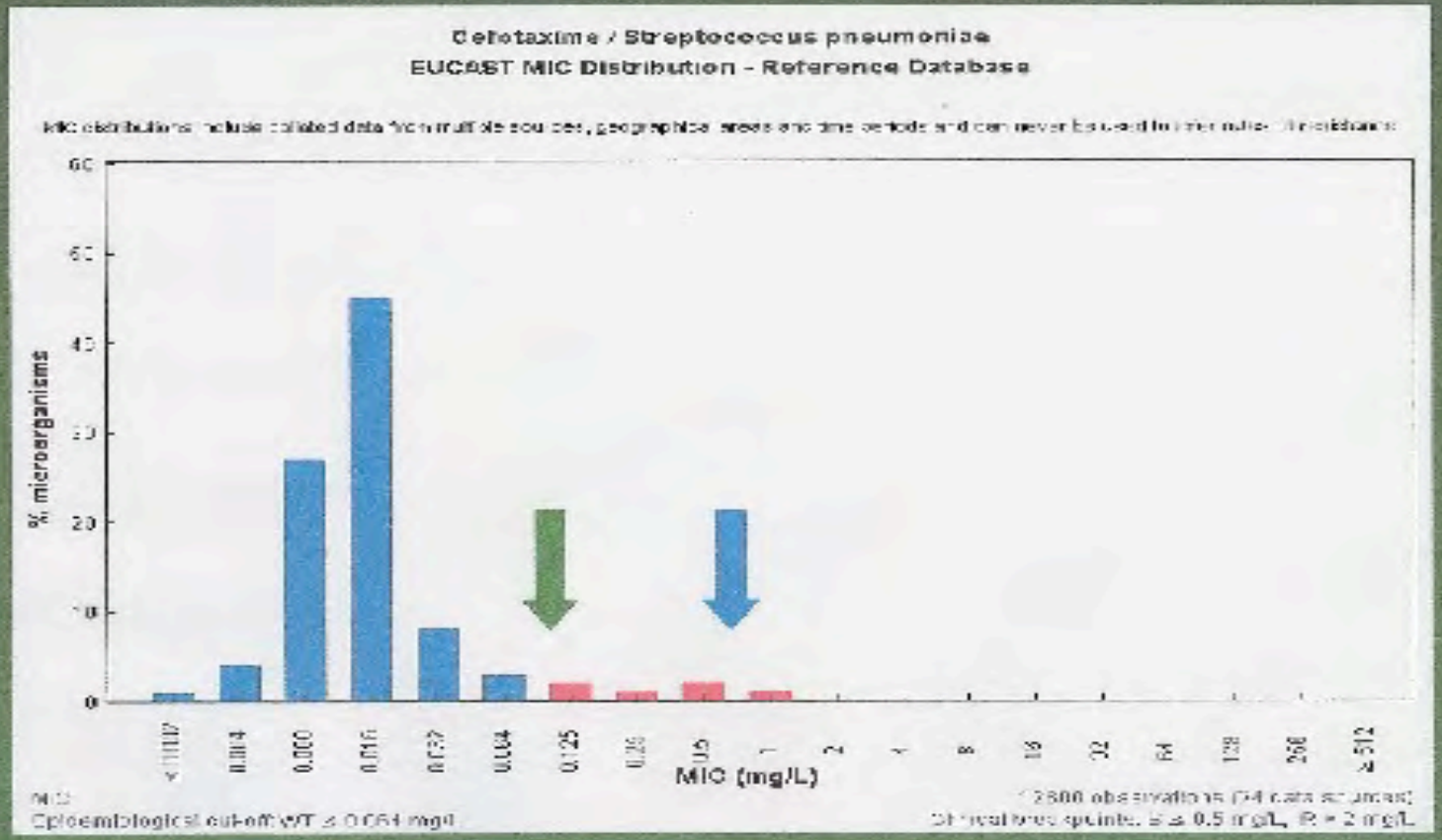
Ciprofloxacin v. *S. pneumoniae*



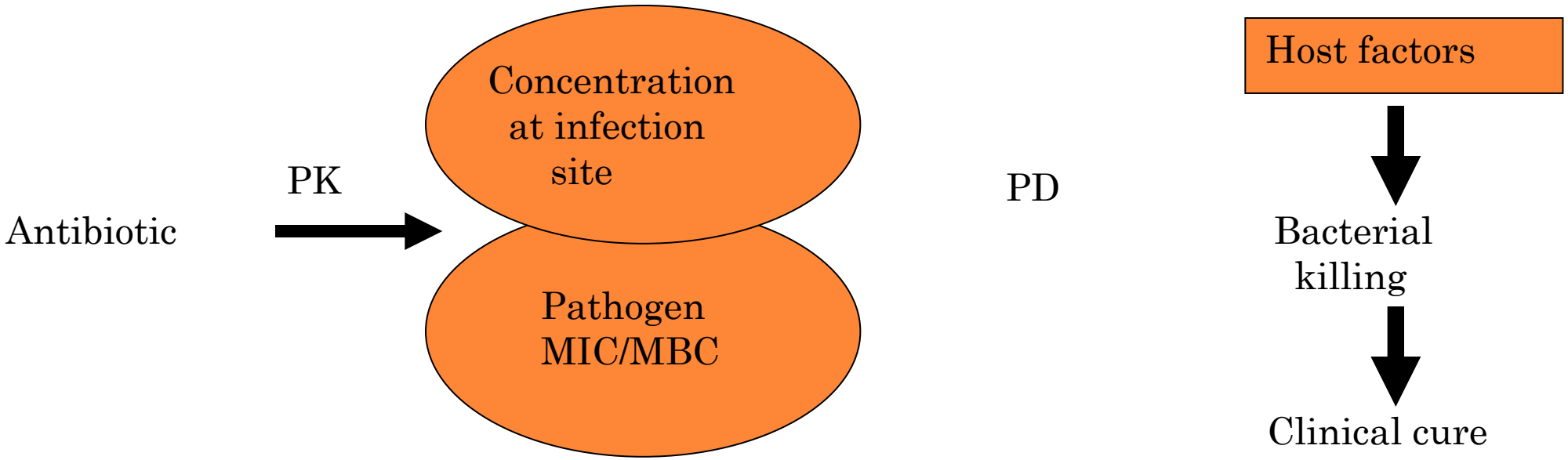
CBP = ECOFF
Ampicillin v. *E. coli*



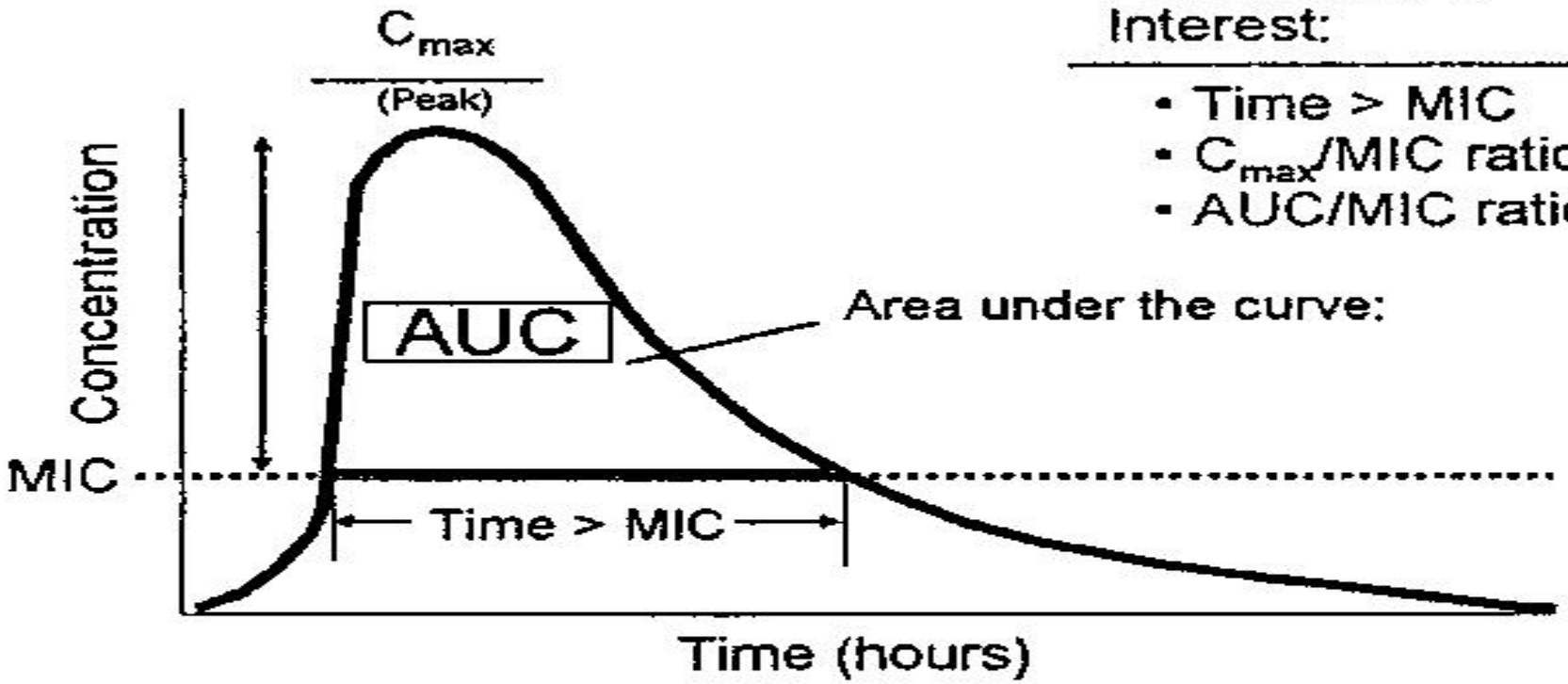
CBP **higher** than ECOFF
Cefotaxime v. *S. pneumoniae*



Optimizing Antimicrobial Therapy



Pharmacokinetics=



Parameters of Interest:

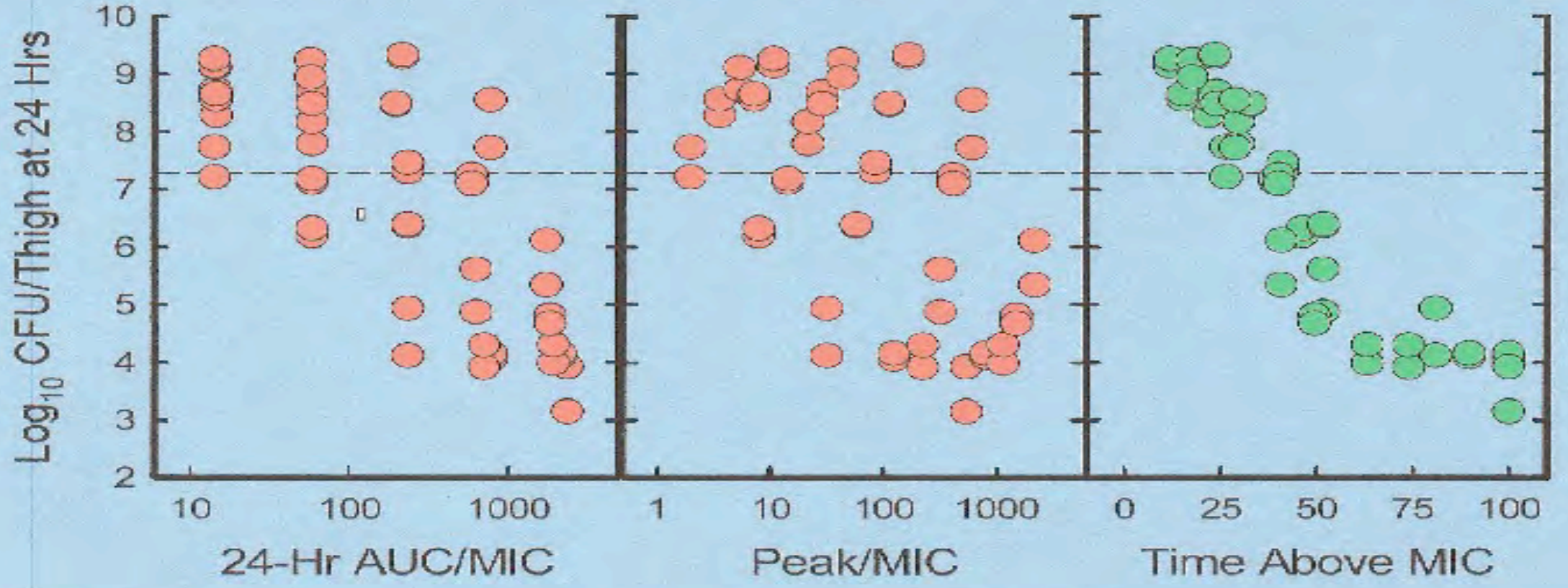
- Time > MIC
- C_{max}/MIC ratio
- AUC/MIC ratio

Area under the curve:

Pharmacodynamics/Pharmacodynamics=

Antimicrobial PK + MIC + Outcome

Relationship Between PK/PD Indices and Efficacy for Ceftazidime against *Klebsiella pneumoniae* in a Murine Pneumonia Model

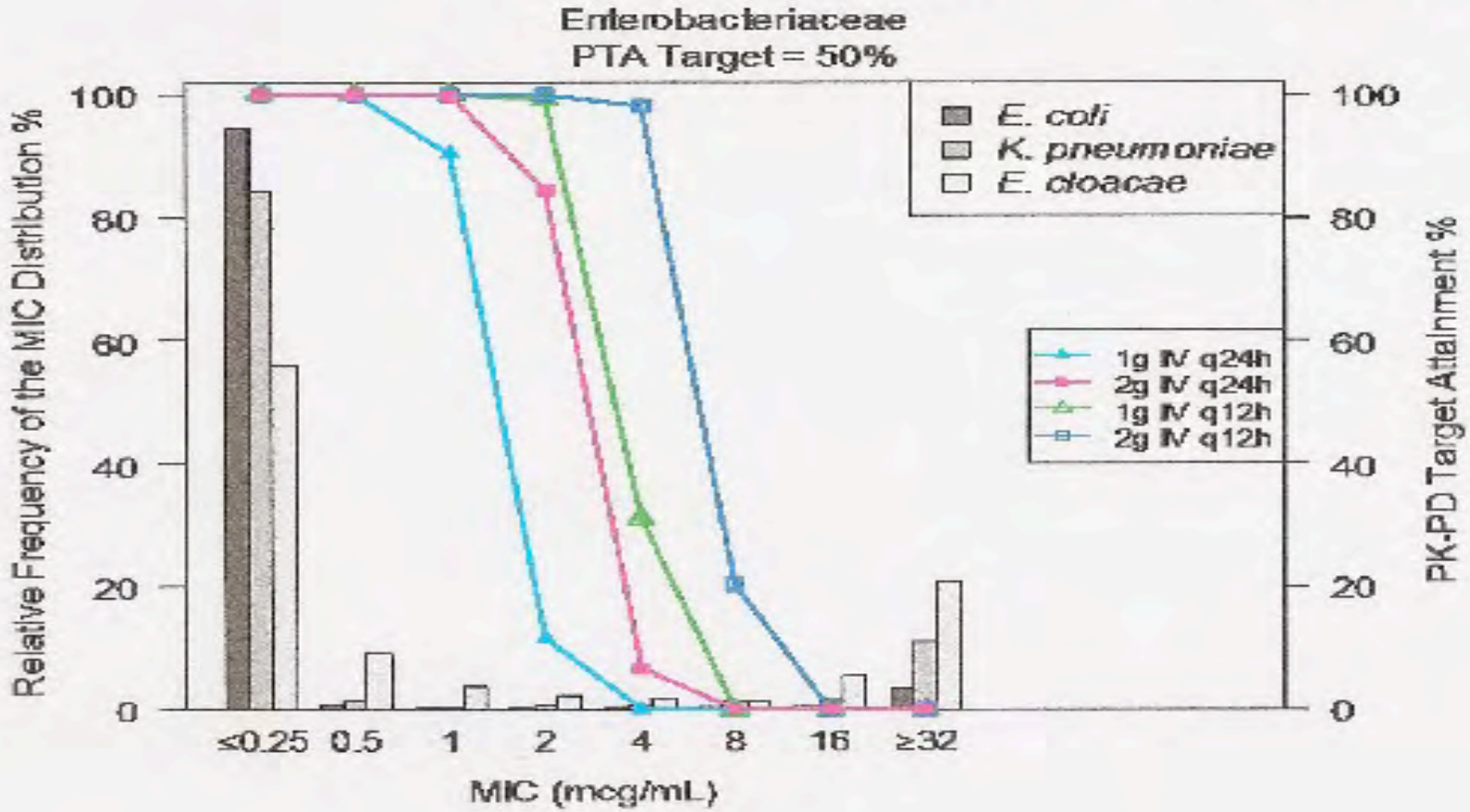


Craig WA: Pharmacodynamics of antimicrobials: General concepts and applications. In: Antimicrobial Pharmacodynamics in Theory and Clinical Practice, 2002:1-22.

Probabilities of PK-PD target attainment for various micro-organisms treated with ceftriaxone.

RESULTS

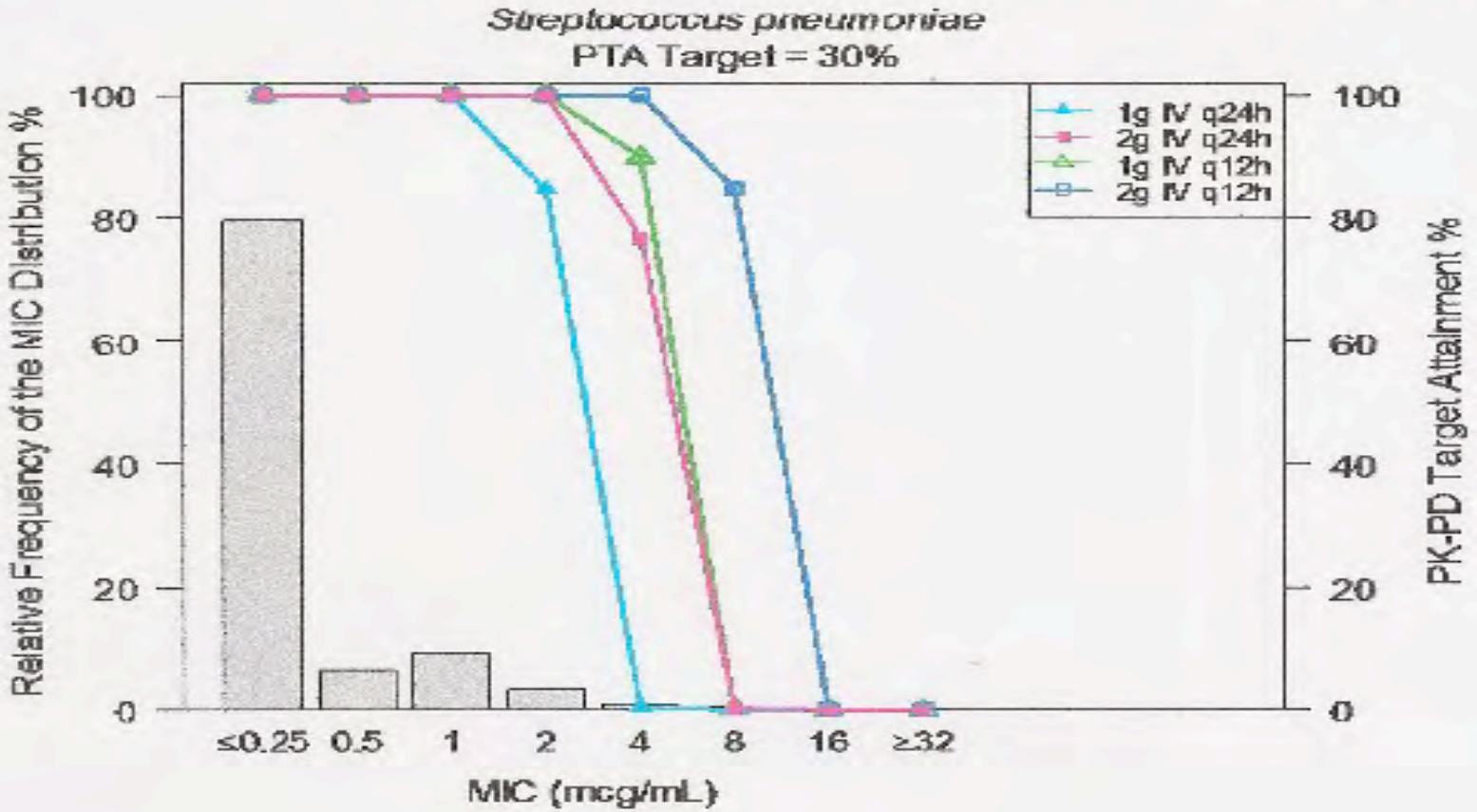
Probability of PK-PD Target Attainment, Enterobacteriaceae



Data from ceftriaxone presentation to CLSI 2012.

RESULTS

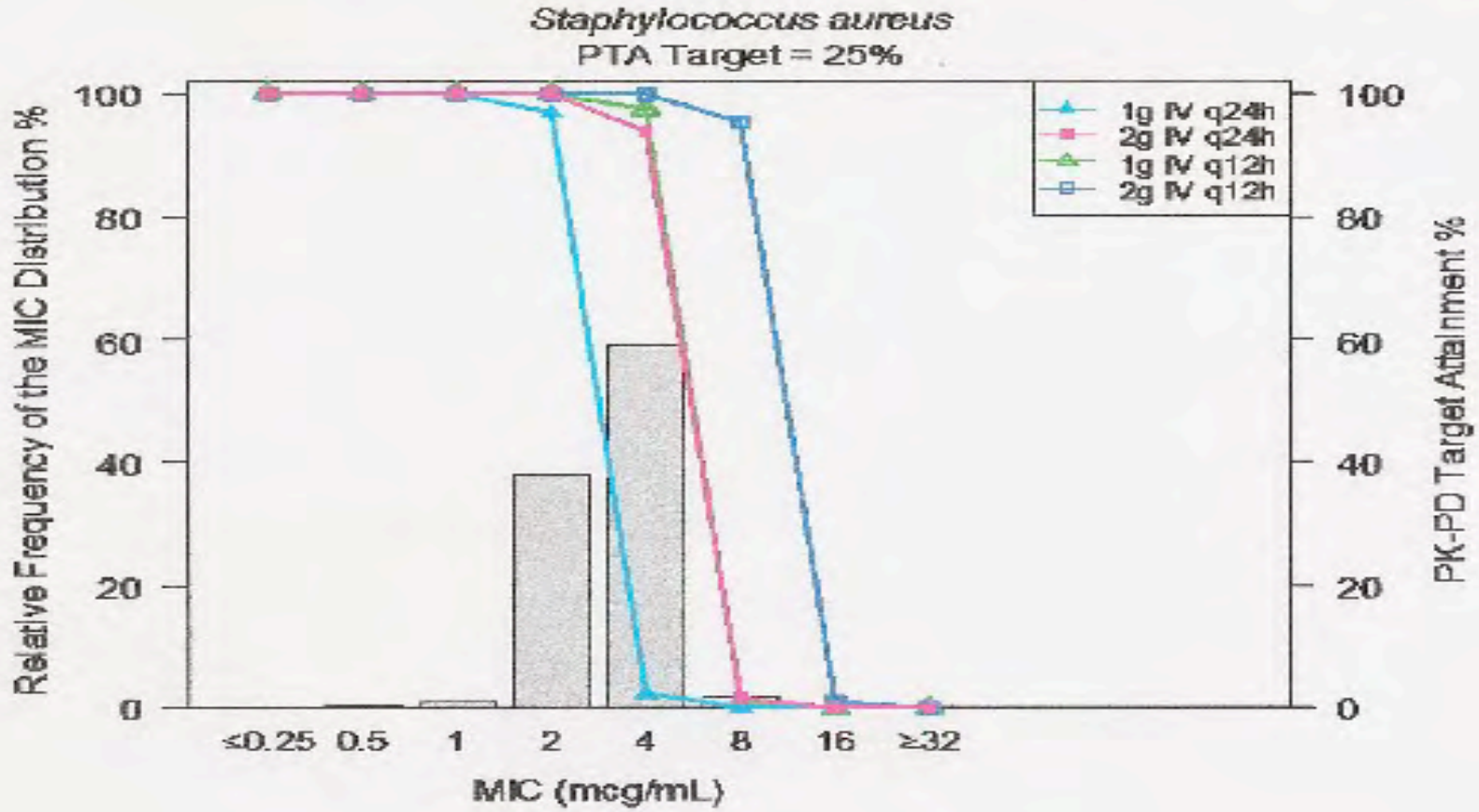
Probability of PK-PD Target Attainment, Streptococcus pneumoniae



Data from ceftriaxone presentation to CLSI 2012.

RESULTS

Probability of PK-PD Target Attainment, Staphylococcus aureus

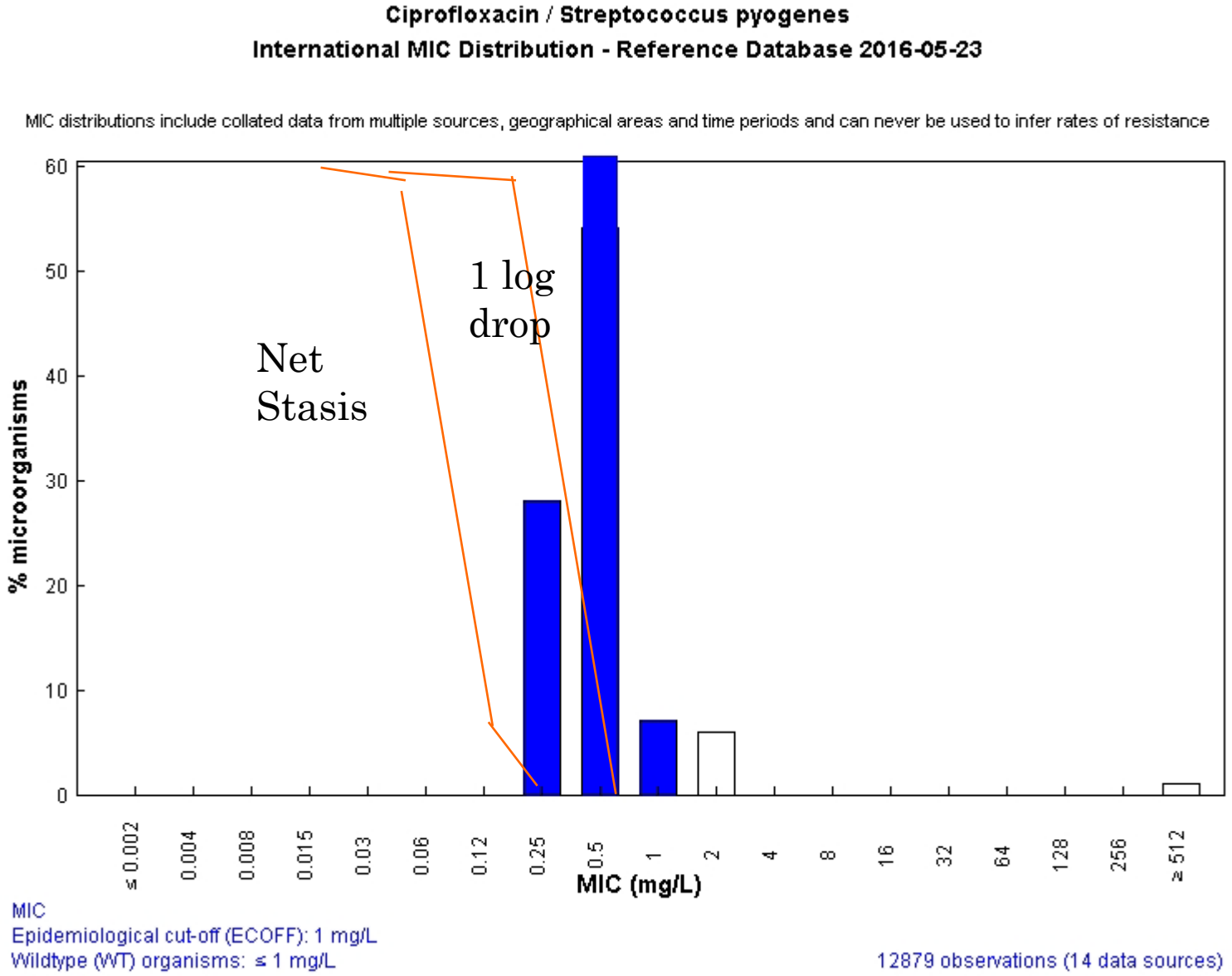


Data from ceftriaxone presentation to CLSI

Another clinical example: ciprofloxacin and beta-haemolytic streptococci

- Middle-aged female patient with extensive cellulitis in the thigh that required fasciotomy and debridement. Primary treatment with intravenous penicillin. Not thought to be necrotizing fasciitis.
- Infection caused by *Streptococcus pyogenes*.
- Discussion about sending patient home on oral ciprofloxacin.
- FDA breakpoints are ≤ 1 – S; 2 – I, ≥ 4 – R.

Target attainment for ciprofloxacin and *S. pyogenes*: 400 mg q12h IV – Net stasis and 1 log CFU decline (Data from EUCAST and USCAST)



Ciprofloxacin and *S. pyogenes*.

- Antimicrobial susceptibility performed to determine MIC.
  - 0.5 mg/L by gradient diffusion endpoint.
- At MIC of 0.5 mg/L modeling (animal studies, Monte Carlo simulations, target attainment), shows that virtually no drug available at 0.5 mg/L even using one log decline in CFU.
- Treatment with ciprofloxacin (IV or oral) will be ineffective
- Outcome:
  - Patient remained in hospital on penicillin and clindamycin until resolution of infection.
  - CLSI, EUCAST, USCAST have no breakpoints for ciprofloxacin and *S. pyogenes*.
  - FDA, TPD are reviewing all the fluoroquinolones for revision of breakpoints.

The importance of global harmonization.

- Same in vitro laboratory testing methodologies. International and ISO based.
- Clear definitions of wild-type strains and species.
- Clear definitions of susceptible, intermediate and resistant.
- Using all the parameters necessary to define a susceptible strain (It's the MIC s-----!).PK, PD, target attainment, dosing, mechanisms of resistance, phenotype, etc.
- Not setting a breakpoint when it is meaningless (e.g. target cuts the wild-type population)
- Provision of a standard base to define and conduct antimicrobial resistance surveillance (singing from the same song book!).
- Establishing a collection of National Antibiotic Committees (NACs) to ensure that these tenets are practiced in each country and globally

EUCAST – The mother ship.

- Created approximately 15 years ago to answer the questions about standardized susceptibility testing and resistance in Europe.
- Previously, Germany (DIN), France (SMF), Sweden(SRGA), UK (BSAC), and others all did their own thing.
- At about the same time Mueller Hinton medium became a global susceptibility medium "standard" – mainly so that isolates from clinical trials tested in various European countries wouldn't have to be repeated for FDA NDAs. Noteworthy – BSAC just recently changed to EUCAST methodology.
- EUCAST now has under it's purview almost all European countries. All providing data, participating in susceptibility development, resistance surveillance – AND – all using the same criteria.

What needs to be done to implement global harmonization in Canada.

- A National Committee to interact with stakeholders and laboratories.
- Buy in from accreditation bodies and linkage with regulatory agencies (drugs and devices)
- Stable ongoing financing for promotion, maintenance, breakpoint evaluations, etc.
- Website.
- Availability and continued rapid updating of breakpoint and quality control tables for laboratories that are free and current.
- Canadian based documents for methodologies: provision of rationale documents to support breakpoints.
- Interaction with antimicrobial resistance surveillance and stewardship groups.

EUCAST – The Mother Ship

- Other countries outside Europe with NACS
  - Australia
  - Brazil
  - Canada
  - Estonia
  - Morocco
  - South Africa
  - Ukraine
  - United States
  - China

Canadian Committee on Antimicrobial Susceptibility Testing (CANCAST)

CANCAST (Canadian Antimicrobial Susceptibility Testing Committee) was established in 2016 with administrative support from Canadian Standards Association (CSA) to provide expert advice in the area of antimicrobial susceptibility testing to Canadian clinical laboratories. A variety of standards and guidelines have been used in Canadian laboratories.

Canadian Committee on Antimicrobial Susceptibility Testing (CANCAST)

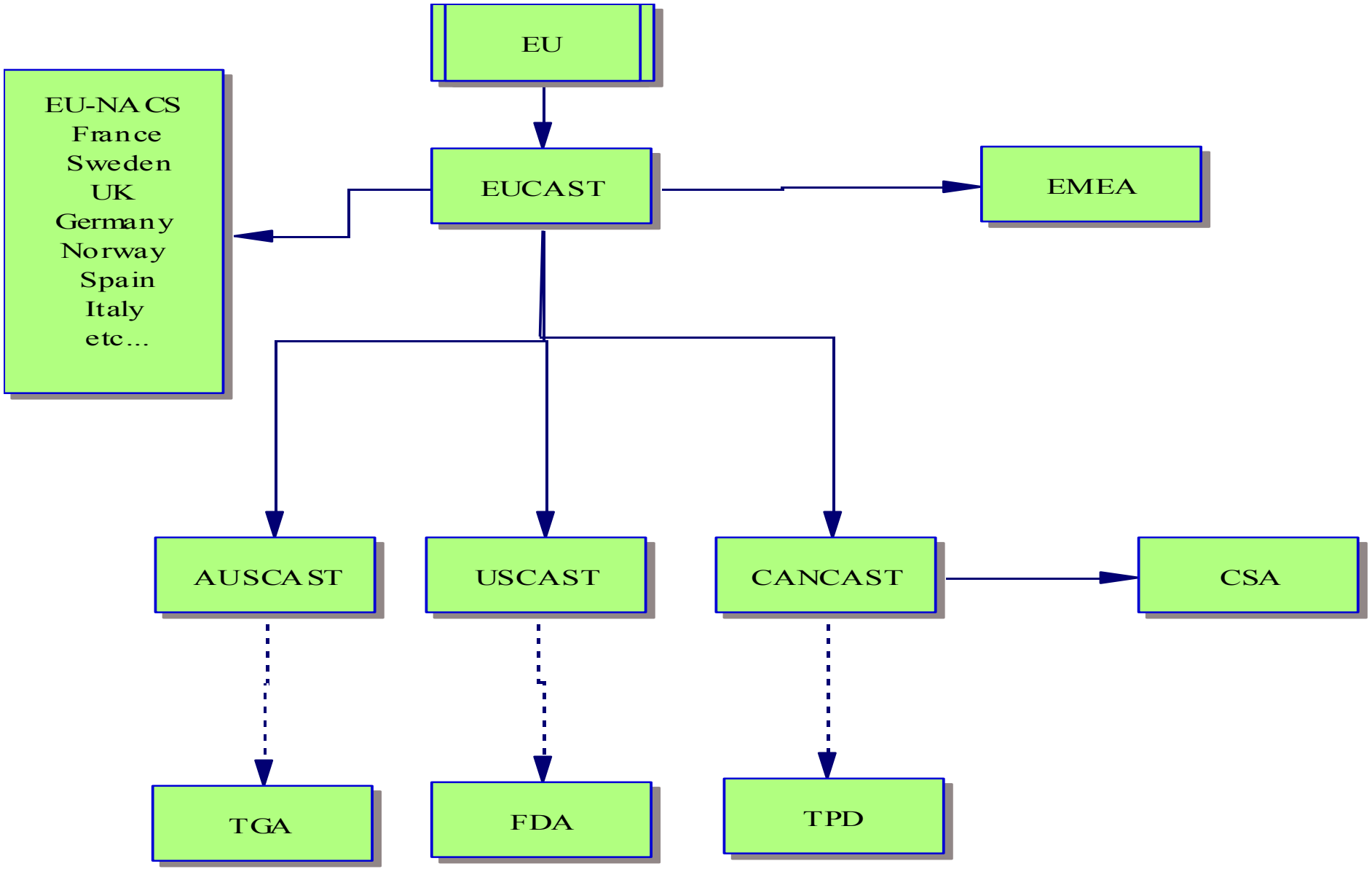
CANCAST is composed at present of an Executive Committee with medical and clinical microbiology and infectious diseases experts and ex officio involvement of the Therapeutics Products Directorate (TPD) of Health Canada. Advisors, representing medical microbiology and infectious diseases experts, national organizations (e.g. AMMI-CACMID) provincial jurisdictions, the Public Health Agency of Canada, National Microbiology Laboratory, external quality assurance agencies, provincial laboratory accreditation bodies, and susceptibility test manufacturers are being recruited to provide encompassing support for CANCAST. The current Chair of CANCAST is Dr. Bob Rennie, from Edmonton, Alberta.

CANCAST Goals and Objectives.

The goals of CANCAST are to:

- Provide standardized antimicrobial susceptibility testing methods and breakpoints for Canadian laboratories (primarily for anti-bacterial and anti-fungal testing) that are consistent with global standards (EUCAST) where available.
- Provide advice to groups conducting resistance surveillance, on laboratory methods, testing concentrations, and on interpretation of data relevant to the type of resistance surveillance being undertaken.
- Provide educational workshops on basic and advanced aspects of susceptibility testing.
- Interact with TPD in Canada and other antimicrobial standards groups (EUCAST, CLSI) to ensure that methodologies and breakpoints are harmonized internationally.

National Antibiotic Committee Structure



Disk diffusion (EUCAST and CLSI standardised disk diffusion method)
Medium: Mueller-Hinton agar
Inoculum: McFarland 0.5
Incubation: Air, 35±1ºC, 18±2h
Reading: Read zone edges as the point showing no growth viewed from the back of the plate against a dark background illuminated with reflected light.
Quality control: *Escherichia coli* ATCC 25922

Penicillins[1]	**MIC breakpoint (µg/ml)**		**Disk content (µg)**	**Zone diameter breakpoint (mm)**		**Notes** Numbers for comments on MIC breakpoints Letters for comments on disk diffusion
	S ≤	**R ≥**		**S ≥**	**R ≤**	
Penicillin	-	-		-	-	**1/A.** Wild type Enterobacteriaceae are categorised as susceptible to aminopenicillins. **2.** For susceptibility testing purposes, the concentration of sulbactam is tested at a 2:1 ratio (ampicillin:sulbactam) per USA criteria. **3.** For susceptibility testing purposes, the concentration of tazobactam is fixed at 4 µg/ml. **4.** For susceptibility testing purposes, the concentration of clavulanic acid is fixed at 2 µg/ml. **B.** Ignore growth that may appear as a thin inner zone on some batches of Mueller-Hinton agar. **C.** Susceptibility inferred from ampicillin.
Ampicillin	8[1]	16	**10**	14[A,B]	13[B]	
Ampicillin-sulbactam	8[1,2]	16[2]	**10-10**	14[A,B]	13[B]	
Amoxicillin	8[1]	16	**-**	Note[C]	Note[C]	
Amoxicillin-clavulanic acid	8[1,2]	16[2]	**20-10**	19[A,B]	18[B]	
Piperacillin	IP	IP	**100**	IP	IP	
Piperacillin-tazobactam	IP[3]	IP[3]	**100-10**	IP	IP	
Ticarcillin	IP	IP	**75**	IP	IP	
Ticarcillin-clavulanic acid	IP[4]	IP[4]	**75-10**	IP	IP	
Oxacillin	-	-		-	-	

Report Date: June 23, 2015
Report Number: USCAST 0001
Version: 1.0

USCAST
THE NATIONAL ANTIMICROBIAL SUSCEPTIBILITY TESTING COMMITTEE FOR THE UNITED STATES

Report Title

Quinolone *In Vitro* Susceptibility Test Interpretive Criteria Evaluations

Antimicrobial Agents

Ciprofloxacin, Levofloxacin, Moxifloxacin, Nalidixic Acid, Norfloxacin, and Ofloxacin

Organisms

Enterobacteriaceae, *Pseudomonas aeruginosa*, *Staphylococcus aureus*, *Streptococcus pneumoniae*, and Other Pathogens

USCAST

Ronald N. Jones, M.D., FIDSA, FCAP, FASCP, FAAM
USCAST Scientific Secretary

Paul G. Ambrose, Pharm.D., FIDSA
USCAST Chairman

This report should be cited as "USCAST, The National Antimicrobial Susceptibility Testing Committee for the United States. Quinolone In Vitro Susceptibility Test Interpretive Criteria Evaluations. Version 1.0, 2015. http://ww.uscast.org

CANCAST Progress

- Executive Committee established.
  - Includes, medical and clinical microbiologists, pharmacologists, regulators, antimicrobial resistance specialists (similar to other NACs).
- Terms of Reference and Business Plan are being completed.
- Health Canada has been engaged to discuss sustainability
- Working on a Website
- General Committee Members and Advisors will be recruited.
- Participation in EUCAST discussions regarding breakpoints, methodology.

Next Steps.

- Once Website developed:
  - Conduct cross-Canada workshops with laboratories regarding use of global breakpoints in clinical laboratories.
  - Work with regulators (national and provincial accreditation bodies) to use CANCAST as the primary antimicrobial testing and reporting system for clinical microbiology laboratories.
  - Work with TPD and Device manufacturers in Canada, and with EUCAST so that breakpoints for new agents are the same, and that important changes to existing breakpoints can be made in a timely manner – not 4-5 years after the need is identified.
  - Provide laboratories and clinicians with freely accessible clinical breakpoint tables and rationale documents that clearly detail how the latest antimicrobial breakpoints were arrived at.

Summary and Benefits

- Laboratories can download and print methodology and breakpoint tables free of charge.
- Laboratories are assured that the same breakpoints are being reported in laboratories in major centres across the globe based on the same testing methodologies.
- Accreditation of susceptibility testing and reporting would be based on systems which have global standards with Canadian input.
- Through CANCAST, laboratories have the ability to provide input into issues with methodology, with testing and reporting issues.
- Antimicrobial resistance surveillance can be standardized globally