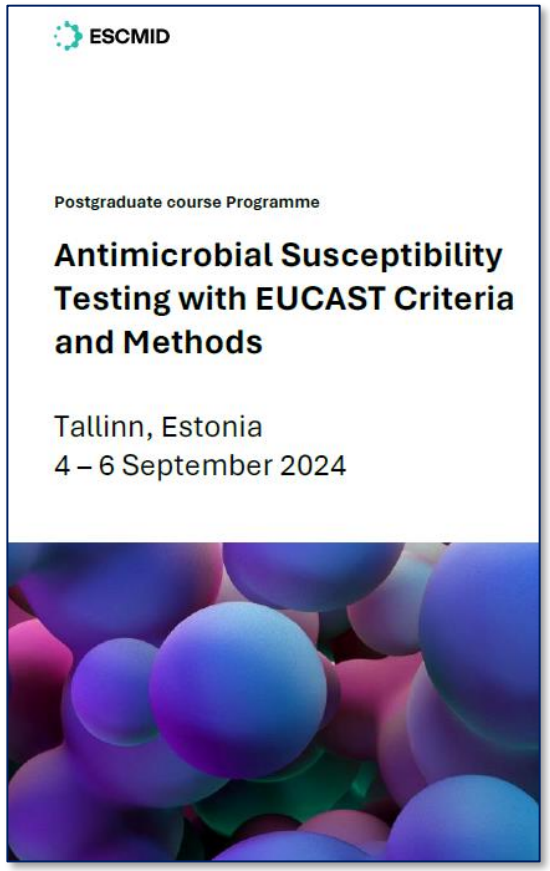


EUCAST EUROPEAN COMMITTEE ON ANTIMICROBIAL SUSCEPTIBILITY TESTING
European Society of Clinical Microbiology and Infectious Diseases

Expected resistant phenotypes, expected susceptible phenotypes and expert rules

RYC MICROBIOLOGÍA HOSPITAL RAMÓN Y CAJAL

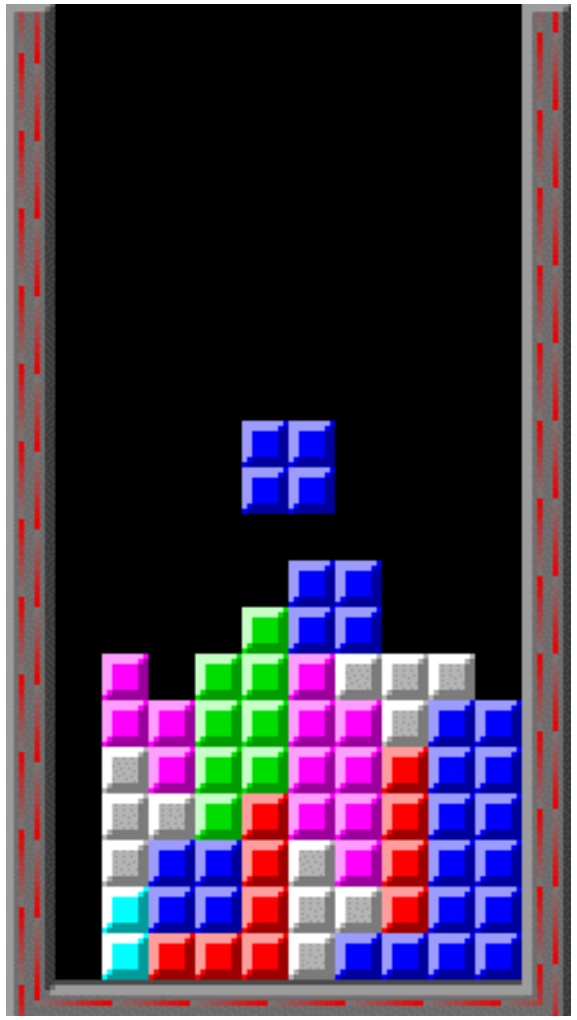
SaludMadrid
Dr. Rafael Cantón
Hospital Universitario Ramón y Cajal
SERVICIO DE MICROBIOLOGÍA Y PARASITOLOGÍA

instituto ramón y cajal de investigación sanitaria irycis

@RafaMCanton
@microRyC

ciberinfec isciii

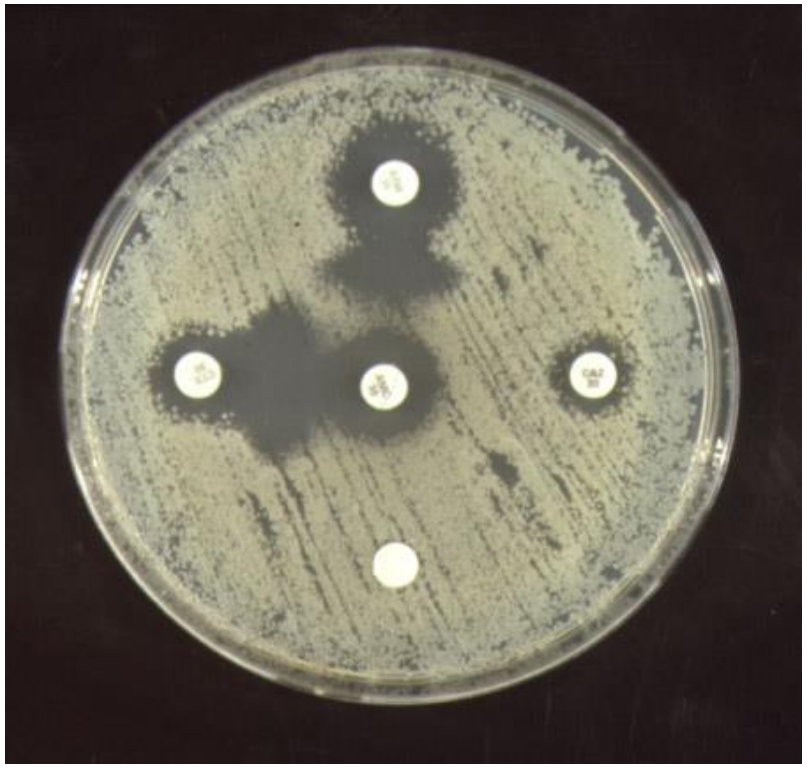
Departamento de Microbiología y Parasitología Universidad Complutense. Madrid



- **Clinical categorization (S, I, R)**
  - Based on clinical breakpoints (*Breakpoint tables*)
- **Interpretive reading**
  - Based on resistance mechanisms knowledge (*Guidelines on detection of resistance mechanisms*)
- **Application of expert rules**
  - Based on clinical evidence, microbiological data and resistance mechanisms knowledge (*Expected phenotypes and expert rules*)

Interpretive reading*

- *The classical example*



Escherichia coli
ESBL producer

- No longer modification of AST results (*report as tested*)
- Alert of the resistance mechanisms for infection control and epidemiological purpose

*Courvalin P. ASM News 19921992;58:368-75;Livermore DM et al. J Antimicrob Chemother 2001;48(Suppl 1):87-102; Cantón R. Enferm Infecc Microbiol Clin 2002; 20: 176-86; Cantón R. Enferm Infecc Microbiol Clin 2010; 28:375-85; Winstanley T, Courvalin P. Clin Microbiol Rev 2011; 24: 515–56; Leclercq R et al. Clin Microbiol infect 2013; 19:141-60

Klebsiella pneumoniae

Antibiotic	MIC (mg/L)	Interpre-tation
Amoxicillin	>16	R
Amoxi-clav	≤4/2	S
Piper-tazo	≤8/4	S
Cefuroxime	≤0.5	S
Cefotaxime	≤0.06	S
Ceftazidime	≤0.06	S
Cefepime	≤0.06	S
Aztreonam	≤0.06	S
Ceftol-Tazo	≤0.5/4	S
Cefta-avib	≤0.5/4	S
Ertapenem	≤0.5	S
Imipenem	≤0.5	S
Meropenem	≤0.5	S

Wild type

Antibiotic	MIC (mg/L)	Interpre-tation
Amoxicillin	>16	R
Amoxi-clav	≤4/2	S
Piper-tazo	≤8/4	S
Cefuroxime	>16	R
Cefotaxime	>16	R
Ceftazidime	2	I
Cefepime	0.5	S
Aztreonam	0.5	S
Ceftol-Tazo	1/4	S
Cefta-avib	1/4	S
Ertapenem	2	R
Imipenem	≤0.5	S
Meropenem	≤0.5	S

ESBL

Antibiotic	MIC (mg/L)	Interpre-tation
Amoxicillin	>16	R
Amoxi-clav	>16/8	R
Piper-tazo	>64/4	R
Cefuroxime	>16	R
Cefotaxime	>16	R
Ceftazidime	>16	R
Cefepime	>16	R
Aztreonam	>4	R
Ceftol-Tazo	>8/4	S
Cefta-avib	4/4	S
Ertapenem	>8	R
Imipenem	<8	R
Meropenem	8	I

Carbapenemase

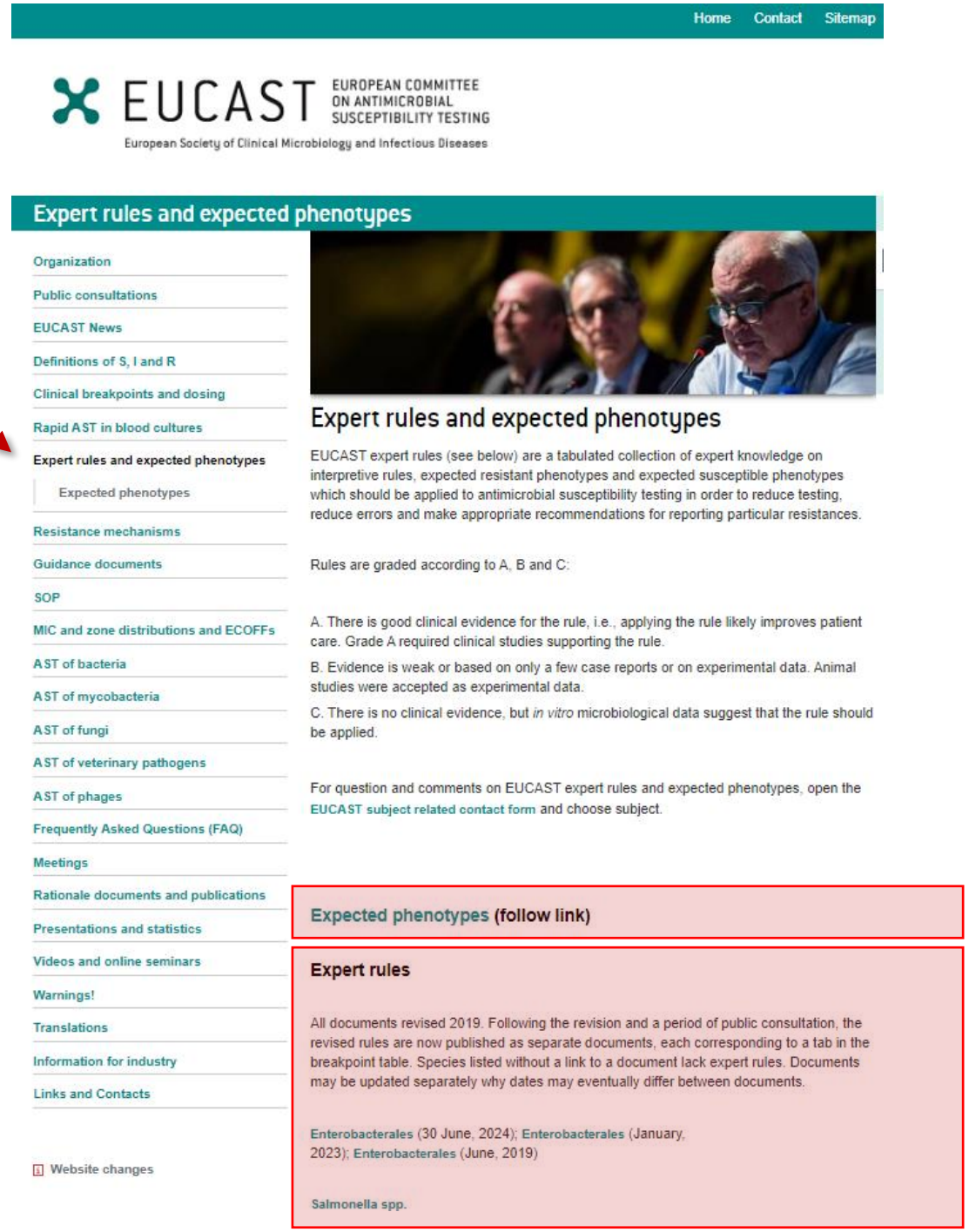
EUCAST, 2024 interpretive criteria (www.eucast.org)

Expert rules and expected phenotypes

To reduce AST testing

To reduce errors

To make appropriate recommendations for reporting particular resistances



Expert rules and expected phenotypes over time

	Document (version)	Content
2008, April 2011, October 2016, September	**Expert rules in antimicrobial susceptibility testing** (v1.0, v2.0, 3,1)	**Intrinsic resistances** **Exceptional resistance phenotypes** **Interpretive / expert rules**
2019, June 2020, February	Intrinsic resistances and unusual phenotypes (v3.2)	Intrinsic resistances Unusual phenotypes
2019, June 2020, February **2023, January /February** **2024, June**	**Expert rules** **(v3.2, v3.3)**	**Expert rules**
2022, February **2022, March** **2023, January**	**Expected phenotypes** **(v1.0, v1.1, v1.2)**	**Expected susceptible phenotypes** **Expected resistant phenotypes**

From “intrinsic resistance” to “expected phenotypes”

EUCAST EUROPEAN COMMITTEE ON ANTIMICROBIAL SUSCEPTIBILITY TESTING

EUCAST Expert rules in antimicrobial susceptibility testing, version 1, April 2008

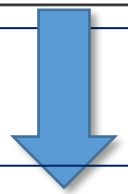
Intrinsic resistance: Inherent (not acquired) resistance which is a characteristic of all or almost all representatives of the species

- The **antimicrobial activity** of the drug is **insufficient** or **antimicrobial resistance innate** or so common as to render **it clinically useless** and **antimicrobial susceptibility testing unnecessary**
- Hence **“susceptible” results should be viewed with caution**, as they most likely indicate an **error in identification or susceptibility testing**. Even if susceptibility is confirmed the drug should be used with caution.
- Intrinsic resistance may be expressed at a low level (MIC close to the S breakpoint), although the antibiotic is not considered clinically active. When the antibiotic is fully active in vitro but in vivo inactive, this is not mentioned as it is a matter of therapeutic recommendations

From “intrinsic resistance” to “expected phenotypes”

EUCAST EUROPEAN COMMITTEE ON ANTIMICROBIAL SUSCEPTIBILITY TESTING

EUCAST Expert rules in antimicrobial susceptibility testing, version 1, April 2008



Exceptional resistance phenotypes

- Resistance of some bacterial species to particular antimicrobial agents has **not yet been reported or is very rare.**
- Exceptional resistance phenotypes should be checked as they **may indicate an error in identification or susceptibility testing**. If they are confirmed locally the isolate should be sent to a reference laboratory for independent confirmation.
- Exceptional resistance phenotypes **may change with time** as resistance may develop and increase over time. There may also be regional or national differences and a very rare resistance in one area may be more common in another.

From “intrinsic resistance” to “expected phenotypes”

REVIEW Clin Microbiol Infect. 2013; 19:141-60 10.1111/j.1469-0691.2011.03703.x

EUCAST expert rules in antimicrobial susceptibility testing

R. Leclercq[1,2], R. Cantón[2,3,4], D. F. J. Brown[4], C. G. Giske[2,4,5], P. Heisig[2,6], A. P. MacGowan[4,7], J. W. Mouton[4,8], P. Nordmann[2,9], A. C. Rodloff[4,10], G. M. Rossolini[2,11], C.-J. Soussy[4,12], M. Steinbakk[4,13], T. G. Winstanley[2,14] and G. Kahlmeter[4,15]

EUCAST EUROPEAN COMMITTEE ON ANTIMICROBIAL SUSCEPTIBILITY TESTING
European Society of Clinical Microbiology and Infectious Diseases

EUCAST Expert Rules Version 3.1

September 2016

Intrinsic Resistance and Exceptional Phenotypes Tables

EUCAST EUROPEAN COMMITTEE ON ANTIMICROBIAL SUSCEPTIBILITY TESTING
European Society of Clinical Microbiology and Infectious Diseases

Intrinsic Resistance and Unusual Phenotypes version 3.2

February 2020

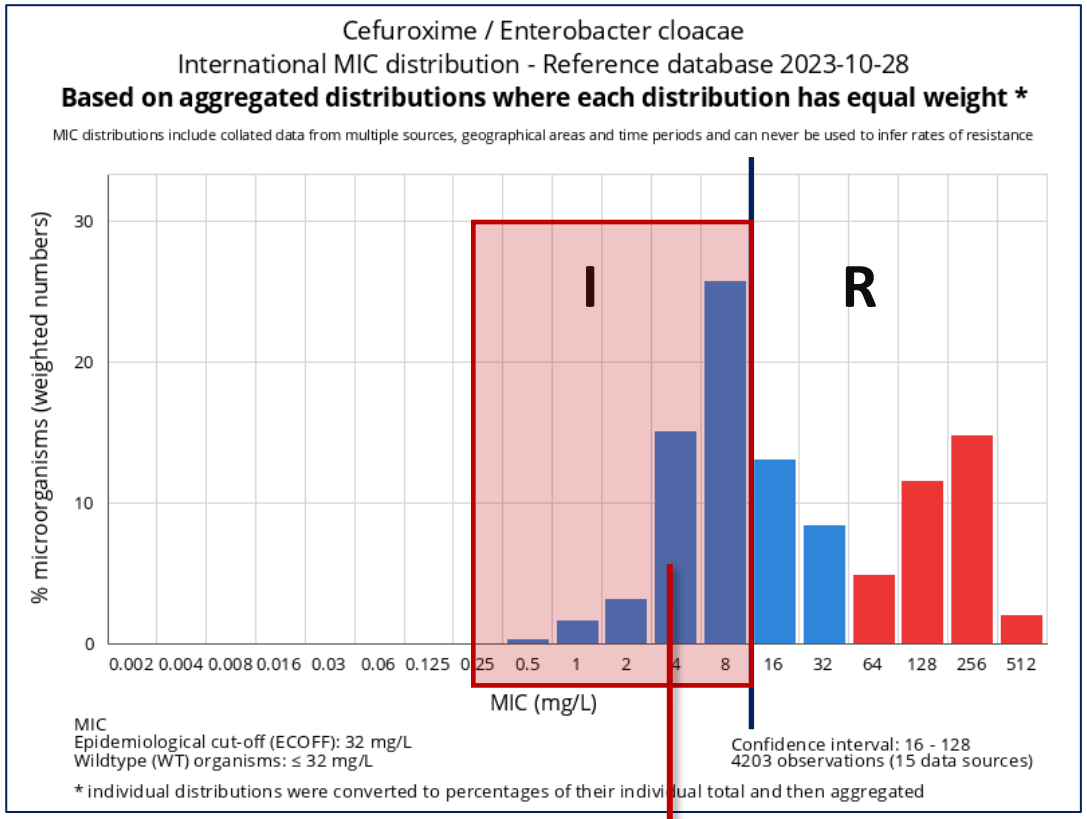
From “intrinsic resistance” to “expected phenotypes”

EUCAST Intrinsic Resistance & Unusual Phenotypes v 3.2 February 2020

- **Intrinsic Resistances** and **Unusual Phenotypes** a **tool for the validation of species identification and/or AST**
- The **absence of intrinsic resistance or an unusual phenotype** in isolates with these expected results indicates that the species identification, the AST or both should be corroborated
- **Microorganisms are only listed as “intrinsically resistant”** when a **vast majority of wild-type isolates exhibit MIC values are high** and **the agent should not be considered for either therapy or clinical susceptibility testing**
- If a significant proportion of the organisms have **MICs below the R breakpoint of species generally susceptible to the agent**, it is not listed as intrinsically resistant. **If the drug is not recommended an expert rule is applied**

e.g. *Enterobacter cloacae* complex and cefuroxime

From “intrinsic resistance” to “expected phenotypes”



Rule	Organisms	Ampicillin/Amoxicillin	Amoxicillin-clavulanic acid	Ampicillin-sulbactam	Ticarcillin	Cefazolin, Cephalothin Cefalexin, Cefadroxil	Cefoxitin[2]	Cefuroxime	Tetracyclines	Tigecycline	Polymyxin B, Colistin	Fosfomycin	Nitrofurantoin
1.1	*Citrobacter koseri, Citrobacter amalonaticus*[3]	R			R								
1.2	*Citrobacter freundii*[4]	R	R	R		R	R						
1.3	*Enterobacter cloacae* complex	R	R	R		R	R						
1.4	*Escherichia hermannii*	R			R								
1.5	*Hafnia alvei*	R	R	R		R	R				R		
1.6	*Klebsiella aerogenes*	R	R	R		R	R						
1.7	*Klebsiella oxytoca*	R			R								
1.8	*Klebsiella pneumoniae* complex[5]	R			R								
1.9	*Leclercia adecarboxylata*											R	
1.10	*Morganella morganii*	R	R	R		R			R		R		R
1.11	*Plesiomonas shigelloides*	R	R	R									
1.12	*Proteus mirabilis*								R	R	R		R
1.13	*Proteus penneri*	R				R		R	R	R	R		R

Expert rule IF susceptible to cefuroxime, THEN report cefuroxime and/or any other 2nd generation cephalosporin as resistant

From “intrinsic resistance” to “expected phenotypes”

When preparing in 2022 a new version of intrinsic resistance phenotypes document and during the review of its publication at CMI, the editor requests to include a definition of intrinsic resistance in the submitted manuscript…*

- No agreed definition of the term **“intrinsic resistance”**
  - Not always associated with the presence of a resistance gene (not always expressed)
  - Difficult in light of “exposure dependent” definition of breakpoints (might be modified with dosage regimens)
- New **“expected phenotype”** definitions
  - Closer to routine antimicrobial susceptibility testing (AST)
  - Allows to report the isolate as resistant or susceptible without performing an AST test
  - Alert inconsistent identification

*Gatermann S, Das S, Dubreuil L, Giske CG, Kahlmeter G, Lina G, Lindemann C, MacGowan A, Meletiadis J, Rossolini GM, Turnidge J, Cantón R. Expected phenotypes and expert rules are Important complements to antimicrobial ausceptibility testing. Clin Microbiol Infect. 2022 Mar 16:S1198-743X(22)00146-X.

From “intrinsic resistance” to “expected phenotypes”

Clinical Microbiology and Infection 28 (2022) 764–767

Contents lists available at ScienceDirect

Clinical Microbiology and Infection

journal homepage: www.clinicalmicrobiologyandinfection.com

Commentary

Expected phenotypes and expert rules are important complements to antimicrobial susceptibility testing

Sören Gatermann [1,*], Shampa Das [2], Luc Dubreuil [3], Christian G. Giske [4], Gunnar Kahlmeter [5], Gerard Lina [6], Christoffer Lindemann [7], Alasdair MacGowan [8], Joseph Meletiadis [9], Gian Maria Rossolini [10], John Turnidge [11], Rafael Cantón [12]

Expected resistant phenotype → *Klebsiella pneumoniae* and ampicillin

- ≥ 90% of population show MIC >PK/PD resistant (R) breakpoint
- Tables show R only, if this condition is met
- Listed with a dash (“-”) in the breakpoint tables

Expected susceptible phenotype → *Streptococcus pyogenes* and benzylpenicillin

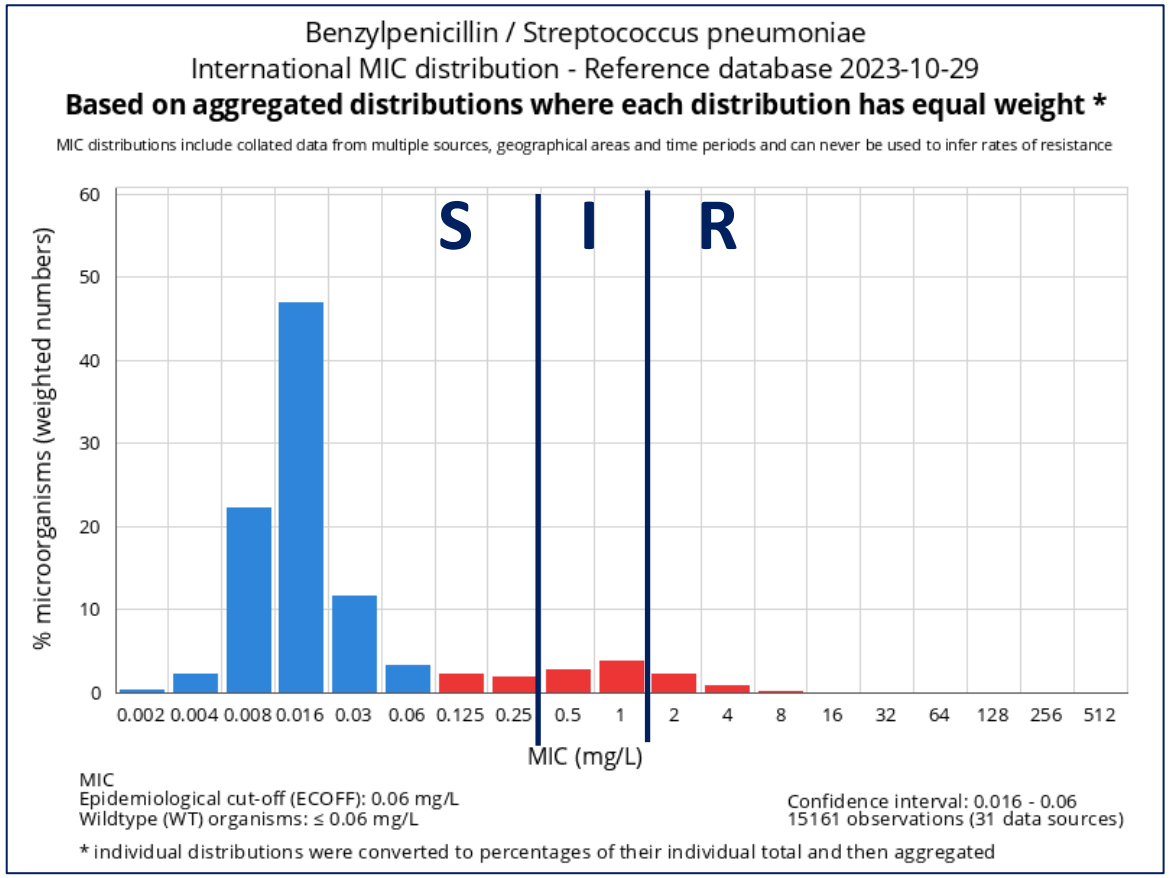
- ≥ 99% of population show MIC ≤ PK/PD susceptible (S) breakpoint

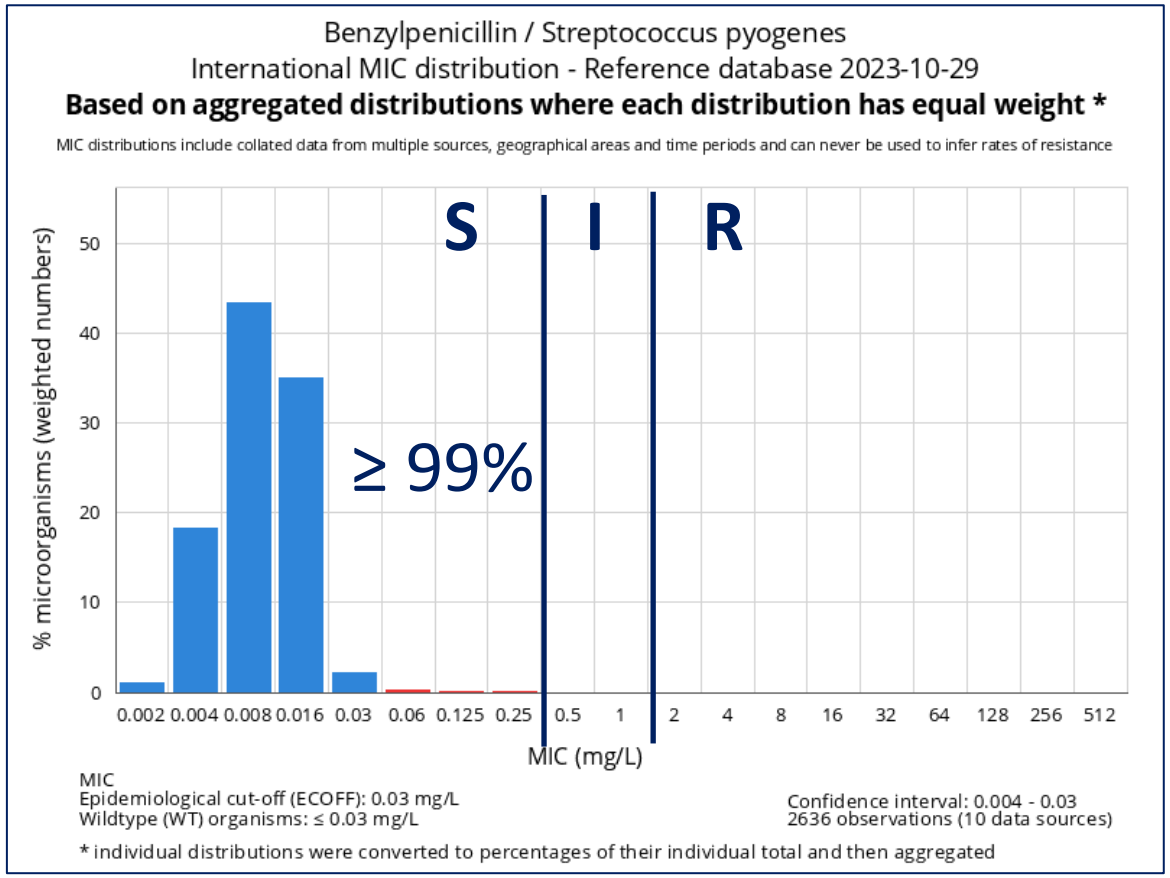
Expected Phenotypes validate identification

Gatermann S, Das S, Dubreuil L, Giske CG, Kahlmeter G, Lina G, Lindemann C, MacGowan A, Meletiadis J, Rossolini GM, Turnidge J, Cantón R. Expected phenotypes and Expert Rules are Important Complements to Antimicrobial Susceptibility Testing. Clin Microbiol Infect. 2022 Mar 16:S1198-743X(22)00146-X.

Expected phenotypes

Expected susceptible phenotype





Expected susceptible phenotype (resistance not expected) in gram-negative bacteria

Rule	Organisms	Unusual phenotypes
2.1	*Staphylococcus aureus*	Resistant to vancomycin, teicoplanin, telavancin, dalbavancin, oritavancin, daptomycin, linezolid, tedizolid, quinupristin-dalfopristin, tigecycline, eravacycline or omadacycline
2.2	Coagulase-negative staphylococci	Resistant to vancomycin, telavancin, dalbavancin, oritavancin, daptomycin, linezolid[1], tedizolid[1], quinupristin-dalfopristin[1], tigecycline, eravacycline or omadacycline
2.3	*Corynebacterium* spp.	Resistant to vancomycin, teicoplanin, telavancin, dalbavancin, oritavancin, daptomycin, linezolid, tedizolid, quinupristin-dalfopristin or tigecycline
2.4	*Streptococcus pneumoniae*	Resistant to carbapenems, vancomycin, teicoplanin, telavancin, dalbavancin, oritavancin, daptomycin, linezolid, tedizolid, quinupristin-dalfopristin, tigecycline, eravacycline, omadacycline or rifampicin.
2.5	Group A, B, C and G β-haemolytic streptococci	Resistant to penicillin, cephalosporins, vancomycin, teicoplanin, telavancin, dalbavancin, oritavancin, daptomycin, linezolid, tedizolid, quinupristin-dalfopristin, tigecycline, eravacycline or omadacycline
2.6	*Enterococcus* spp.	Resistant to daptomycin, linezolid, tigecycline, eravacycline or omadacycline Resistant to teicoplanin but not vancomycin
2.7	*Enterococcus faecalis*	Resistant to ampicillin
2.8	*Enterococcus faecalis, Enterococcus gallinarum, Enterococcus casseliflavus, Enterococcus avium*	Susceptible to quinupristin-dalfopristin, consider misidentification. If also resistant to ampicillin it is almost certainly *E. faecium*

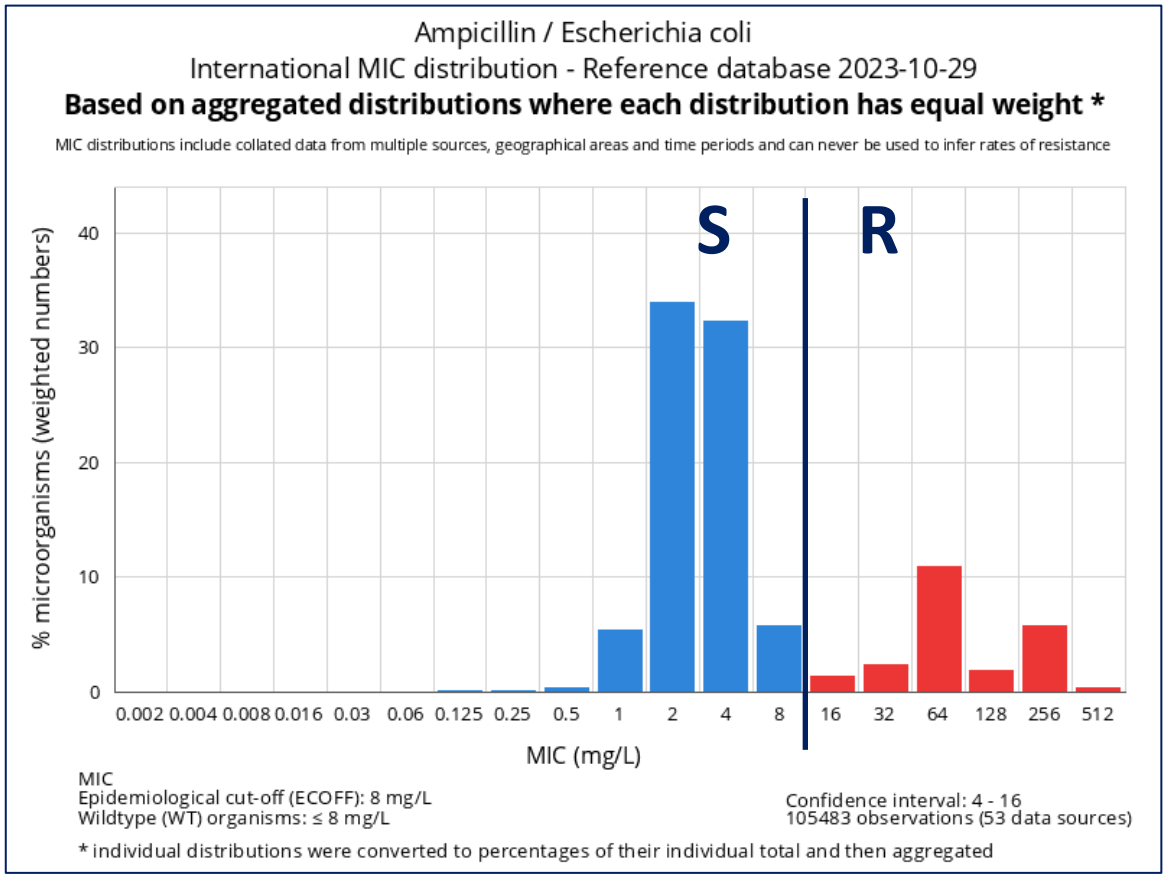
1 Except in countries where linezolid, tedizolid or quinupristin-dalfopristin resistant coagulase-negative staphylococci are not rare

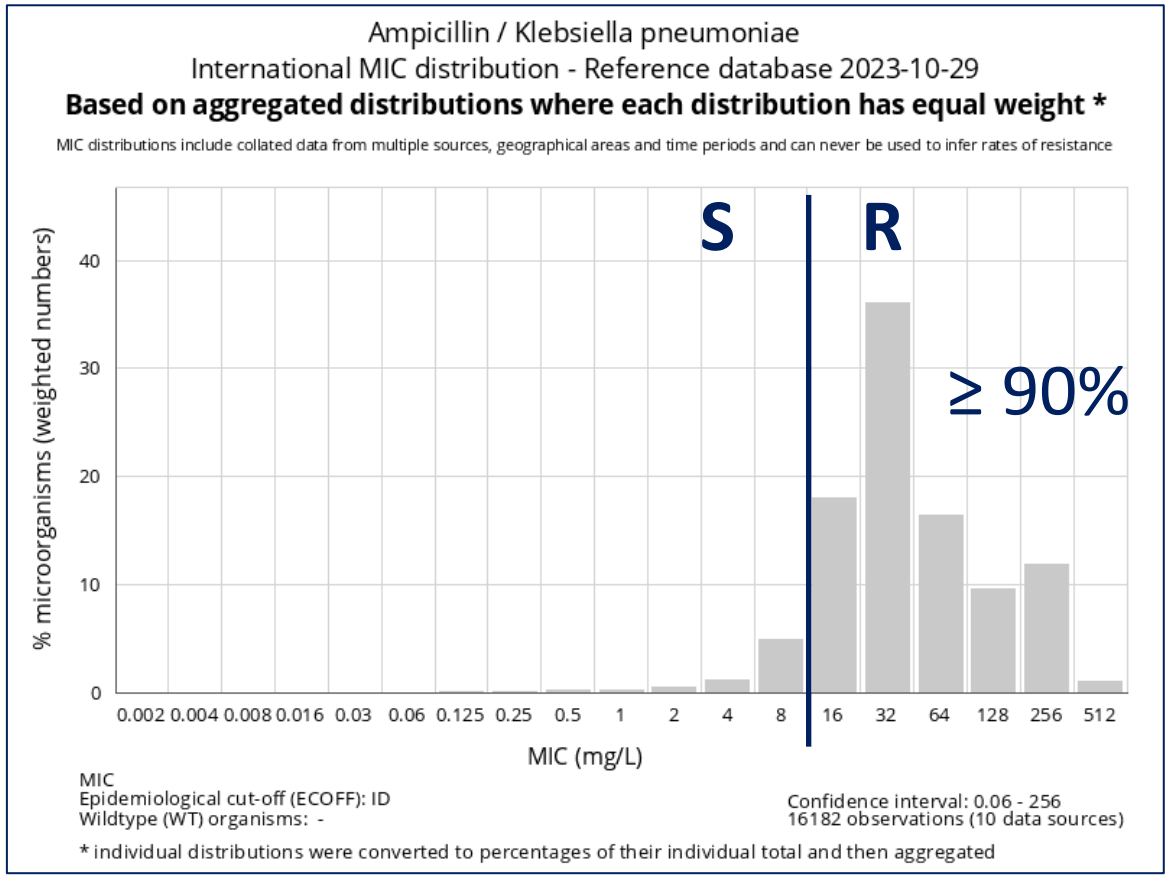
EUCAST Expected Susceptible Phenotypes v 1.1 March 2022

Only includes frequently isolated bacteria in clinical samples!

Expected phenotypes

Expected resistant phenotype





Expected resistant (susceptibility not expected) phenotypes in Enterobacterales/*Aeromonas* spp.

(also expected to be resistant to benzylpenicillin, glycopeptides,lipoglycopeptides, fusidic acid, macrolides, lincosamides, streptogramins, rifampicin, and oxazolidinones)

Rule	Organisms	Ampicillin/Amoxicillin	Amoxicilin-clavulanic acid	Ampicillin-sulbactam	Ticarcillin	Cefazolin, Cephalothin Cefalexin, Cefadroxil	Cefoxitin[2]	Cefuroxime	Tetracyclines	Tigecycline	Polymyxin B, Colistin	Fosfomycin	Nitrofurantoin
1.1	*Citrobacter koseri, Citrobacter amalonaticus*[3]	R			R								
1.2	*Citrobacter freundii*[4]	R	R	R		R	R						
1.3	*Enterobacter cloacae* complex	R	R	R		R	R						
1.4	*Escherichia hermannii*	R			R								
1.5	*Hafnia alvei*	R	R								R		
1.6	*Klebsiella aerogenes*	R	R	R		R	R						
1.7	*Klebsiella pneumoniae* complex	R			R								
1.8	*Klebsiella oxytoca*	R			R								
1.9	*Leclercia adecarboxylata*											R	
1.10	*Morganella morganii*	R	R	R		R			R		R		R
1.11	*Plesiomonas shigelloides*	R	R	R									
1.12	*Proteus mirabilis*								R		R		R
1.13	*Proteus penneri*	R				R		R	R		R		R
1.14	*Proteus vulgaris*	R				R		R	R		R		R
1.15	*Providencia rettgeri*	R	R	R		R			R		R		R

Only includes frequently isolated bacteria in clinical samples!

European Committee on Antimicrobial Susceptibility Testing

Breakpoint tables for interpretation of MICs and zone diameters

Version 14.0, valid from 2024-01-01

7. Dash in breakpoint tables indicates that the agent is unsuitable for treatment of infections caused by the organism or group of organisms and that testing and clinical use should be avoided. If included, report resistant without prior testing.

- When the expected phenotype of the organisms is resistant (*always listed with a dash*)
- It can denote an "implicit expert rule" that discourages use of the antimicrobial (e.g. moxifloxacin and *P. aeruginosa*)

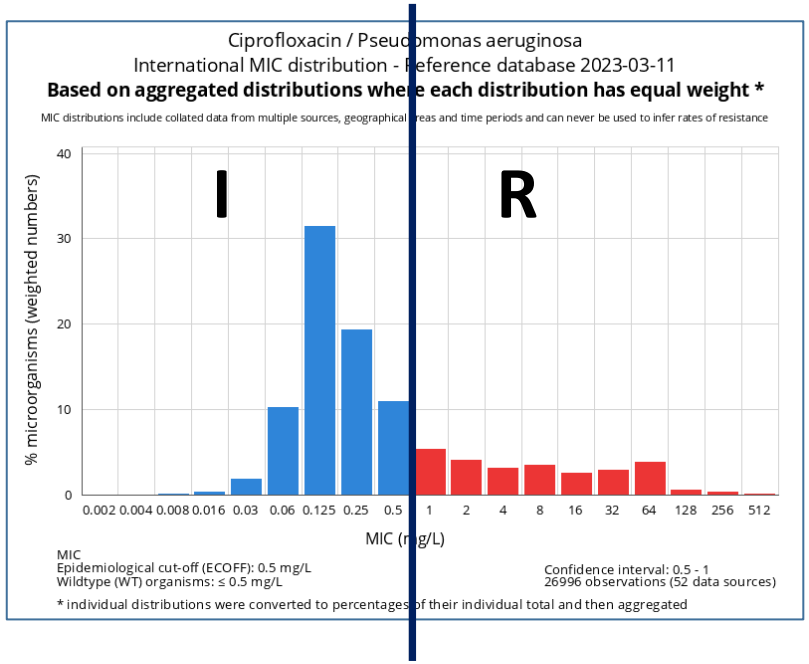
Pseudomonas aeruginosa and fluoroquinolones

Fluoroquinolones	MIC breakpoints (mg/L)			Disk content (µg)	Zone diameter breakpoints (mm)		
	S ≤	R >	ATU		S ≥	R <	ATU
Ciprofloxacin	0.001	0.5		5	50	26	
Delafloxacin	IE	IE			IE	IE	
Levofloxacin	0.001	2		5	50	18	
Moxifloxacin	-	-			-	-	
Nalidixic acid (screen only)	NA	NA			NA	NA	
Norfloxacin (uncomplicated UTI only)	-	-			-	-	
Ofloxacin	-	-			-	-	

https://www.eucast.org/clinical_breakpoints

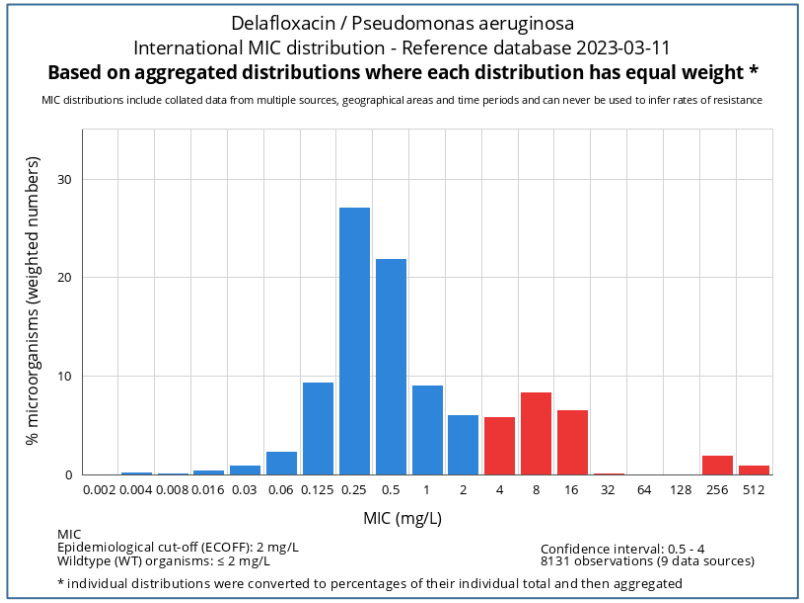
Pseudomonas aeruginosa

Ciprofloxacin



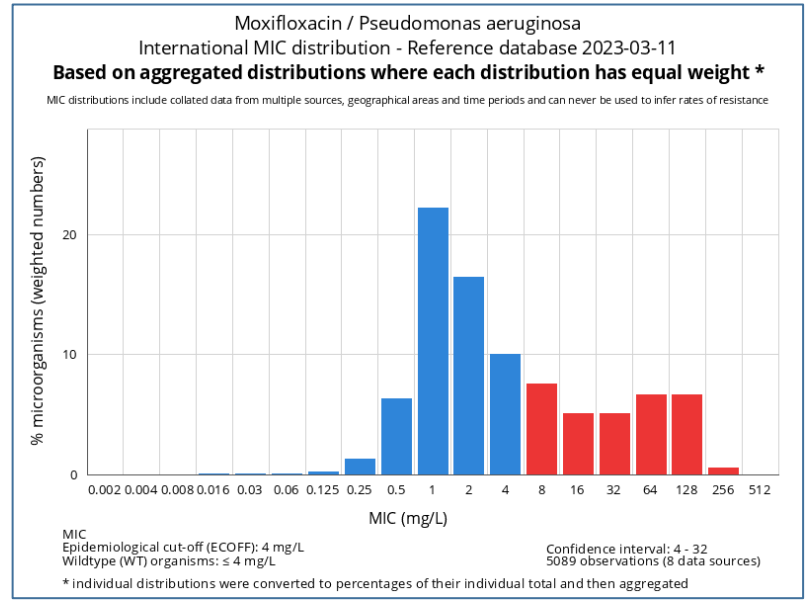
I ≤ 0.001 mg/L **R** >0.5 mg/L

Delafloxacin



IE (insufficient evidence)

Moxifloxacin



" – "

https://mic.eucast.org/

Expert rules

- Use identification and susceptibility testing results to deduce **recommendations for therapy**
- Represent **advice for antimicrobial therapy**, most often indicating when to avoid the use of antimicrobials that are likely to result in treatment failure
- Give **recommendations** how to handle situations that are currently **controversial or unresolved**
- Depend on **clinical breakpoints** and not on ECOFFs (if they differ)
- They can be **based on phenotypic screening tests** (e.g. nalidixic ac./*H. influenzae*) or detection of the expression of resistance (e.g., b-lactamase/*H. influenzae*)
- **Not based on molecular tests** [gene detection does not imply its expression (e.g. *ampC/E. coli*)]
- Expert rules **might change over time** when new evidence are available
- Organized in a similar way than breakpoint tables
- **Grade in clinical and microbiological evidences**

Gatermann S, Das S, Dubreuil L, Giske CG, Kahlmeter G, Lina G, Lindemann C, MacGowan A, Meletiadis J, Rossolini GM, Turnidge J, Cantón R.
Expected phenotypes and expert rules are Important complements to antimicrobial ausceptibility testing. Clin Microbiol Infect. 2022 Mar 16:S1198-743X(22)00146-X.

Expert rules

- All expert rules have **similar structure**

Rule No.	Organisms	Indicator Agent*	Agents affected*	Rule	Remarks	Grade	References
Beta-Lactams							
1	*E. coli, P. mirabilis*	ampicillin	piperacillin	IF resistant to ampicillin, THEN report resistant to piperacillin regardless of test result IF susceptible to ampicillin, THEN report as susceptible to piperacillin		A	Drusano, Schimpff, & Hewitt, 1984

Agents tested

Agents affected

IF … THEN …

Exceptions, scientific rational and comments

Evidence

References

Expert rules: Grade of the evidence

Evidence	2008	2022 - . . .
A	There is clinical evidence that reporting the test result as susceptible leads to clinical failures	There is **good clinical evidence** for the rule, i.e. applying the rule likely improves patient care. Grade A required clinical studies supporting the rule
B	Evidence is weak and based only on a few case reports or on experimental models. It is presumed that reporting the test result as susceptible may lead to clinical failures	**Evidence is weak** or **based on only a few case reports or on experimental data.** Animal studies are accepted as experimental data.
C	There is no clinical evidence, but microbiological data suggest that clinical use of the agent should be discouraged.	There is **no clinical evidence**, but ***in vitro* microbiological data** suggest that the rule should be applied.

Expert rules

Rule No.	Organisms	Indicator Agent*	Agents affected*	Rule	Remarks	Grade	References
Beta-Lactams							
1	*E. coli, P. mirabilis*	ampicillin	piperacillin	IF resistant to ampicillin, THEN report resistant to piperacillin regardless of test result IF susceptible to ampicillin, THEN report as susceptible to piperacillin		A	Drusano, Schimpff, & Hewitt, 1984

IF … .. … … THEN

Evidence

There is **good clinical evidence** for the rule (i.e. applying the rule likely improves patient care). Requires clinical studies supporting the rule

Expert rules

Rule No.	Organism(s)	Indicator Agent	Agent(s) Affected*	Rule	Remarks	Grade	References
6	*Enterococcus faecalis* *Enterococcus faecium*	vancomycin teicoplanin	teicoplanin	IF vancomycin resistant AND teicoplanin susceptible THEN report with a warning of resistance development to teicoplanin during therapy; IF vancomycin susceptible but *van*A is detected by molecular methods THEN report resistant to vancomycin and teicoplanin;	Enterococci harbouring *vanB* may appear susceptible to teicoplanin, but resistance may develop during therapy; the same is true if in phenotypically susceptible isolates harbour *vanA* or *vanB*	B	Holmes et al., 2013; Thaker et al., 2015

Evidence is weak or based on **only a few case reports** or on **experimental data**. Animal studies are accepted as experimental data

Expert rules

Rule No.	Organisms	Indicator Agent*	Agents affected*	Rule	Remarks	Grade	References
5	*Enterobacter* spp., *K. aerogenes*, *Citrobacter freundii*[†], *Serratia* spp., *Morganella morganii*, *Hafnia alvei*, *Providencia* spp.	cefuroxime	cefuroxime other 2nd generation cephalosporins	IF susceptible to cefuroxime, THEN report cefuroxime and/or any other 2nd generation cephalosporin as resistant	Although the breakpoint table does not list cefuroxime breakpoints for species other than *E. coli*, *P. mirabilis*, *Klebsiella* spp. (except *K. aerogenes*) and *Raoultella* spp., isolates may appear susceptible in vitro but the MICs tend to be higher than with the mentioned species and therapy with cefuroxime is not recommended. In addition, de-repressed mutants may be selected as with a third-generation cephalosporin.	C	

There is **no clinical evidence**, but in **vitro microbiological data** suggest that the rule should be applied.

Expert rules

- Expert rules also based in screen tests

Rule No.	Organisms	Indicator Agent	Agents affected	Rule	Remarks	Grade	References
5	*Staphylococcus* spp.	norfloxacin screening test	all fluoroquinolones	IF susceptible in norfloxacin screening test, THEN report as susceptible to ciprofloxacin, levofloxacin, moxifloxacin and ofloxacin IF resistant in norfloxacin screening test, THEN report individual agents as tested, and IF susceptible to either of ciprofloxacin, levofloxacin or moxifloxacin, THEN report agent as tested with a warning of risk for development of resistance during therapy with quinolones.	The screening test detects first step mutants and other mechanisms (e.g. efflux) that cause reduced susceptibility. Since mutants with increased efflux may still be susceptible to other fluoroquinolones these must be tested	C	Kaatz & Seo, 1997; Sierra et al., 2005

There is **no clinical evidence**, but ***in vitro*** **microbiological data** suggest that the rule should be applied.

Expert rules

- Expert rules are **continuously updated** when new information is available or other documents are updated (…. *June 2024*)

EUCAST Expert Rules v 3.3 on Enterobacterales

Rule No.	Organisms	Indicator Agent*	Agents affected*	Rule	Remarks	Grade	References
Beta-Lactams							
3	*Enterobacter* spp., *K. aerogenes, Citrobacter freundii*†, *Hafnia alvei*	cefotaxime, ceftriaxone, ceftazidime	cefotaxime, ceftriaxone, ceftazidime, piperacillin±tazobactam	IF susceptible in vitro to cefotaxime, ceftriaxone, ceftazidime, or piperacillin±tazobactam THEN EITHER add a note that monotherapy with cefotaxime, ceftriaxone, ceftazidime or piperacillin±tazobactam as well as combination therapy of these agents with an aminoglycoside should be discouraged owing to risk of selecting resistance, OR suppress the susceptibility testing results for these agents	Selection of AmpC de-repressed cephalosporin-resistant mutants may occur during therapy. The risk is relatively high in *Enterobacter* spp, *K. aerogenes* and *C. freundii* and low in *M, morganni* and *S, macescens*. For *Hafnia alvei in-vitro* mutation rates are similar to *Enterobacter* spp. or *C. freundii*. The use of a 3rd generation cephalosporin in combination with an aminoglycoside may also lead to failure by selection of resistant mutants. The combination with a quinolone, however, has found to be protective, although the clinical utility of this combination is not known The selection risk is absent or much diminished for cefepime	A	Sanders & Sanders, 1988; Choi et al., 2008; Harris & Ferguson, 2012; Kohlmann, Bähr, & Gatermann, 2018 Maillard et al 2023

Expert rules

- Expert rules are **continuously updated** when new information is available or other documents are updated (…. *June 2024*)

EUCAST Expert Rules v 3.3 on *Enterococcus* spp. – update January 2023

Rule No.	Organism(s)	Indicator Agent	Agent(s) Affected*	Rule	Remarks	Grade	References
	Fluoroquinolones						
4	*Enterococcus* spp.	norfloxacin screening test	ciprofloxacin levofloxacin	IF susceptible in the norfloxacin screening test THEN report susceptible to ciprofloxacin and levofloxacin IF resistant in the norfloxacin screening test THEN report ciprofloxacin and levofloxacin resistant or test the desired agent individually NOTE: this rule applies to isolates from uncomplicated UTI only	As with other gram-positive organisms, first step mutants as well as overexpressed efflux pumps are detected with norfloxacin; therefore, norfloxacin-susceptible isolates can be reported as susceptible to the other fluoroquinolones. In most cases, a positive result in the screening test also indicates resistance to other fluoroquinolones.	C	Oyamada, Ito, Inoue, & Yamagishi, 2006

Link to expert rules and Expected Phenotypes in the breakpoint tables

Pseudomonas spp.

Expert Rules and Expected Phenotypes
For abbreviations and explanations of breakpoints, see the Notes sheet

EUCAST Clinical Breakpoint Tables v. 14.0, valid from 2024-01-01

MIC determination (broth microdilution according to ISO standard 20776-1 ~~except for fosfomycin where agar dilution is used~~)
Medium: Cation-adjusted Mueller-Hinton broth (for cefiderocol, see https://www.eucast.org/eucastguidancedocuments/
Inoculum: $5x10^5$ CFU/mL
Incubation: Sealed panels, air, 35±1ºC, 18±2h
Reading: Unless otherwise stated, read MICs at the lowest concentration of the agent that completely inhibits visible growth. See "EUCAST Reading Guide for broth microdilution" for further information.
Quality control: *Pseudomonas aeruginosa* ATCC 27853. For agents not covered by this strain and for control of the inhibitor component of beta-lactam inhibitor combinations, see EUCAST QC Tables.

Disk diffusion (EUCAST standardised disk diffusion method)
Medium: Mueller-Hinton agar
Inoculum: McFarland 0.5
Incubation: Air, 35±1ºC, 18±2h
Reading: Unless otherwise stated, read zone edges as the point showing no growth viewed from the back of the plate against a dark background illuminated with reflected light. See "EUCAST Reading Guide for disk diffusion" for further information.
Quality control: *Pseudomonas aeruginosa* ATCC 27853. For agents not covered by this strain and for control of the inhibitor component of beta-lactam inhibitor-combination disks, see EUCAST QC Tables.

***Pseudomonas aeruginosa* is the most frequent species of this genus. Other less frequent *Pseudomonas* species recovered in clinical samples are: *P. fluorescens* group, *P. putida* group and *P. stutzeri* group.**

Penicillins	MIC breakpoints (mg/L)			Disk content (µg)	Zone diameter breakpoints (mm)			Notes
	S ≤	R >	ATU		S ≥	R <	ATU	Numbered notes relate to general comments and/or MIC breakpoints. Lettered notes relate to the disk diffusion method.
Benzylpenicillin	-	-			-	-		1. For susceptibility testing purposes, the concentration of tazobactam is fixed at 4 mg/L.
Ampicillin	-	-			-	-		2. For susceptibility testing purposes, the concentration of clavulanic acid is fixed at 2 mg/L.
Ampicillin-sulbactam	-	-			-	-		
Amoxicillin	-	-			-	-		
Amoxicillin-clavulanic acid	-	-			-	-		
Piperacillin	0.001	16		30	50	18	18-19	
Piperacillin-tazobactam	0.001[1]	16[1]		30-6	50	18	18-19	
Ticarcillin-clavulanic acid	0.001[2]	16[2]		75-10	50	18		
Temocillin	-	-			-	-		
Phenoxymethylpenicillin	-	-			-	-		
Oxacillin	-	-			-	-		
Cloxacillin	-	-			-	-		
Dicloxacillin	-	-			-	-		
Flucloxacillin	-	-			-	-		
Mecillinam oral (pivmecillinam) (uncomplicated UTI only)	-	-			-	-		

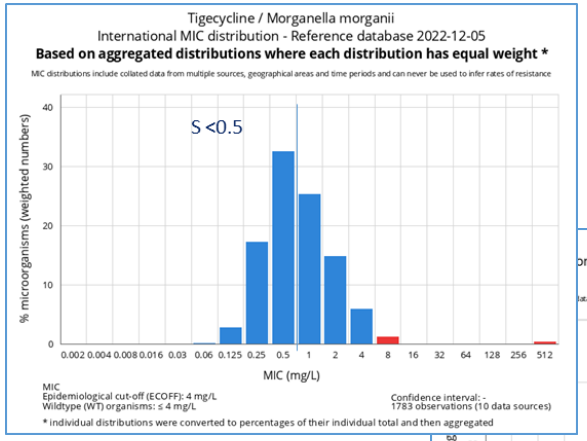
Summary: Expert rules and Expected Phenotypes

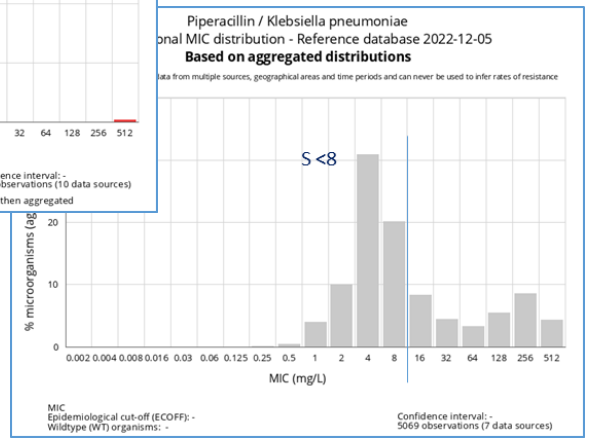
Expected Phenotypes → validate identification

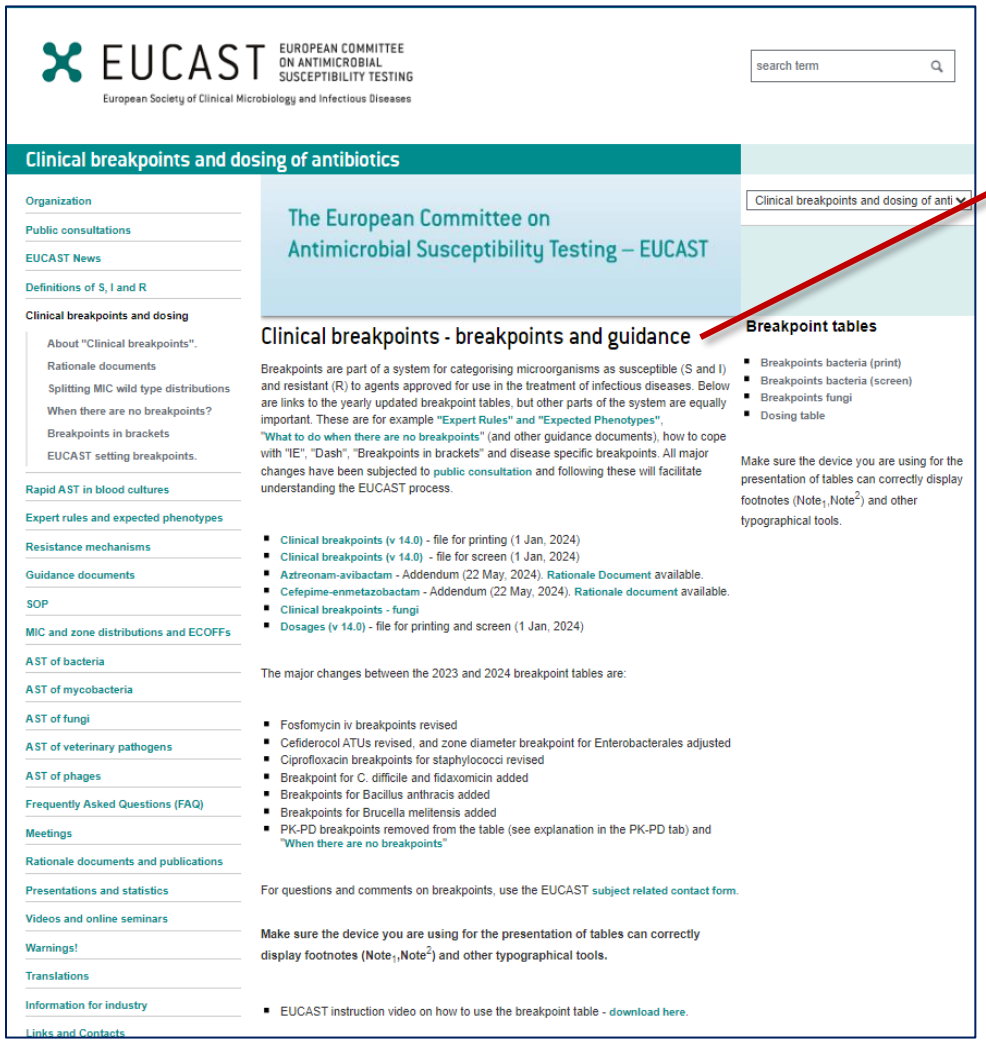
- Presumptive identification: *Klebsiella pneumoniae*
  - If test result: ampicillin S… likely misidentification
- Presumptive identification: *Streptococcus pyogenes*
  - If test result: penicillin R … likely misidentification

Expert Rules → improve therapy

- *Morganella morganii*
  - Tigecycline, not recommended although >10% test S
  - Expert rule: Report R regardless of test result
- *Klebsiella pneumoniae*
  - ~50% test S to piperacillin
  - Expert rule: Report R (several reports of clinical failure)







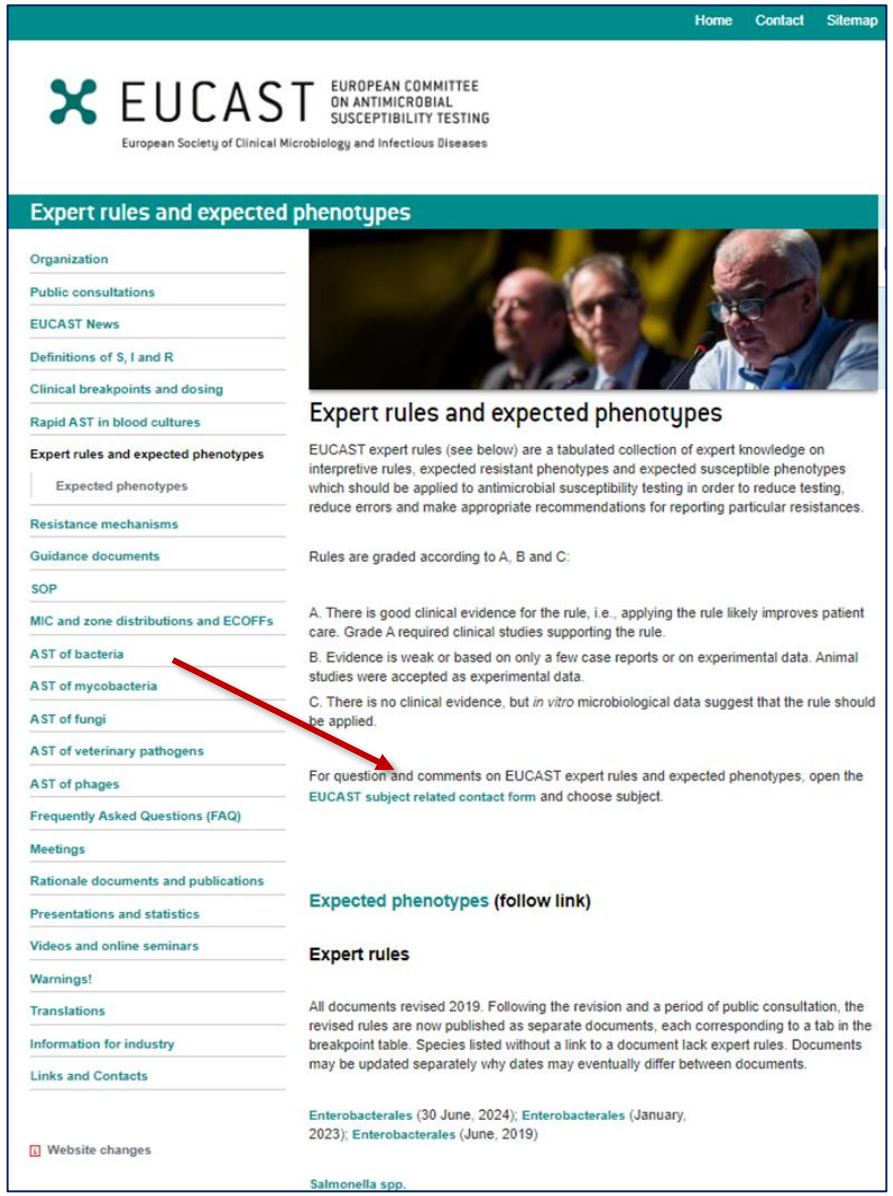
Clinical breakpoints - breakpoints and guidance

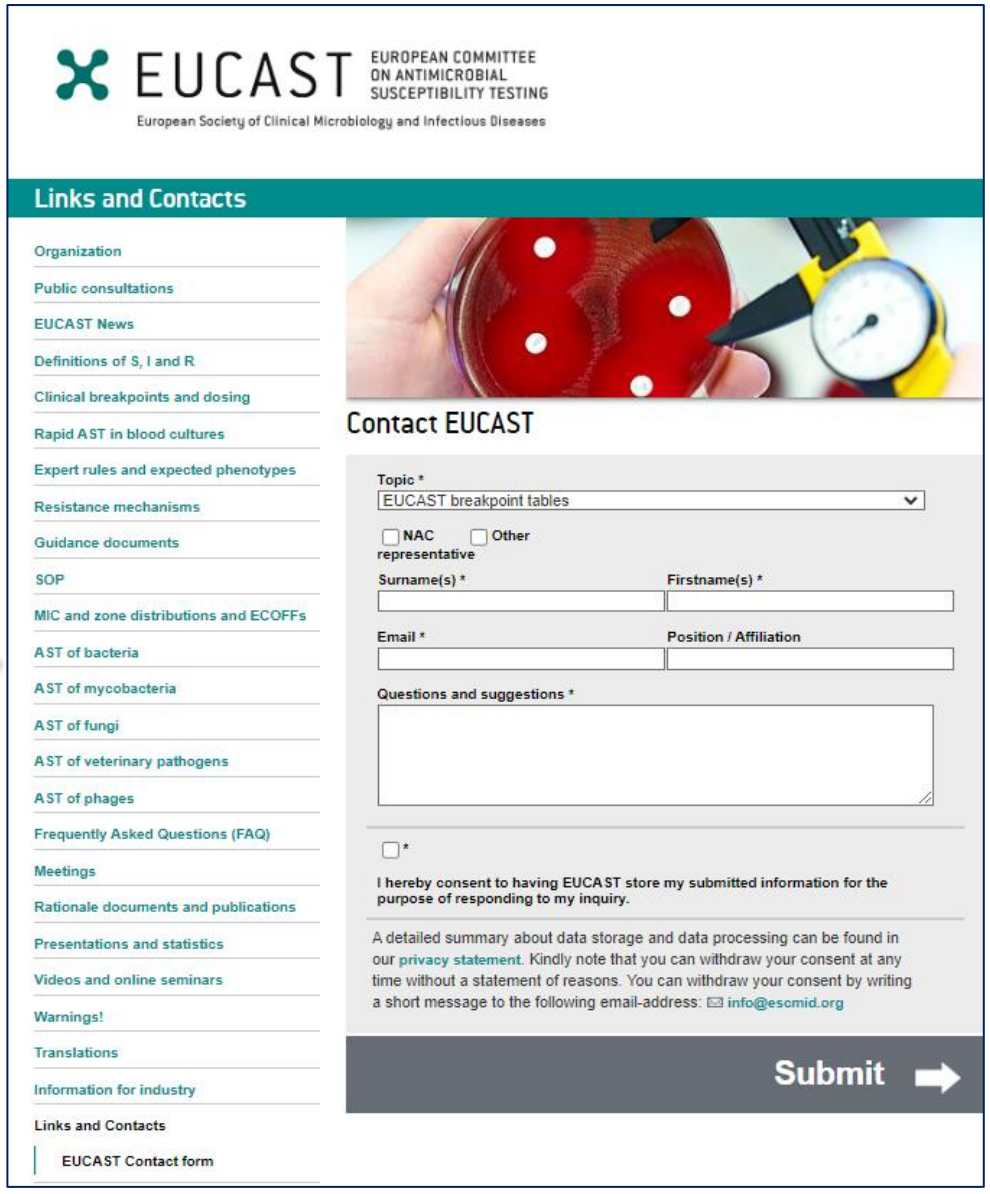
Breakpoints are part of a system for categorising microorganisms as susceptible (S and I) and resistant (R) to agents approved for use in the treatment of infectious diseases. Below are links to the yearly updated breakpoint tables, but other parts of the system are equally important. These are for example "Expert Rules" and "Expected Phenotypes", "What to do when there are no breakpoints" (and other guidance documents), how to cope with "IE", "Dash", "Breakpoints in brackets" and disease specific breakpoints. All major changes have been subjected to public consultation and following these will facilitate understanding the EUCAST process.

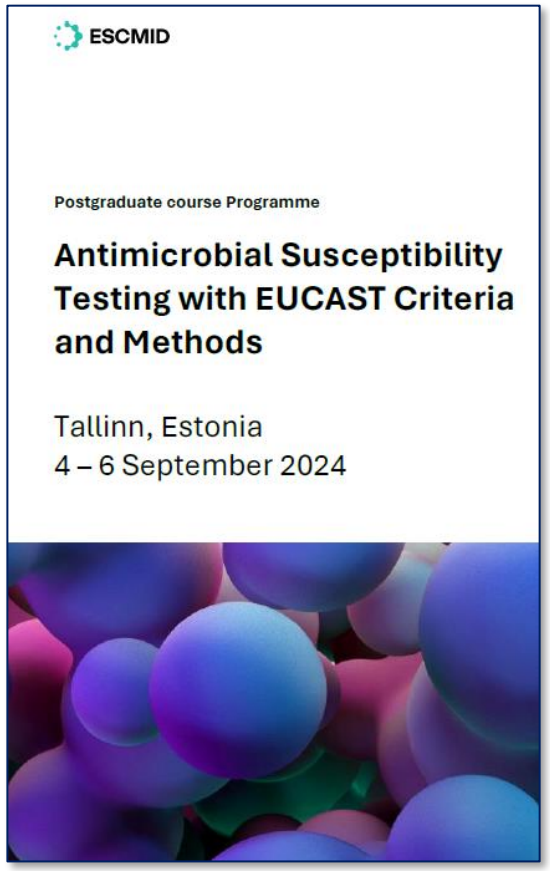
- Clinical breakpoints (v 14.0) - file for printing (1 Jan, 2024)
- Clinical breakpoints (v 14.0) - file for screen (1 Jan, 2024)
- Aztreonam-avibactam - Addendum (22 May, 2024). Rationale Document available.
- Cefepime-enmetazobactam - Addendum (22 May, 2024). Rationale document available.
- Clinical breakpoints - fungi
- Dosages (v 14.0) - file for printing and screen (1 Jan, 2024)

https://www.eucast.org/clinical_breakpoints

If you discover inconsistencies between expert rules/expected phenotypes, please, **alert us!**







EUCAST EUROPEAN COMMITTEE ON ANTIMICROBIAL SUSCEPTIBILITY TESTING
European Society of Clinical Microbiology and Infectious Diseases

Expected resistant phenotypes, expected susceptible phenotypes and expert rules

RYC MICROBIOLOGÍA HOSPITAL RAMÓN Y CAJAL

SaludMadrid
Dr. Rafael Cantón
Hospital Universitario Ramón y Cajal
SERVICIO DE MICROBIOLOGÍA Y PARASITOLOGÍA

instituto ramón y cajal de investigación sanitaria irycis

@RafaMCanton
@microRyC

ciberinfec isciii

Departamento de Microbiología y Parasitología Universidad Complutense. Madrid