

# **Carbapenemase-producing Enterobacterales (CPE): preparedness of Nordic laboratories**

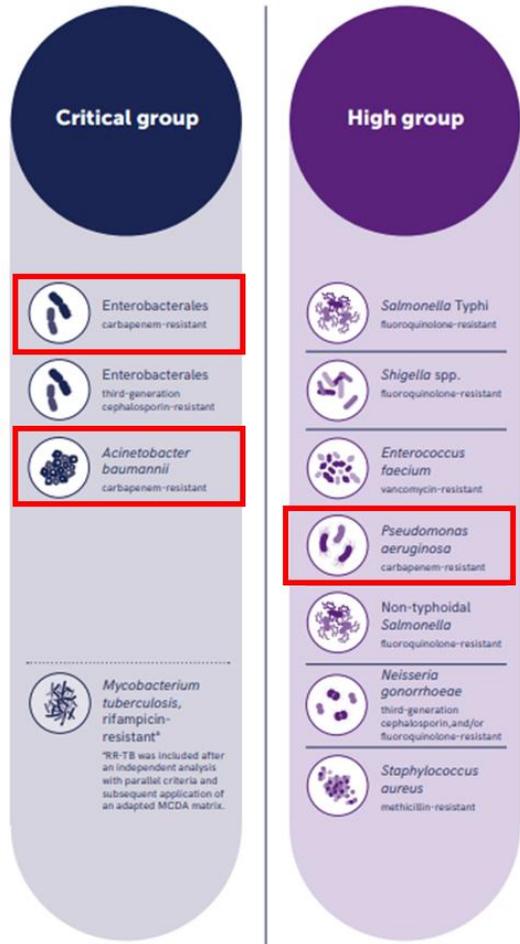
Meet-the-expert session  
NordicAST Workshop 2025

# **Part I: Carbapenemases – general overview and current epidemiology**

Ørjan Samuelsen, Norwegian Centre for Detection of AMR  
University Hospital of North Norway

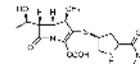
# Carbapenem resistance mechanisms - *Enterobacterales*

Fig 4. WHO Bacterial Priority Pathogens List, 2024



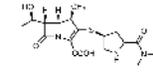
## ➤ Reduced permeability

- OmpK36 – *K. pneumoniae*
- OmpC/F – *E. coli*
- OmpC/F – *Enterobacter*



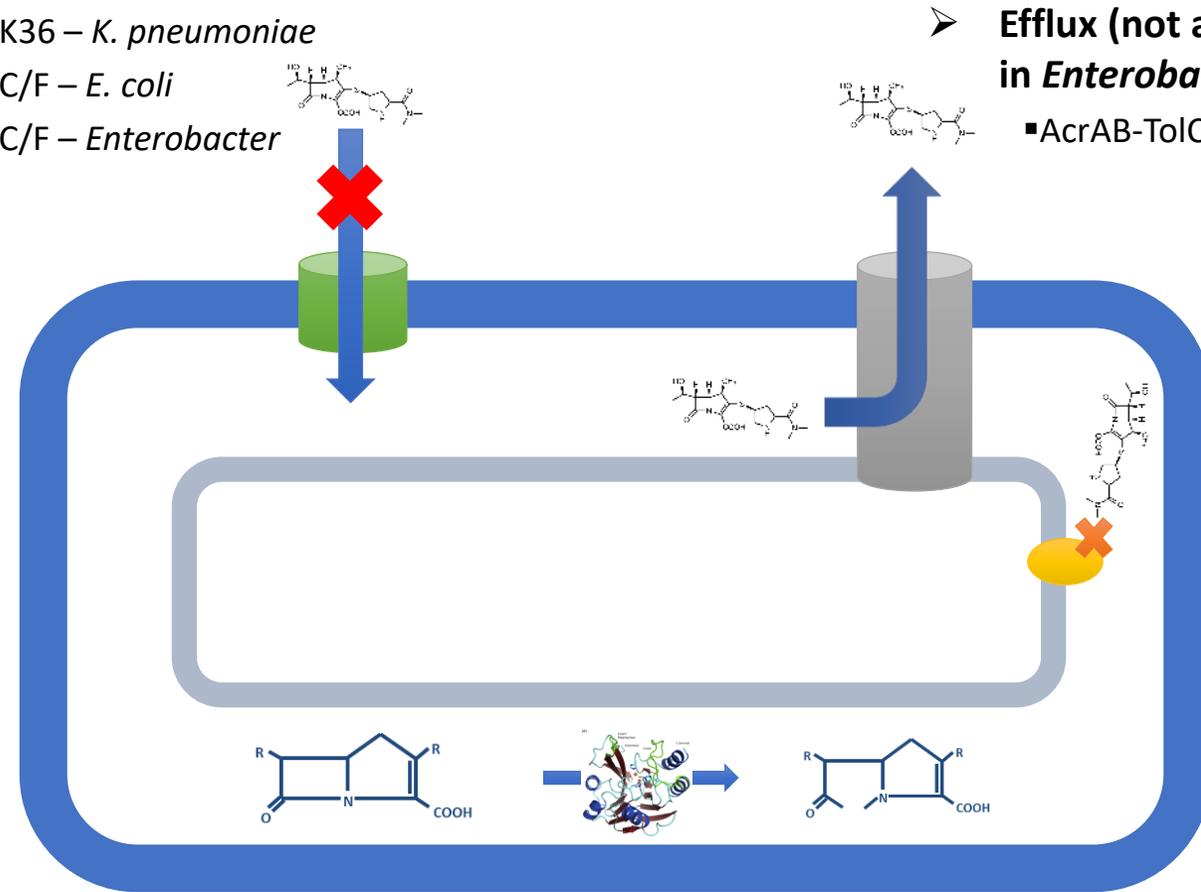
## ➤ Efflux (not a major contributor in *Enterobacterales*):

- AcrAB-TolC



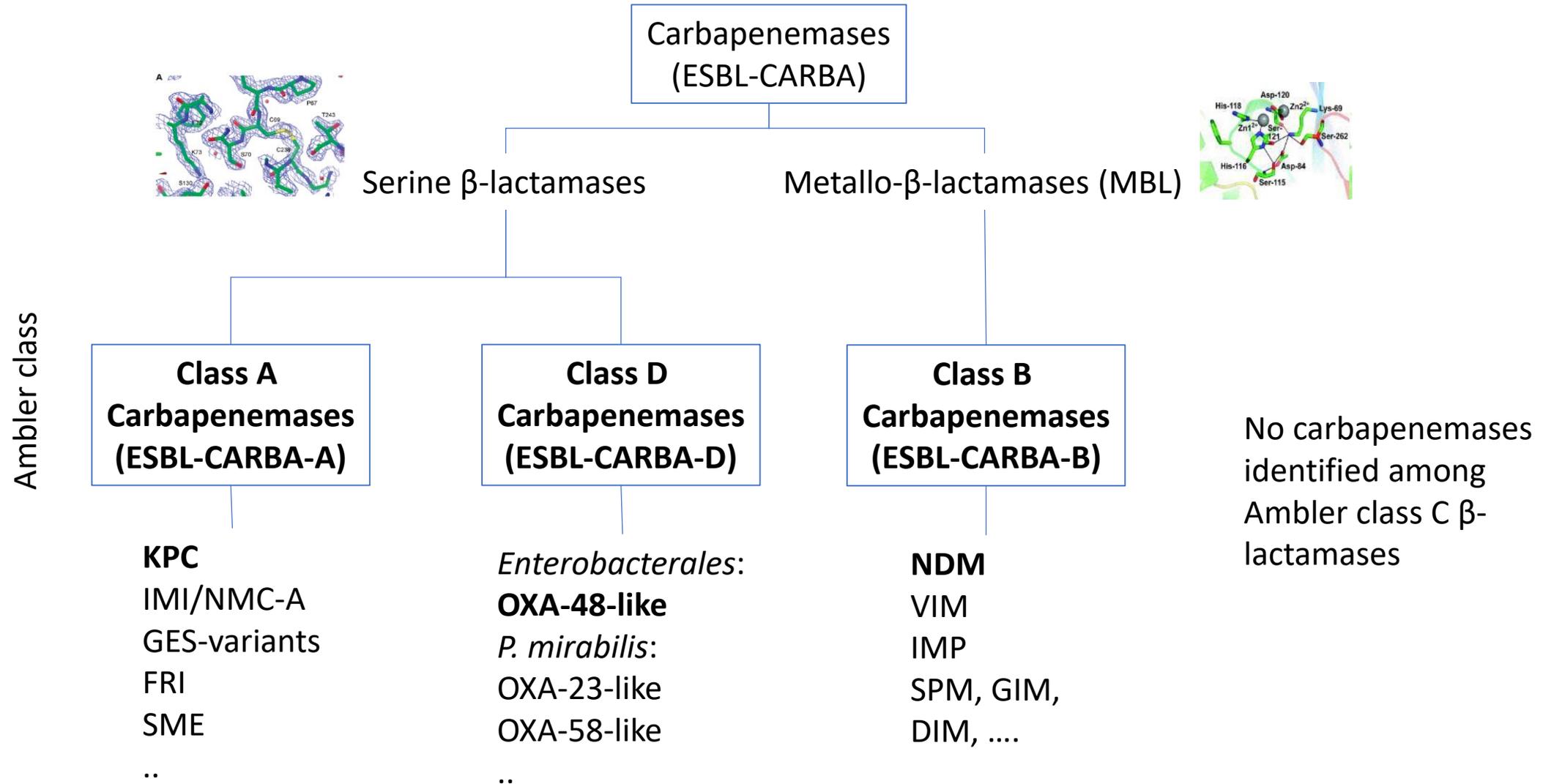
## ➤ PBP:

- Mutations -> ↓ affinity
- ↓ transcription



## ➤ β-lactamases – carbapenemases (ESBL-CARBA) – alternatively ESBL/AmpC + reduced permeability

# Carbapenemases - classification



# Carbapenemases – within-type variation

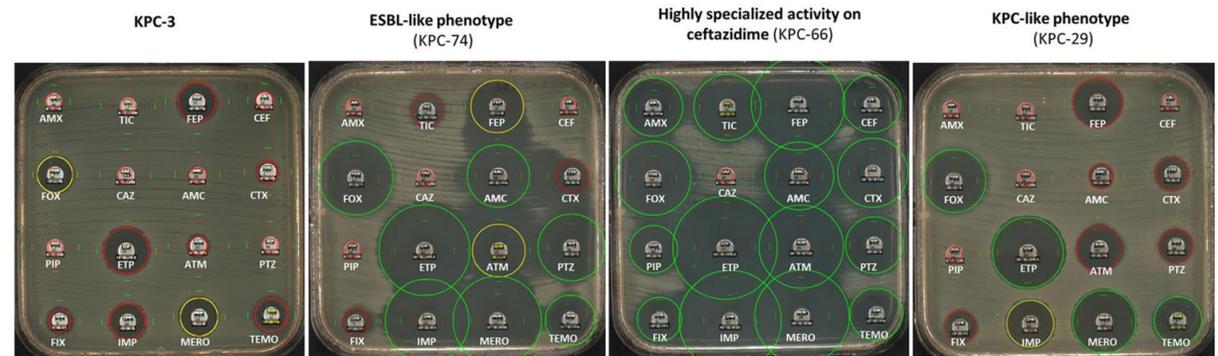
- KPC: ~249 variants
- NDM: ~81 variants
- OXA-48-like: ~20 variants

Note – not all variants are carbapenemases

Some ceftazidime-avibactam resistant variants of KPC have lost carbapenemase activity

<https://www.ncbi.nlm.nih.gov/pathogens/refgene/#>

Isolate	Starting KPC variant	Replicate no.	Meropenem MIC ( $\mu\text{g/ml}$ ) at <sup>a</sup> :						Ending KPC variant	ompK36 variant <sup>b</sup>	Ceftazidime-avibactam MIC ( $\mu\text{g/ml}$ ) at end of protocol (change from baseline) <sup>c</sup>	
			Baseline	Wk 1	Wk 2	Wk 3	Wk 4	Wk 5				Wk 6
3-A	KPC-3	1	32	>64	>64	—	—	—	KPC-3	ISS ins	4 (↔)	
		2	32	>64	>64	—	—	—	KPC-3	—	4 (↔)	
		3	32	>64	>64	—	—	—	KPC-3	—	4 (↔)	
4-A	KPC-3	1	16	>64	>64	—	—	—	KPC-3	ISS ins	4 (↔)	
		2	16	>64	>64	—	—	—	KPC-3	—	2 (↔)	
		3	16	>64	>64	—	—	—	KPC-3	—	4 (↔)	
2-B	V240G	1	8	>64	>64	—	—	—	V240G	WT	32 (↔)	
		2	8	>64	>64	—	—	—	V240G	—	16 (↔)	
		3	8	>64	>64	—	—	—	V240G	—	16 (↔)	
2-D	T243A	1	4	16	16	>64	>64	—	T243A	WT	8 (↔)	
		2	4	64	32	>64	>64	—	T243A	WT	16 (↔)	
		3	4	64	32	>64	>64	—	T243A	WT	8 (↔)	
3-B	D179Y	1	0.25	8	16	>64	>64	—	KPC-3	ISS ins	128 (↔)	
		2	0.25	4	16	>64	>64	—	T243A	F362D	16 (↓ 8-fold)	
		3	0.25	8	16	>64	>64	—	T243A	WT	128 (↔)	
4-C	A177E, D179Y	1	0.25	16	16	>64	>64	—	KPC-3	ISS ins	2 (↓ 64-fold)	
		2	0.25	0.25	16	>64	>64	—	T243A	WT	64 (↔)	
		3	0.25	0.5	8	32	>64	—	A177E	WT	2 (↓ 64-fold)	
1-B	D179Y, T243 M	1	0.5	8	8	16	16	32	64	D179Y, T243 M	Deletion	512 (↔)
		2	0.5	8	8	16	64	64	>64	T243A	ISS ins	128 (↔)
		3	0.5	8	16	16	32	32	32	D179Y, T243 M	ISS ins	512 (↔)



Hobson CA. et al. Antimicrob. Agents Chemother. 2022



Shields RK. et al. Antimicrob. Agents Chemother. 2017

# Activity spectrum

<b>β-lactam group</b>	<b>Class A</b>		<b>Class D</b>	<b>Class B</b>
	<b>KPC</b>	<b>IMI</b>	<b>OXA-48-gr., OXA-23-gr.</b>	<b>NDM, VIM, IMP</b>
Penicillins/1-2. gen cephalosporins	+	+	+	+
3-4. gen./extended spectrum cephalosporins	+	-	-	+
Monobactams	+	+	-	-
Carbapenems	+	+	+	+
<b>β-lactamase inhibitors</b>				
Clavulanic acid, tazobactam, enmetazobactam	+	+ / -	-	-
Avibactam	+	+	+	-
Vaborbactam, Relebactam	+	+	-	-
Boronic acid	+	+	-	-
Cloxacillin	-	-	-	-
Dipicolinic acid, EDTA	-	-	-	+

⊕ = hydrolytically active

⊖ = no/limited hydrolytic activity

⊕ = inhibitory activity

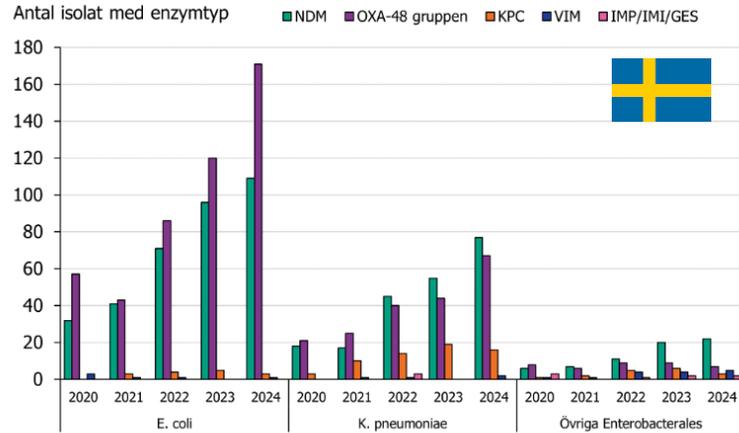
⊖ = no/limited inhibitory activity

} Used for diagnostic purposes

Note: variable activity within each class and variant

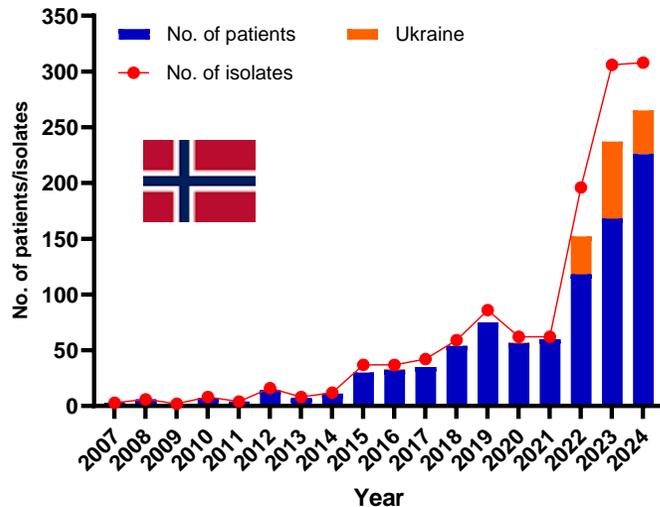
# CPE in the Nordic countries

Figur 4. Antalet isolat och enzymtyper av ESBL-CARBA-producerande *E. coli*, *K. pneumoniae* och övriga arter inom familjen Enterobacterales under åren 2019-2024.



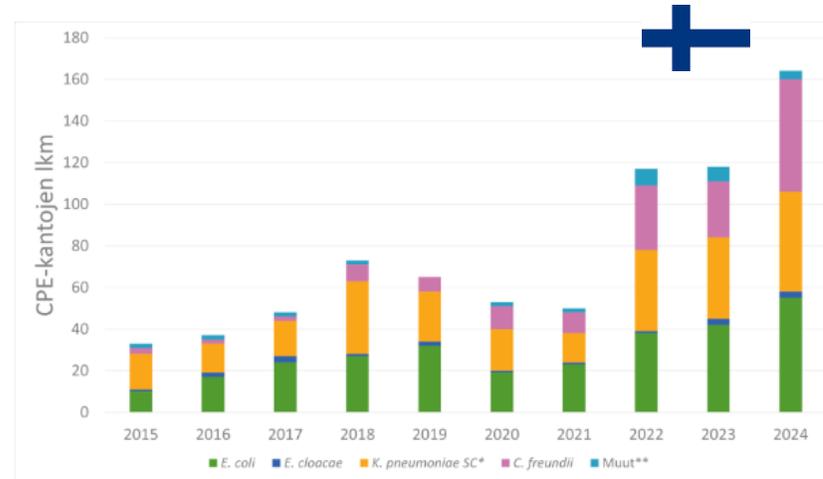
Källa: Folkhälsomyndigheten

<https://www.folkhalsomyndigheten.se/folkhalsorapportering-statistik/statistik-a-o/sjukdomsstatistik/esblcarba/?tab=tab-report>



<https://www.unn.no/fag-og-forskning/k-res/nyheter/rapport-forekomst-og-molekylargenetiske-analyser-av-vre-lre-og-cpo-i-norge-2024/>

## CPE-esiintyvys Suomessa

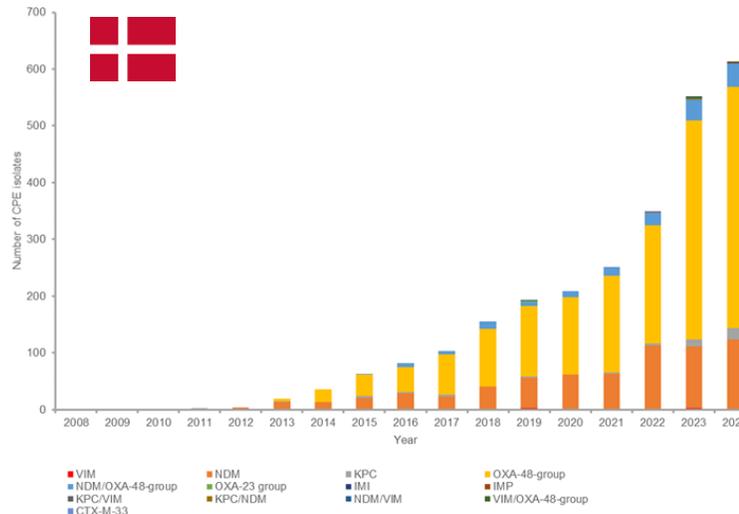


<https://thl.fi/aiheet/infektiotaudit-ja-rokotukset/taudit-ja-torjunta/taudit-ja-taudinaiheuttajat-a-o/cpe/cpe-esiintyvys-suomessa>



Mynd 54. Fjöldi einstaklinga sem greindist í fyrsta sinn með karbapenemasa myndandi *Enterobacterales* sýkla á Íslandi eftir ári og tegund rannsóknar árin 2014–2023. Upplýsingar frá sýkla- og veirufræðideild Landspítala.

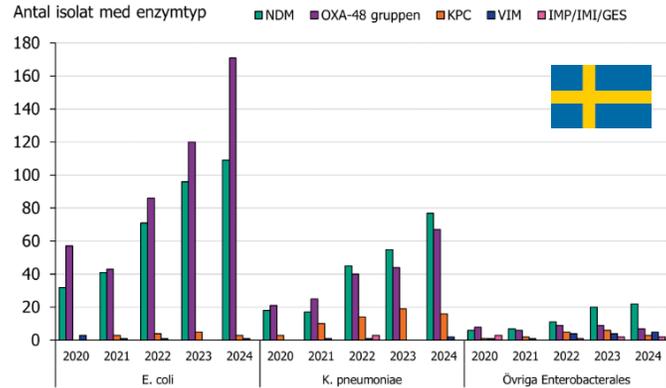
[https://assets.ctfassets.net/8k0h54kbe6bj/61U5AHm38hLnGanHpPhT5m/2762cc1e65ec78fa6ba9d85701b4f59f/S\\_klalyfjask\\_rsla\\_2023.pdf](https://assets.ctfassets.net/8k0h54kbe6bj/61U5AHm38hLnGanHpPhT5m/2762cc1e65ec78fa6ba9d85701b4f59f/S_klalyfjask_rsla_2023.pdf)



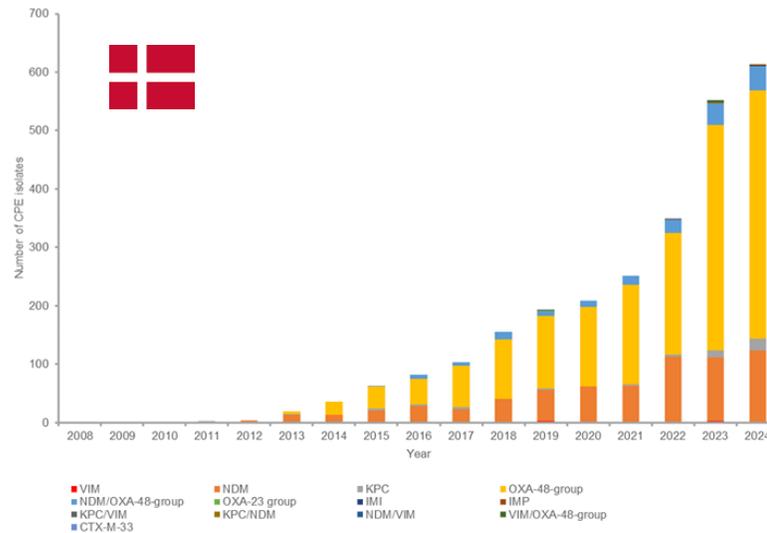
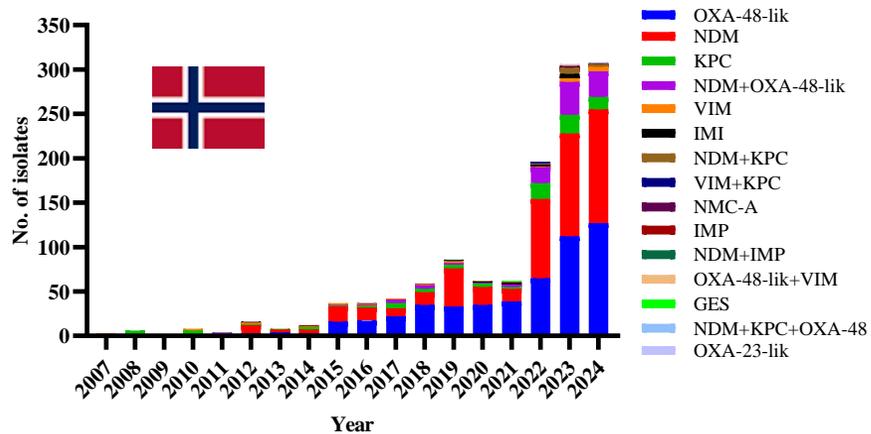
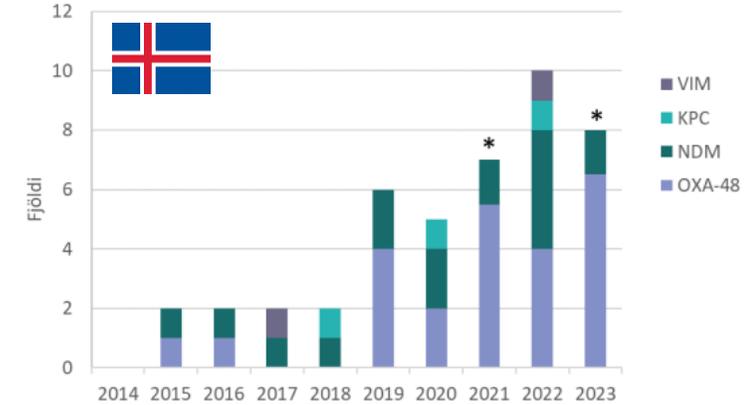
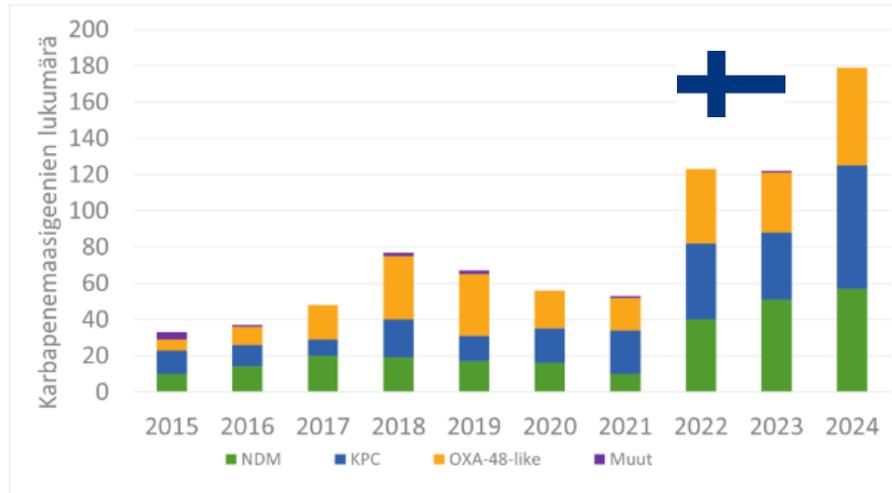
Anette Hammerum, SSI

# Carbapenemase variants

Figur 4. Antalet isolat och enzymtyper av ESBL-CARBA-producerande E. coli, K. pneumoniae och övriga arter inom familjen Enterobacterales under åren 2019-2024.



Källa: Folkhälsomyndigheten

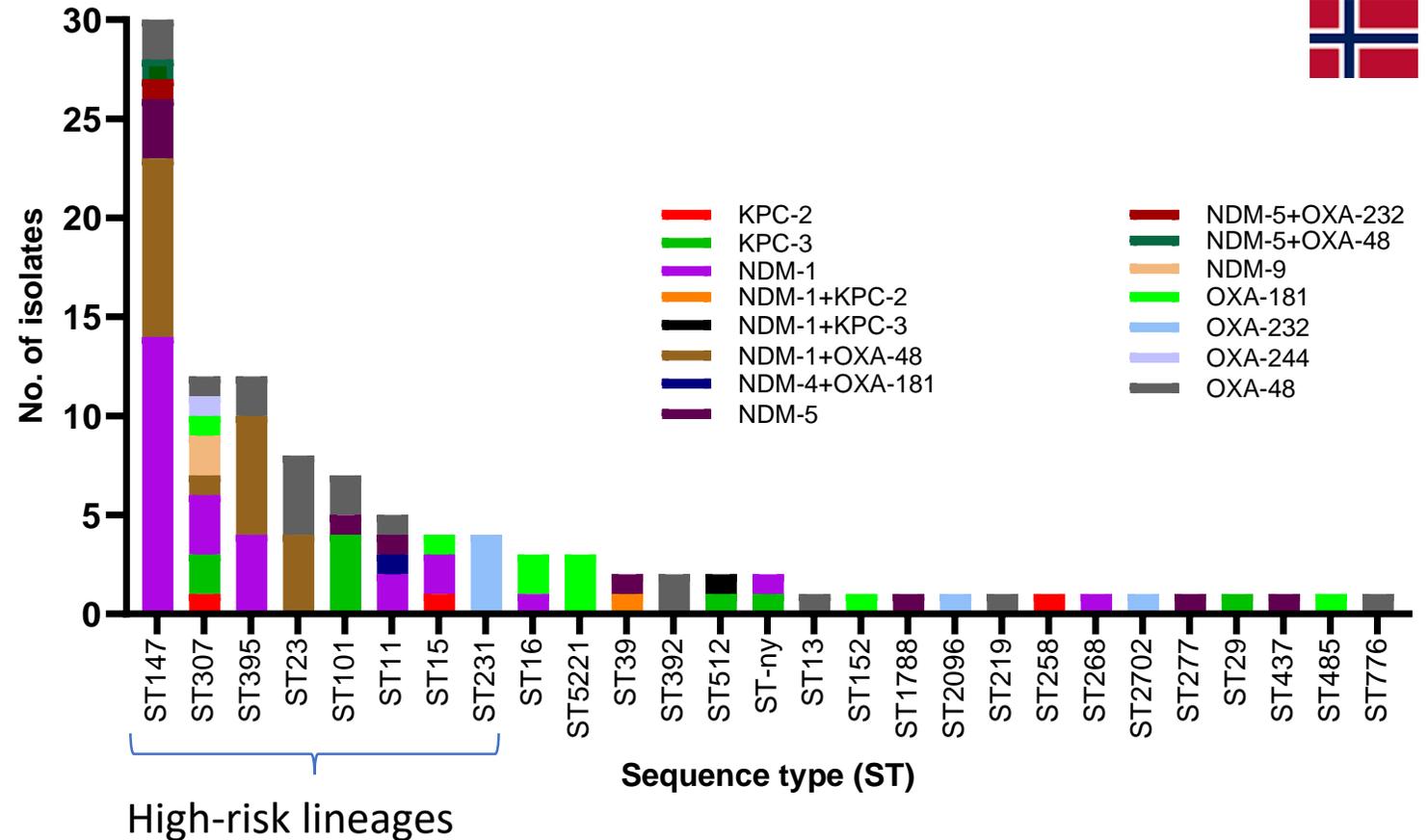


- NDM & OXA-48-like dominating
- Finland: more KPC
- Double carbapenemase producers
  - NDM+OXA-48-like

# High-risk lineages – *K. pneumoniae*



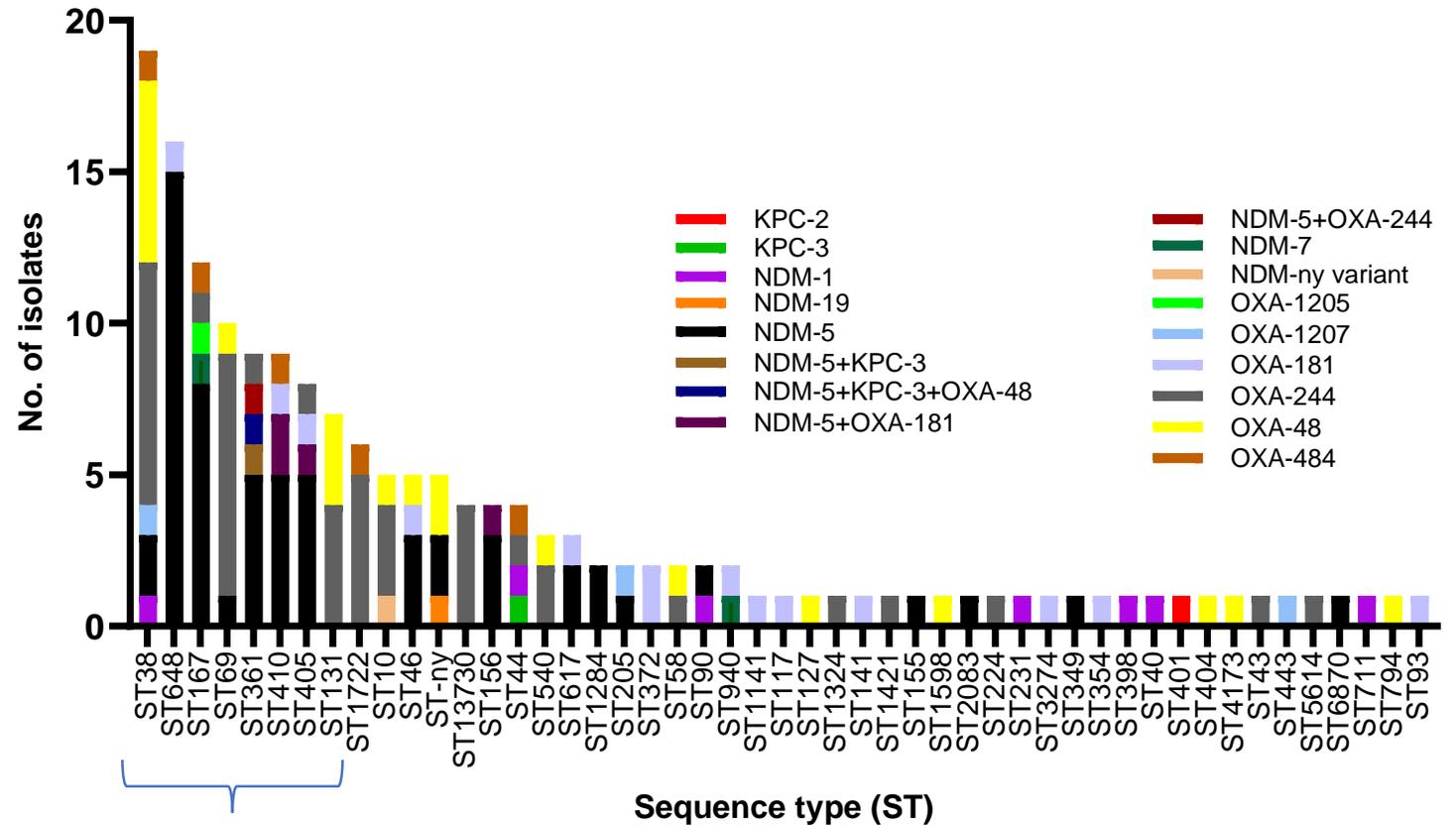
«..genomic evidence of continued transmission and outbreaks of **high-risk lineages** of carbapenemase-producing *K. pneumoniae* in hospitals in the EU/EEA»



# High-risk lineages - *E. coli*



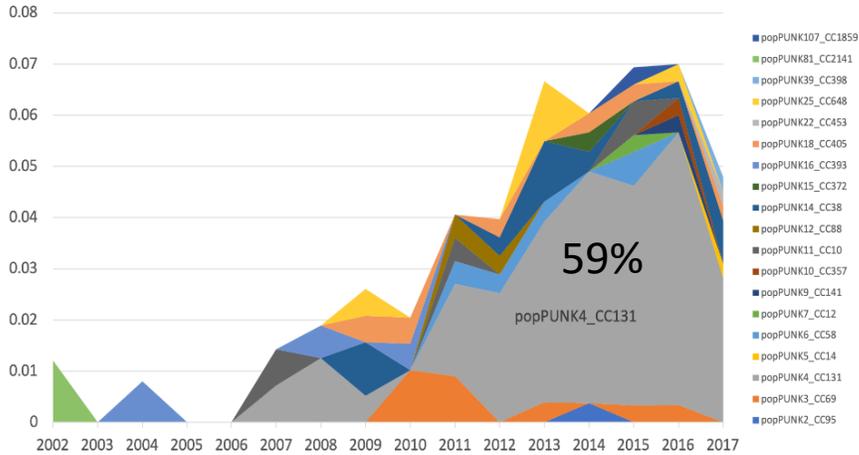
«..increasing detection of high-risk lineages of *E. coli* carrying carbapenemase genes in the EU/EEA with a risk for transmission in the community»



High-risk lineages

# Carbapenemase-producing *E. coli* ST131

- ST131 – dominant *E. coli* ESBL lineage globally



Gladstone RA. et al. Lancet Microbe 2021

## RAPID COMMUNICATION

Emergence of *Escherichia coli* ST131 carrying carbapenemase genes, European Union/European Economic Area, August 2012 to May 2024

Anke Kohlenberg<sup>1</sup>, Olov Svartström<sup>1</sup>, Petra Apfalter<sup>2</sup>, Rainer Hartl<sup>1</sup>, Pierre Bogaerts<sup>3</sup>, Te-Din Huang<sup>3</sup>, Katerina Chudejova<sup>4</sup>, Lucia Malisova<sup>4,5</sup>, Jessica Eisfeld<sup>6</sup>, Mirco Sandfort<sup>6</sup>, Anette M Hammerum<sup>6</sup>, Louise Roer<sup>6</sup>, Kati Räisänen<sup>6,7</sup>, Laurent Dortet<sup>6,8</sup>, Rémy A Bonnin<sup>6,9,10</sup>, Ákos Tóth<sup>11</sup>, Kinga Tóth<sup>11</sup>, Christina Clarke<sup>12</sup>, Martin Cormican<sup>13</sup>, Algirdas Griškevičius<sup>14</sup>, Kirstin Khomyongwa<sup>15</sup>, Marie Mee<sup>16</sup>, Baiba Niedre-Otomere<sup>17</sup>, Reins Vangravs<sup>18</sup>, Antoni PA Hendrickx<sup>19</sup>, Dan W Notermans<sup>20</sup>, Brjan Samuelssen<sup>21</sup>, Manuela Canica<sup>22</sup>, Vera Manageiro<sup>23</sup>, Vilhelm Müller<sup>24</sup>, Barbro Mäkitalo<sup>25</sup>, Urška Kramar<sup>26</sup>, Mateja Pirs<sup>26</sup>, Daniel Patm<sup>27</sup>, Dominique L Monnet<sup>28</sup>, Erik Alm<sup>29</sup>, Marius Linkevicius<sup>30</sup>

Kohlenberg A. et al. Euro Surveill. 2024

FIGURE 1

Number of *Escherichia coli* ST131 isolates, including its single locus variants<sup>a</sup>, carrying carbapenemase genes, by year, EU/EEA, 2012–2023<sup>b</sup> (n = 535)

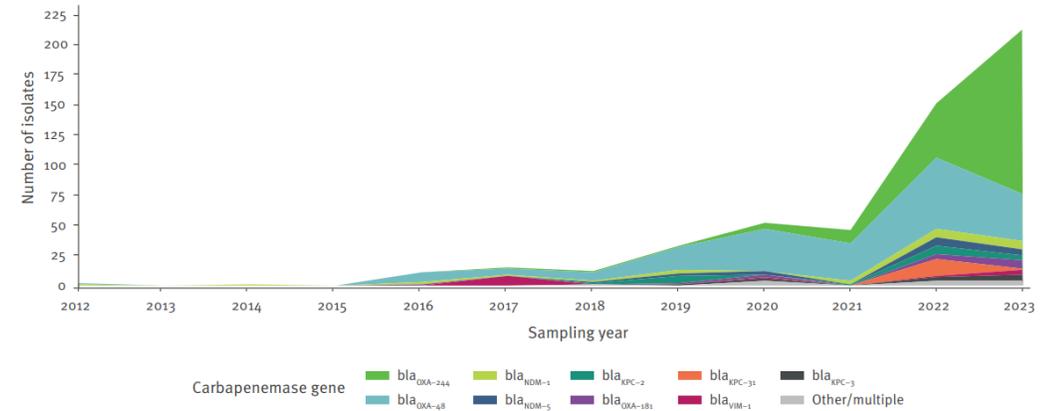


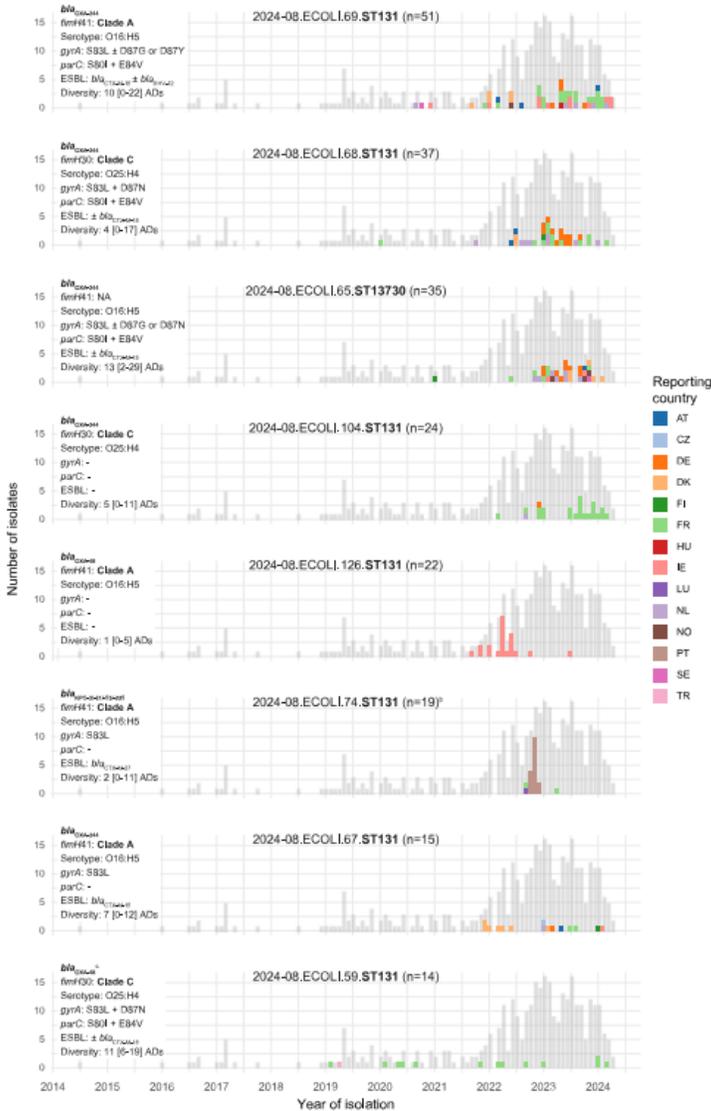
TABLE 1

Isolates of *Escherichia coli* ST131 and its single locus variants carrying carbapenemase genes, by country, EU/EEA, August 2012–May 2024 (n = 594)

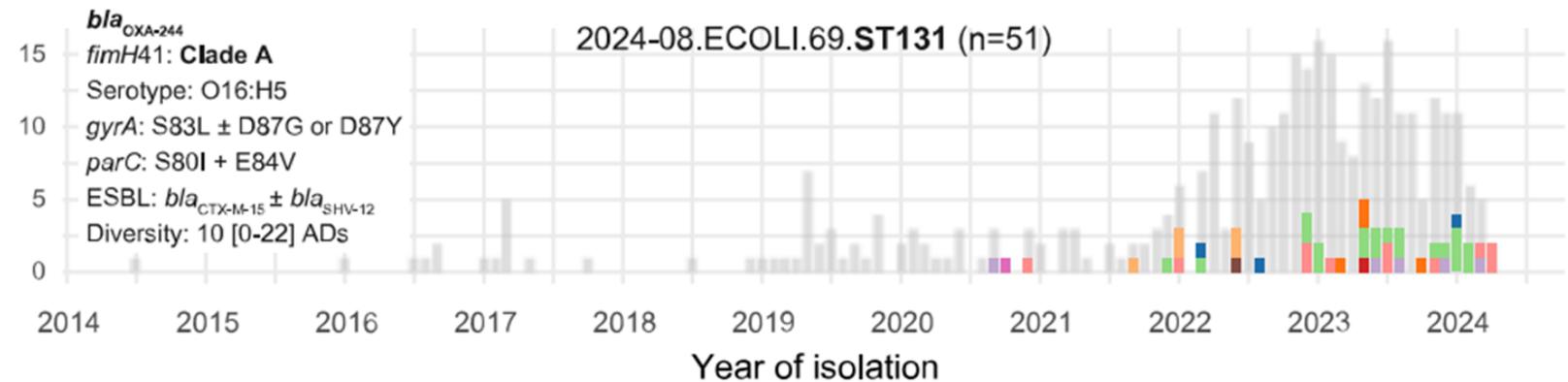
Carbapenemase gene	Number of isolates by country and period covered																	Total
	AT	BE	CZ	DE	DK	FI	FR	HU	IE	LT	LU	LV	NL	NO	PT	SE	SI	
	2022–2024	2023–2024	2021–2023	2022–2023	2014–2024	2021–2024	2019–2024	2023	2016–2024	2019–2023	2019–2024	2023	2012–2024	2012–2023	2016–2022	2017–2024	2022–2023	
bla <sub>OXA-244</sub>	8	3	1	32	25	3	85	1	14	0	2	0	28	6	0	21	1	230
bla <sub>OXA-48</sub>	0	2	0	1	9	0	82	0	101	1	1	1	13	3	0	9	1	224
bla <sub>NDM-1</sub>	0	2	2	1	3	1	7	1	4	0	0	0	4	2	3	0	1	31
bla <sub>NDM-5</sub>	0	0	0	1	3	0	8	0	2	0	1	0	3	0	0	2	0	20
bla <sub>KPC-2</sub>	0	0	0	1	0	0	1	0	3	11	0	0	1	0	0	2	0	19
bla <sub>OXA-181</sub>	0	0	0	1	3	0	8	0	3	0	0	0	0	1	0	0	0	16
bla <sub>KPC-31</sub>	0	0	0	0	0	0	1	0	0	0	0	0	0	0	14	0	0	15
bla <sub>VIM-1</sub>	0	1	0	1	0	0	3	0	0	0	0	0	9	0	0	0	0	14
bla <sub>KPC-3</sub>	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	12
bla <sub>NDM-2</sub>	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	2
bla <sub>OXA-48a</sub>	0	1	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	2
bla <sub>NDM-18</sub>	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	1
bla <sub>VIM-4</sub>	0	0	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	1
bla <sub>KPC-31</sub>	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	1
bla <sub>KPC-25</sub>	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	1
bla <sub>OXA-204</sub>	0	0	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	1
bla <sub>OXA-232</sub>	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0	1
bla <sub>OXA-244</sub> <sup>b</sup>	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0	1
bla <sub>NDM-1</sub> /bla <sub>OXA-232</sub>	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0	1
bla <sub>NDM-1</sub> /bla <sub>OXA-244</sub>	0	0	0	0	0	0	0	0	1	0	0	0	0	0	0	0	0	1
Total	8	10	3	40	44	6	202	2	129	12	5	2	60	13	20	35	3	594

Nordic countries 16% of all isolates

# Carbapenemase-producing *E. coli* ST131



- Isolates from Nordic countries intermingled within European clusters

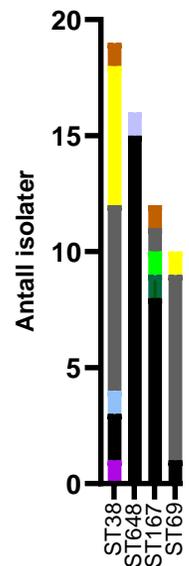


- Denmark, Sweden & Norway

# Spread in the community?

«..increasing detection of high-risk lineages of *E. coli* carrying carbapenemase genes in the EU/EEA with a risk for transmission in the community»

- High proportion in urine
- Women > men
- Geographical spread of cases without epidemiological links

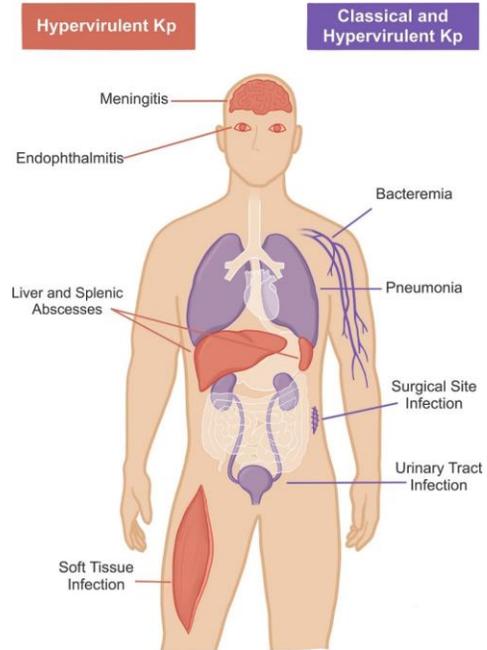


2015-2024:

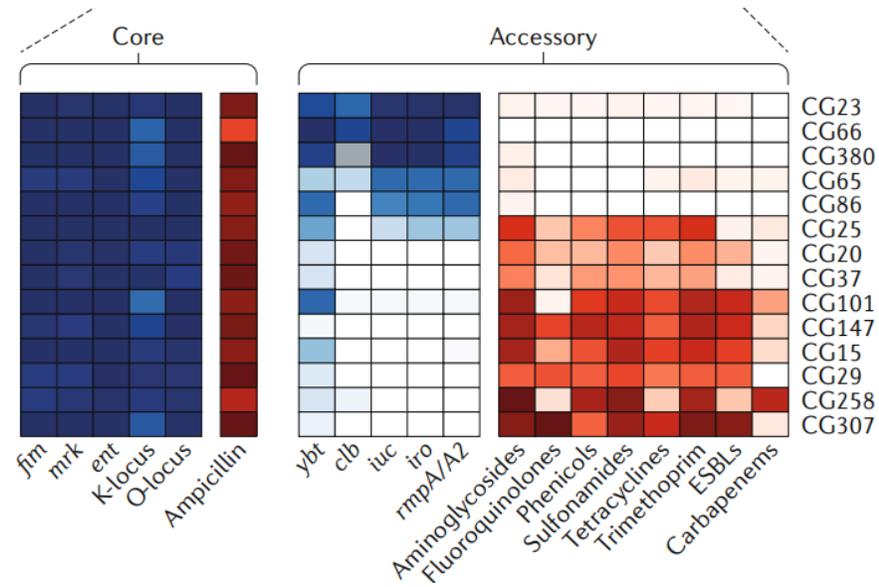
	ST38* (n=88)	ST69 (n=34)	ST167 (n=60)	ST648 (n=39)
<b>Import</b>	<b>35 %</b>	<b>32 %</b>	<b>77 %</b>	<b>74 %</b>
No import	36 %	24 %	3 %	10 %
Import unknown	28 %	44 %	20 %	16 %
<b>Screening sample</b>	<b>52 %</b>	<b>35 %</b>	<b>73 %</b>	<b>92 %</b>

\* outbreak 2020: n=12

# Carbapenemase-producing hypervirulent *K. pneumoniae*



Gonzalez-Ferrer S, *et al.* Infect. Immun. 2021



Wyres K, *et al.* Nat. Rev. Microbiol. 2020

**ecdc**  
EUROPEAN CENTRE FOR DISEASE PREVENTION AND CONTROL

RAPID RISK ASSESSMENT

**Emergence of hypervirulent *Klebsiella pneumoniae* ST23 carrying carbapenemase genes in EU/EEA countries, first update**

14 February 2024

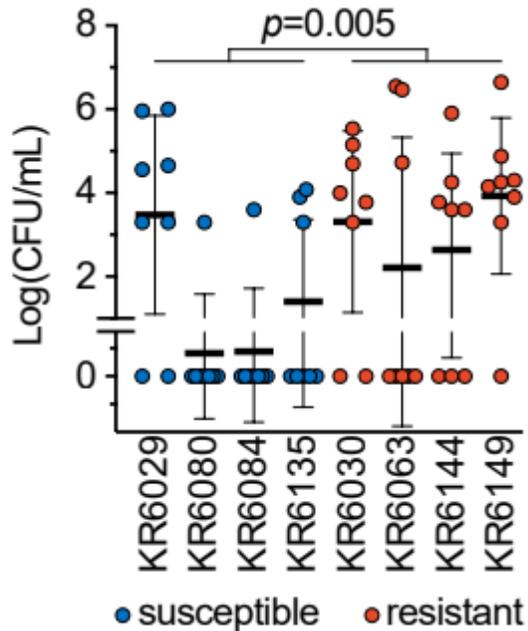
Challenge – no definitive genetic definition

# Carbapenemase-producing hypervirulent *K. pneumoniae*

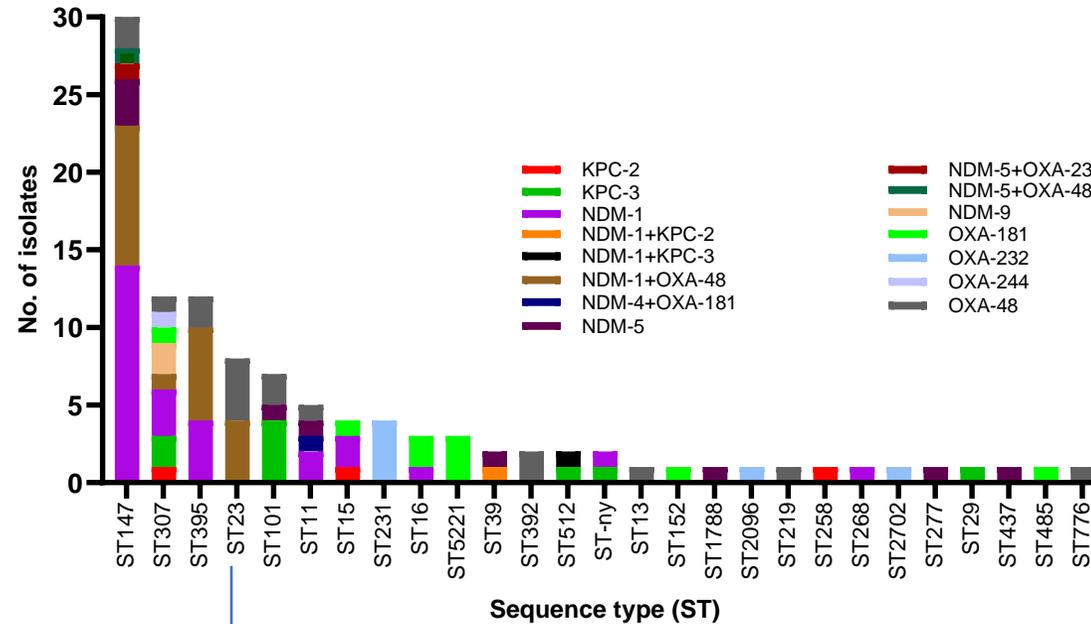
Bacteria and Bacterial Diseases

Pandrug-resistant *Klebsiella pneumoniae* isolated from Ukrainian war victims are hypervirulent

Oskar Ljungquist<sup>a,1</sup>, Michal Magda<sup>b,1</sup>, Christian G. Giske<sup>c,d</sup>, Chaitanya Tellapragada<sup>d</sup>, Oleksandr Nazarchuk<sup>c,f</sup>, Dmytro Dmytriiev<sup>c,g</sup>, Oskar Thofte<sup>h</sup>, Valdemar Öhnström<sup>h</sup>, Erika Matuschek<sup>i</sup>, Anna M. Blom<sup>b</sup>, Kristian Riesbeck<sup>h,j,k,2</sup>



Ljungquist O, *et al.* J. Infect. 2024



2024:

- No isolates interpreted genotypically as HvKp
- 8 ST23 isolates – all import Ukraine
- All had KL57 – not KL1

No evidence to suggest that ST23-KL57 causes 'hypervirulent' disease

*J Antimicrob Chemother* 2023; **78**: 1132–1134  
<https://doi.org/10.1093/jac/dkad028>  
Advance Access publication 10 February 2023

**Comment on: MDR carbapenemase-producing *Klebsiella pneumoniae* of the hypervirulence-associated ST23 clone in Poland, 2009–19**

Margaret M. C. Lam<sup>1\*</sup>, Kathryn E. Holt<sup>1,2</sup> and Kelly L. Wyres<sup>1†</sup>

# CPE from Ukraine -> Nordic countries



Letter to the Editor

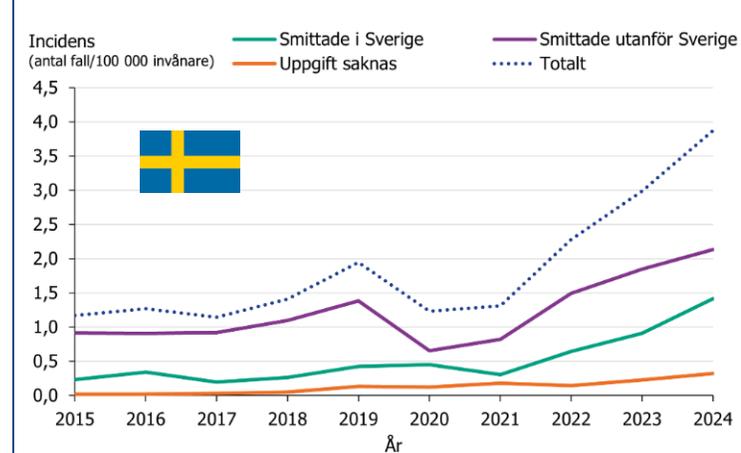
Genotypic characterisation of carbapenemase-producing organisms obtained in Denmark from patients associated with the war in Ukraine | ilar sequencing data from a recent Dutch study | comparison was made to Danish CPO isolates from a riot, to investigate nosocomial spread in Danish h

 Stolberg RS, *et al.* J. Glob. Antimicrob. Resist. 2023

Febr. 24 2022 -> Jan. 23 2023:

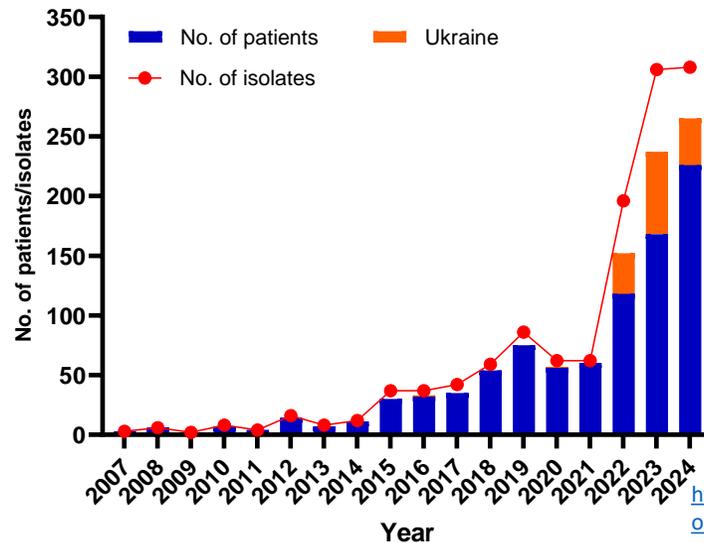
- 15% of patients
- 21% of isolates

Figur 1. Incidensen av ESBL-CARBA smittade i Sverige, utanför Sverige, där uppgift saknas samt totalt smittade under åren 2015-2024.



2024: total 410 cases - ~4% Ukraine

<https://www.folkhalsomyndigheten.se/folkhalsorapportering-statistik/statistik-a-o/sjukdomsstatistik/esblcarba/?tab=tab-report>



2024: 15% of cases -> Ukraine

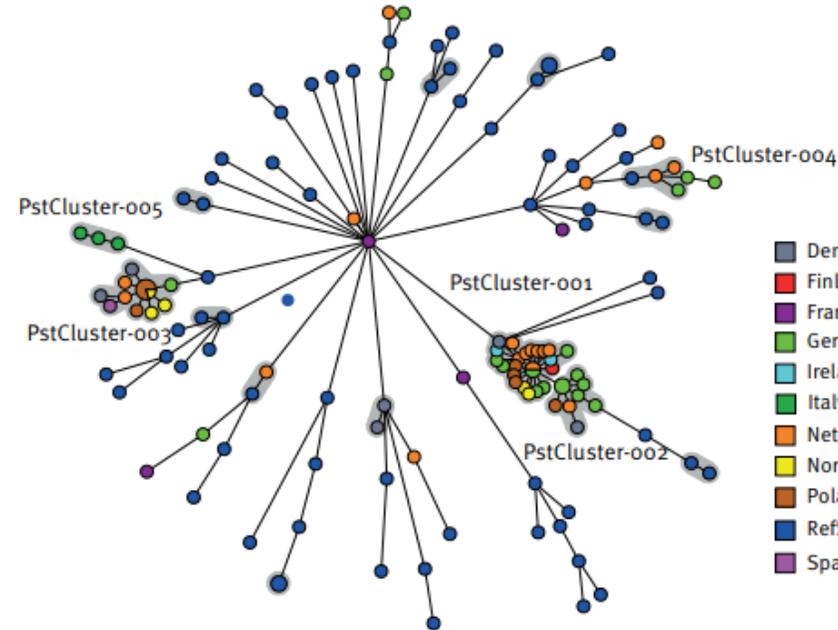
<https://www.unn.no/fag-og-forskning/k-res/nyheter/rapport-forekomst-og-molekylargenetiske-analyser-av-vre-lre-og-cpo-i-norge-2024/>

# Not only *E. coli* & *K. pneumoniae* – *P. stuartii*

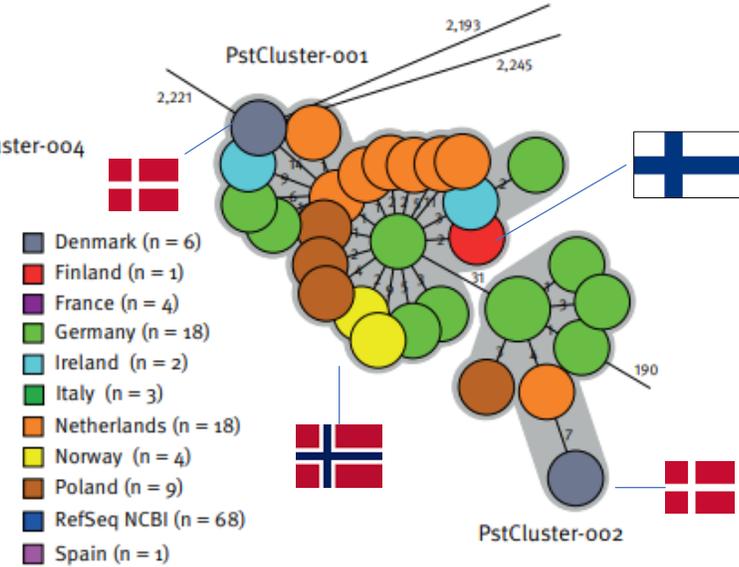
Dissemination of extensively drug-resistant NDM-producing *Providencia stuartii* in Europe linked to patients transferred from Ukraine, March 2022 to March 2023

Sandra Witteveen<sup>1</sup>, Jörg B Hans<sup>2</sup>, Radosław Izdebski<sup>3</sup>, Henrik Hasman<sup>4</sup>, Brian Samuelsen<sup>5</sup>, Laurent Dortet<sup>6</sup>, Yvonne Pfäfer<sup>7</sup>, Niall Delaghe<sup>8</sup>, Jesús Oteo-Iglesias<sup>9</sup>, Dorota Zabicka<sup>10</sup>, Martin Cormican<sup>11</sup>, Mirco Sandfort<sup>12</sup>, Felix Reichert<sup>13</sup>, Anna K Pösthöefer<sup>14</sup>, Martine A Fischer<sup>15</sup>, Nellanne Verkaik<sup>16</sup>, María Pérez-Vazquez<sup>17</sup>, Niels Pfennigwerth<sup>18</sup>, Anette M Hammerum<sup>19</sup>, Sören Hallström<sup>20</sup>, Marta Biedrzycka<sup>21</sup>, Kati Räsänen<sup>22</sup>, Cornelia CH Wielders<sup>23</sup>, Paweł Urbaniowski<sup>24</sup>, Angela de Haan<sup>25</sup>, Karin Westö<sup>26</sup>, Fabian Landman<sup>27</sup>, Han GJ van der Heide<sup>28</sup>, Simon Lansu<sup>29</sup>, Romy D Zwißlink<sup>30</sup>, Daan W Notermans<sup>31</sup>, Aneta Guzek<sup>32</sup>, Viacheslav Kondratiuk<sup>33</sup>, Aidyn Salmano<sup>34</sup>, Sebastian Haller<sup>35</sup>, Marius Linkevicius<sup>36</sup>, Sören Gatermann<sup>37</sup>, Anke Kohlenberg<sup>38</sup>, Marek Gniadkowski<sup>39</sup>, Guido Werner<sup>40</sup>, Antoni PA Hendrickx<sup>41</sup>

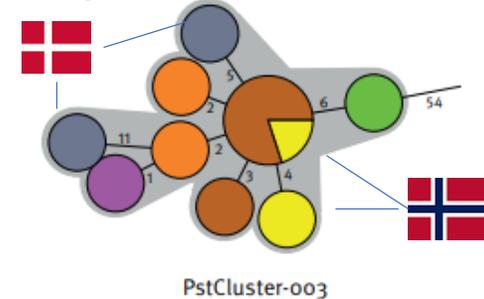
A. MST based on sequences of *P. stuartii* isolates from Europe and from the NCBI RefSeq collection



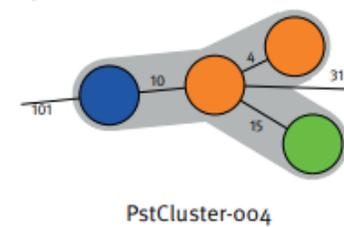
B. Genetic relationship between isolates of *bla*<sub>NDM-1</sub>-carrying PstCluster-001 and PstCluster-002



C. Relationship between *bla*<sub>NDM-5</sub>-carrying PstCluster-003 isolates



D. Relationship between *bla*<sub>NDM-1</sub>/*bla*<sub>NDM-18</sub>-carrying PstCluster-004 isolates



	Number of isolates	Mean allelic distance	Min-max distance
PstCluster-001	22	7.8	0-27
PstCluster-002	8	5.2	0-5
PstCluster-003	13	5.6	0-8
PstCluster-004	4	13.8	4-24

# Summary

- The prevalence of CPE is increasing in the Nordic countries
- War in Ukraine -> partly responsible for increase
- Dominance of NDM and OXA-48-like enzymes – Finland more KPC than the other Nordic countries
- Dominance of *K. pneumoniae* and *E. coli* high-risk clones



# Part II

## CPE: Screening and detection in the current setting

Kaisu Rantakokko-Jalava, Turku University Hospital, Finland

# What are we talking about?



## I. **Detection** of reduced susceptibility to carbapenems $\approx$ screening strains

- An isolate in routine AST
- Test indicator antibiotics (meropenem, ertapenem)
- I.1 Results beyond screening cutoffs raise suspicion of presence of carbapenemase
- I.2 The mechanism is confirmed by a separate test

## II. **Screening people** for carriage of CPE

- Methods (selective culture/molecular) with aim of searching MDR pathogens from among normal microbiota
- If appropriate colonies grow in selective culture, go to 1

# What are we talking about?

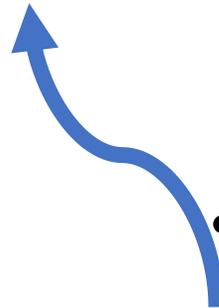


## I. **Detection** of reduced susceptibility to carbapenems $\approx$ screening strains

- An isolate in routine AST
- Test indicator antibiotics (meropenem, ertapenem)
- I.1 Results beyond screening cutoffs raise suspicion of presence of carbapenemase
- I.2 The mechanism is confirmed by a separate test

## II. **Screening people** for carriage of CPE

- Methods (selective culture/molecular) with aim of searching MDR pathogens from among normal microbiota
- If appropriate colonies grow in selective culture, go to 1



# I.1 EUCAST screening cutoffs for CPE

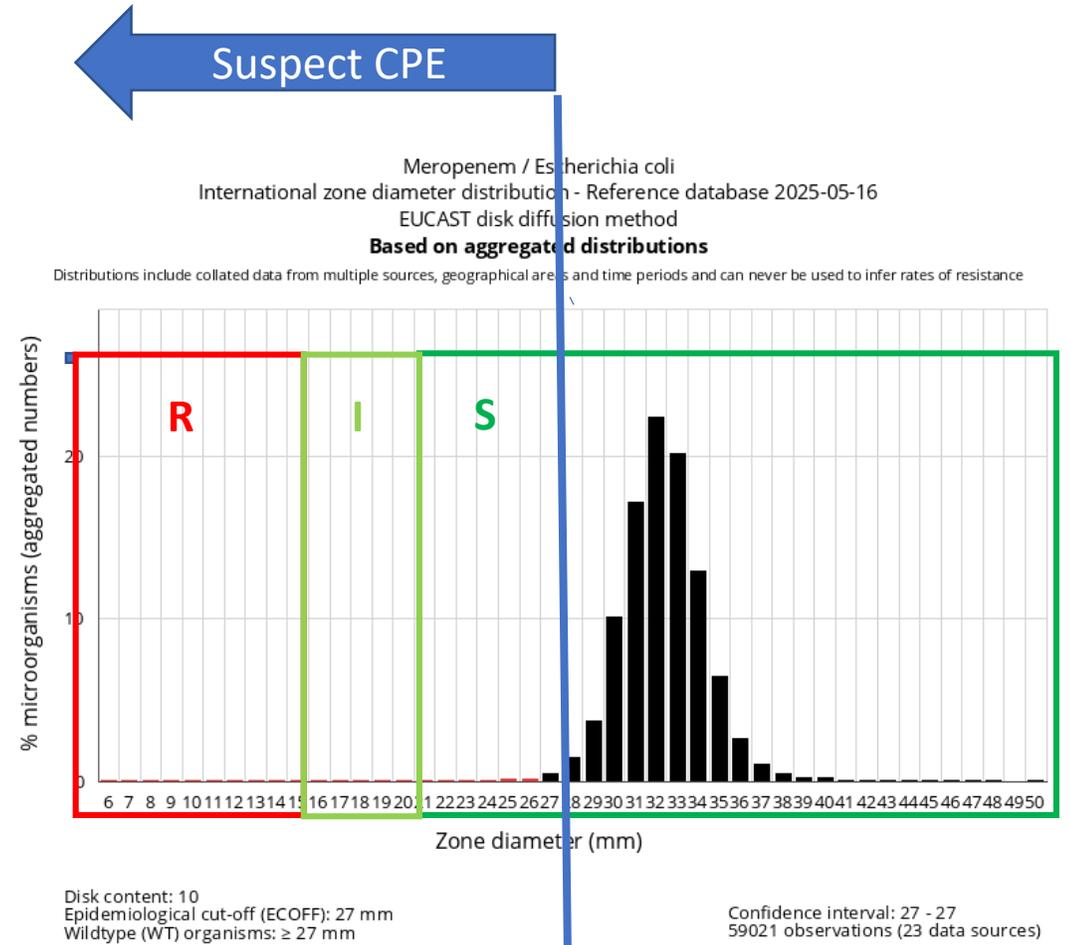
Table 1. Clinical breakpoints and screening cut-off values for carbapenemase-producing Enterobacteriaceae (according to EUCAST methodology).

Carbapenem	MIC (mg/L)		Disk diffusion zone diameter (mm) with 10 µg disks	
	S/I breakpoint	Screening cut-off	S/I breakpoint	Screening cut-off
Meropenem <sup>1</sup>	≤2	>0.125	≥22	<28 <sup>2</sup>
Ertapenem <sup>3</sup>	≤0.5	>0.125	≥25	<25

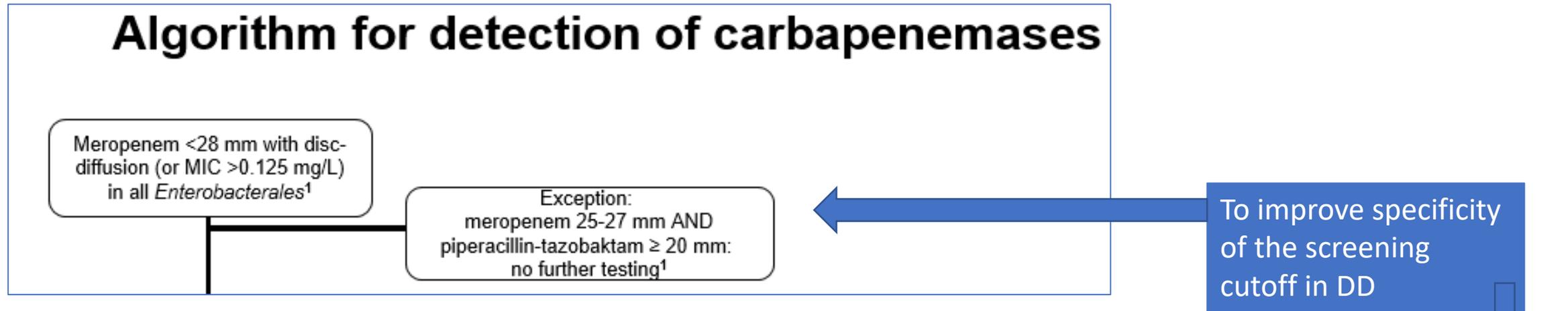
<sup>1</sup>Best balance of sensitivity and specificity

<sup>2</sup>Isolates with 25-27 mm only need to be investigated for carbapenemase-production if they are resistant to piperacillin-tazobactam and/or temocillin (temocillin contributes more to the specificity). Investigation for carbapenemases is always warranted if zone diameter of meropenem is <25 mm.

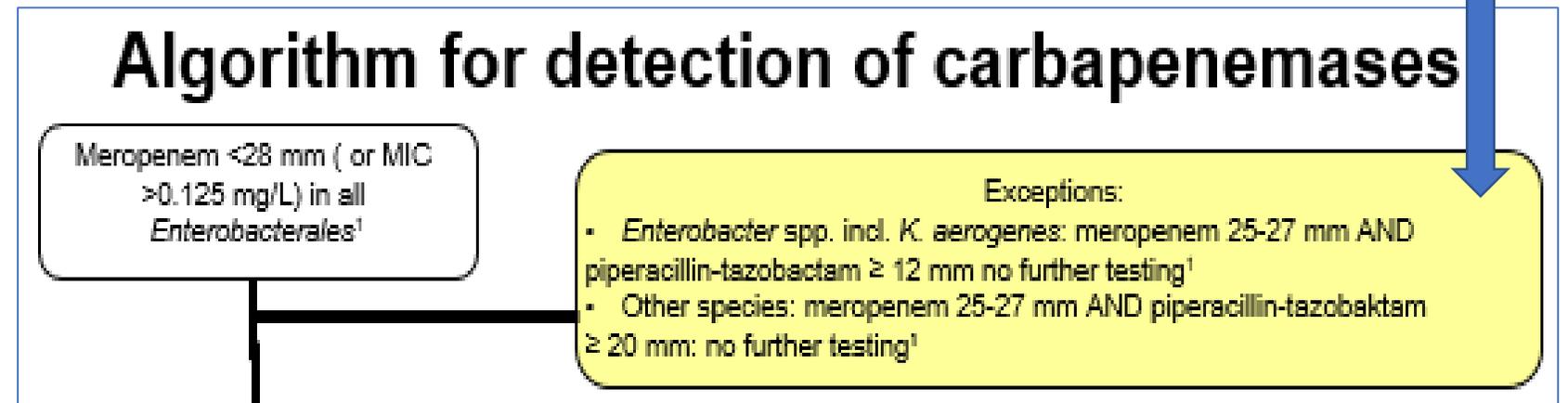
<sup>3</sup>High sensitivity but low specificity. Can be used as an alternative screening agent, but isolates with ESBL and AmpC may be resistant without having carbapenemases.



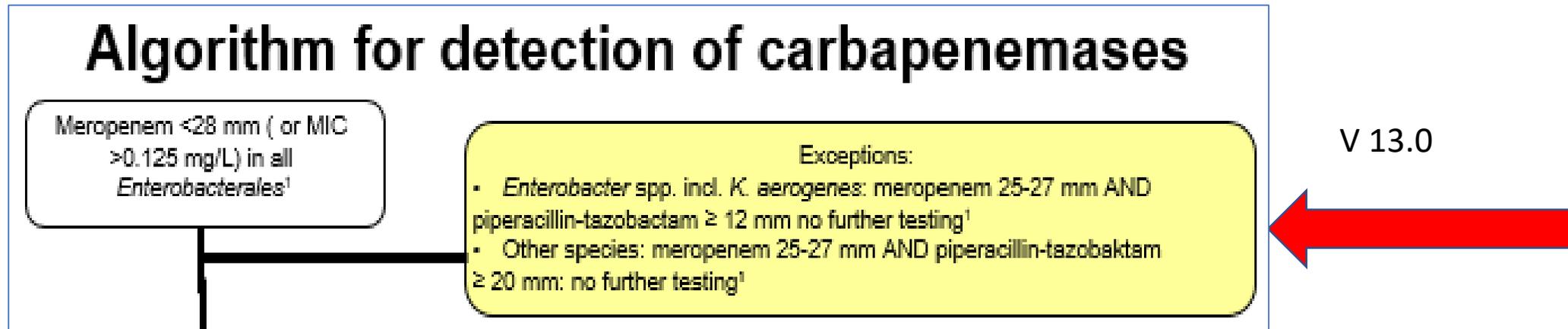
# Current EUCAST flow chart



## NordicAST



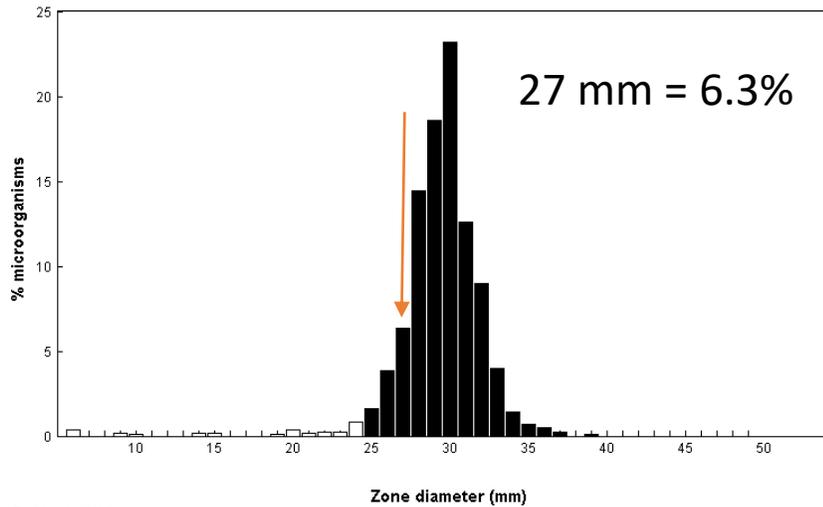
# Current NordicAST flowchart



**Purpose of the change:** To reduce the workload in the laboratories regarding the most common species with derepressed AmpC-production as the reason for meropenem < 28 mm. But at the same time reduce the risk of missing any OXA-48 positive isolates

**Meropenem / *Klebsiella pneumoniae***  
**International wild type zone diameter distribution - Reference database 2018-04-18**  
**EUCAST disk diffusion method**

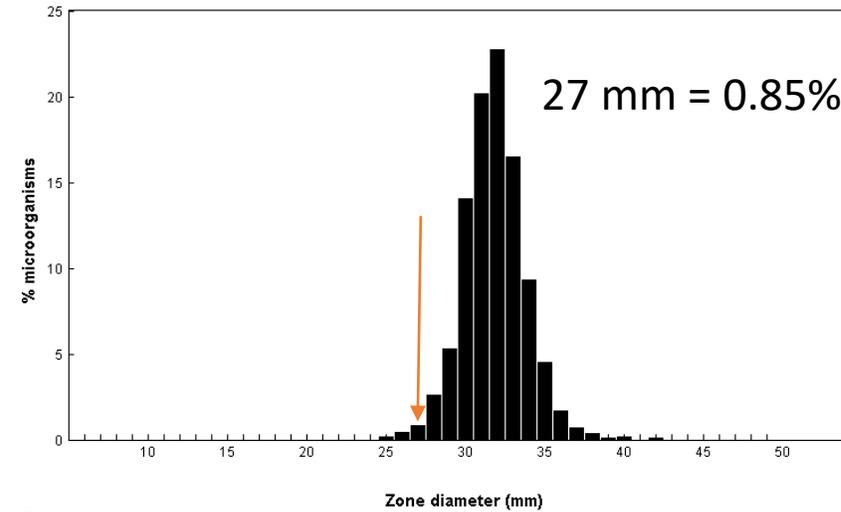
Distributions include collated data from multiple sources, geographical areas and time periods and can never be used to infer rates of resistance



Disk content: 10  
 Epidemiological cut-off (ECOFF): 25 mm (MIC = 0.125 mg/L)  
 Wildtype (WT) organisms:  $\geq 25$  mm (MIC = 0.125 mg/L)  
 1671 observations (10 data sources)

**Meropenem / *Escherichia coli***  
**International wild type zone diameter distribution - Reference database 2018-04-18**  
**EUCAST disk diffusion method**

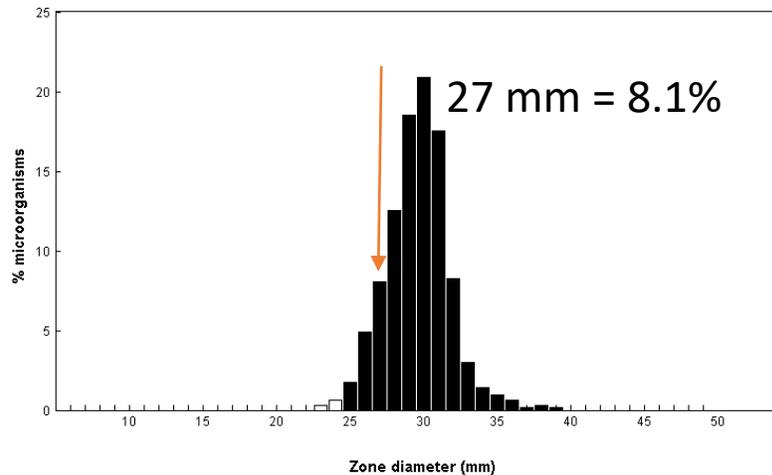
Distributions include collated data from multiple sources, geographical areas and time periods and can never be used to infer rates of resistance



Disk content: 10  
 Epidemiological cut-off (ECOFF): 25 mm (MIC = 0.125 mg/L)  
 Wildtype (WT) organisms:  $\geq 25$  mm (MIC = 0.125 mg/L)  
 11131 observations (14 data sources)

**Meropenem / *Enterobacter cloacae***  
**International wild type zone diameter distribution - Reference database 2018-04-18**  
**EUCAST disk diffusion method**

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



Disk content: 10  
 Epidemiological cut-off (ECOFF): 25 mm (MIC = 0.125 mg/L)  
 Wildtype (WT) organisms:  $\geq 25$  mm (MIC = 0.125 mg/L)  
 632 observations (7 data sources)

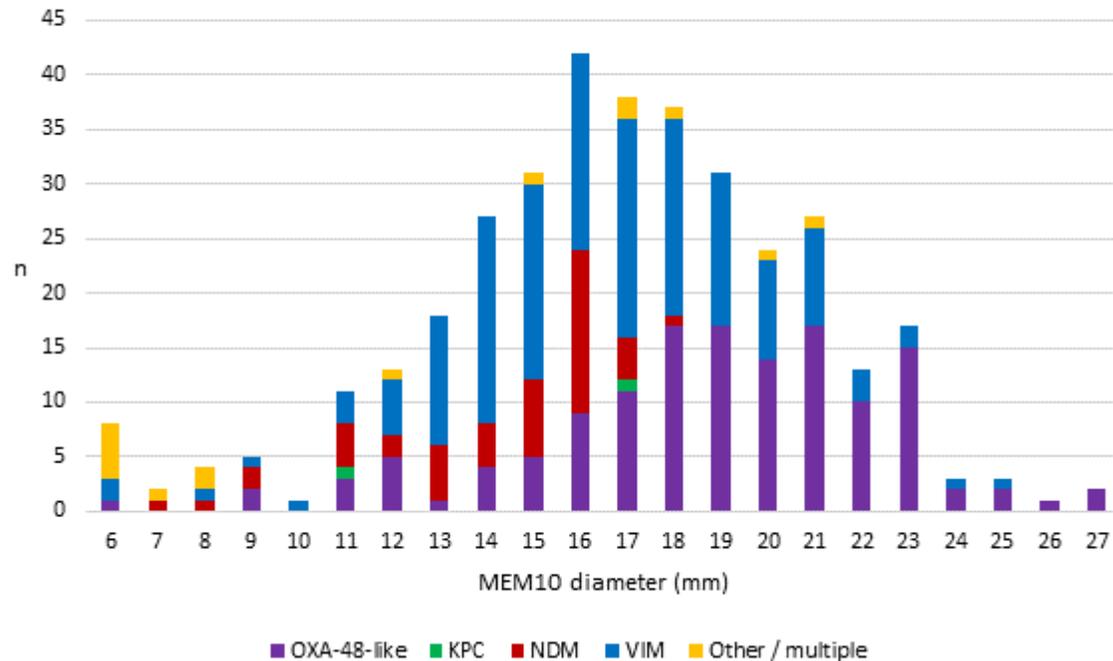
➤ ECOFF:  $\geq 25$  mm

➤ Impact – specificity of the algorithm is species dependent

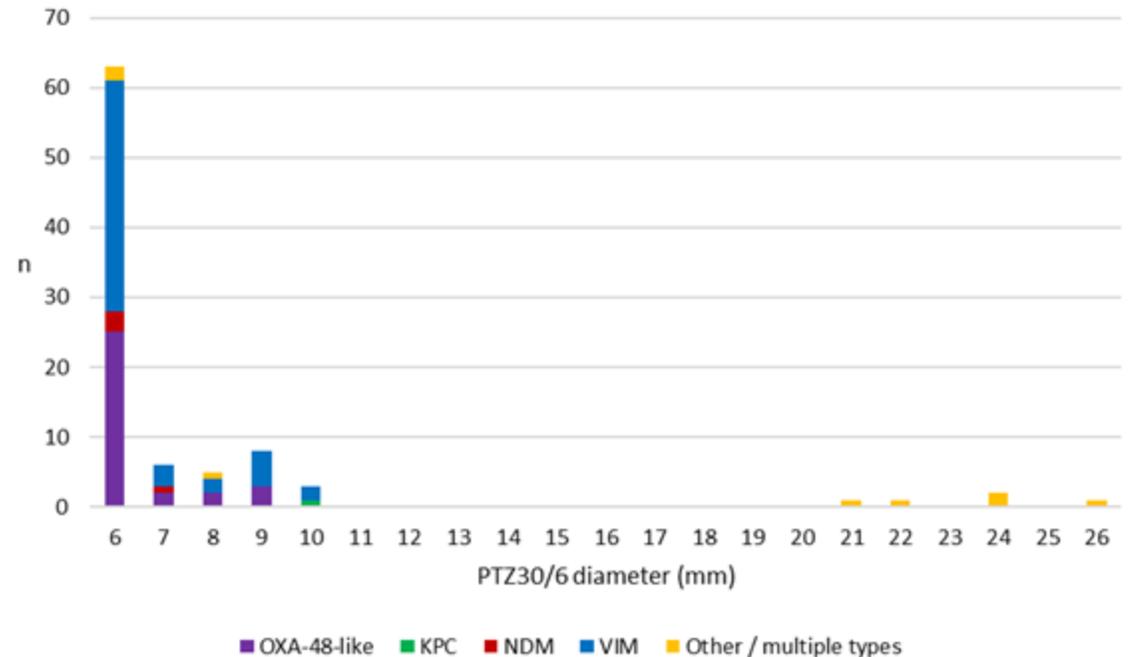
# Enterobacterales- Revised flowchart

## ESBL<sub>CARBA</sub>

*E. cloacae* complex CPE (2015-2021; n=358)



*E. cloacae* complex CPE (2018-2021; n=90)



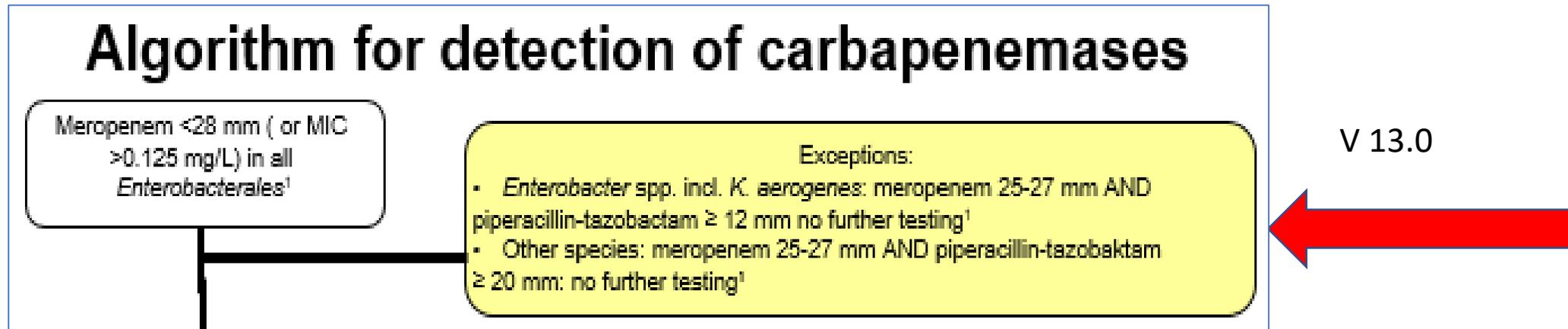
Background to the revised flowchart!

**Pr. Dr. Te-Din Daniel Huang, MD, PhD**

Chef de Laboratoire Associé / Biologie Clinique

National Reference Center for Antibiotic-Resistant Gram-Negative Bacilli

# 2023 v 13.0, Revised flowchart



The first exception does not seem to work for other *Enterobacterales* species like *Providencia* spp and *Proteus* spp. but data are very limited!

Sattler et al. (2024) tested 13 OXA-48 –producing *P. mirabilis* strains isolated in Germany, Switzerland or Austria.  
All had ertapenem and meropenem MICs >0.125 mg/L (but 10 were S to ertapenem and 11 to meropenem).  
Three were S to piperacillin-tazobactam (MIC 4, and one R had MIC 16 mg/L)

# What about sensitivity vs. current epidemiology?

*J Antimicrob Chemother* 2024; **79**: 1194–1196  
<https://doi.org/10.1093/jac/dkae054>  
Advance Access publication 27 February 2024

## **Comparison of the French novel disc diffusion-based algorithm and the current EUCAST guidelines for the screening of carbapenemase-producing Enterobacterales**

Mathilde Duque<sup>1,2</sup>, Rémy A. Bonnin<sup>1,2,3</sup>  
and Laurent Dortet <sup>1,2,3\*</sup>

518 strains collected in France 2022-23

- 364 (70.6%) CPE
- 229 (62.9%) OXA-48, of which 37 (16.9%) OXA-244

- According to the letter, EUCAST meropenem screening cutoffs
  - MEM zone <28 mm
    - sensitivity 70.6 %
  - MIC > 0.125
    - Sensitivity 96%

# What about sensitivity vs. current epidemiology?

*J Antimicrob Chemother* 2024; **79**: 1194–1196  
<https://doi.org/10.1093/jac/dkae054>  
Advance Access publication 27 February 2024

## Comparison of the French novel disc diffusion-based algorithm and the current EUCAST guidelines for the screening of carbapenemase-producing Enterobacterales

Mathilde Duque<sup>1,2</sup>, Rémy A. Bonnin<sup>1,2,3</sup>  
and Laurent Dortet <sup>1,2,3\*</sup>

518 strains collected in France 2022-23

- 364 (70.6%) CPE
- 229 (62.9%) OXA-48, of which 37 (16.9%) OXA-244

- According to the letter, EUCAST meropenem screening cutoffs
  - MEM zone <28 mm
    - sensitivity 70.6 %

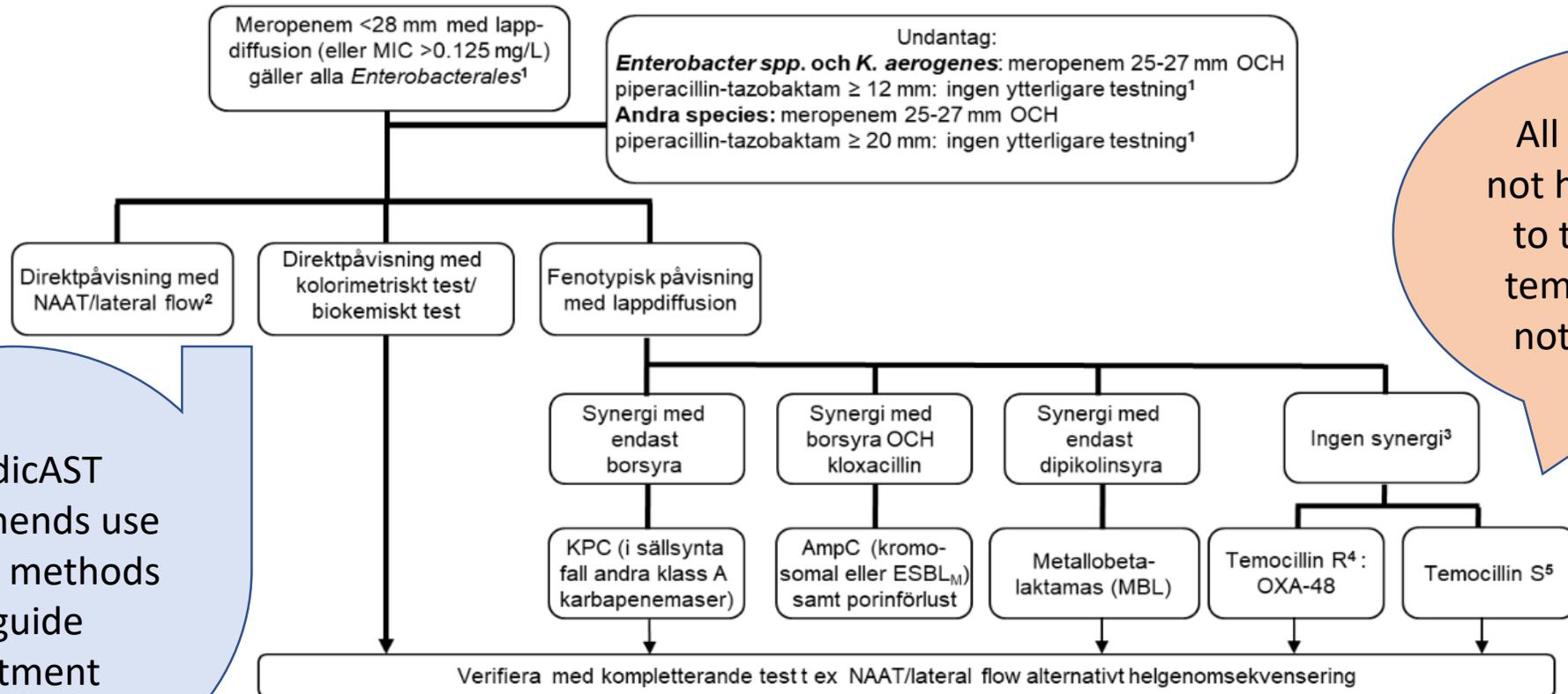
However, EDL could not reproduce MEM inhibition zones <28 mm with the same strains.

**No evidence that many strains would be missed if EUCAST method is followed.**

Yet, there may be issues with some media – disc combinations

# I.2 Confirmatory testing

## *Enterobacterales* ALGORITM FÖR DETEKTION AV KARBAPENEMASER (ESBL<sub>CARBA</sub>)



All OXA-244 are not highly resistant to temocillin, so temocillin S does not exclude CPE

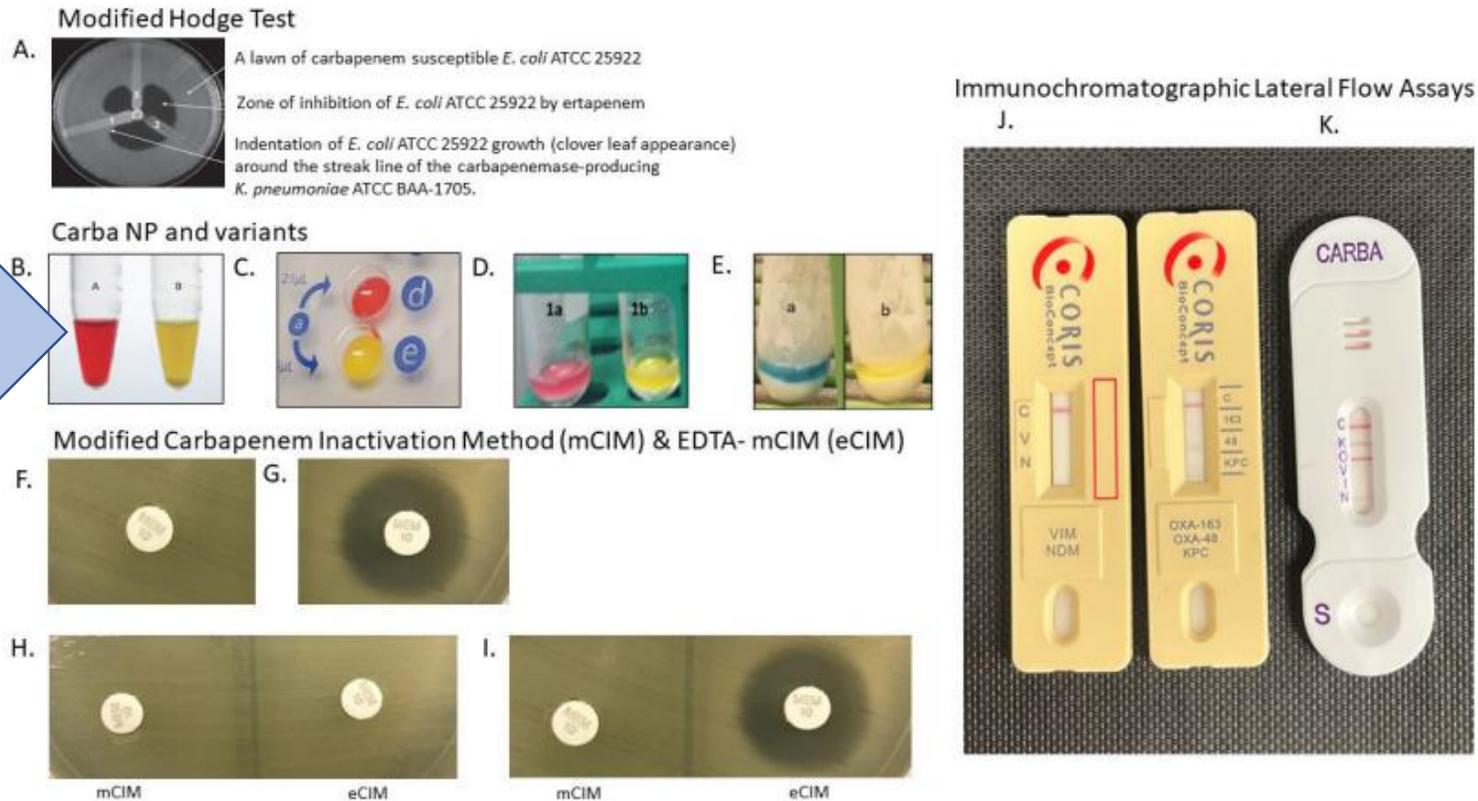
NordicAST recommends use of rapid methods to guide treatment

## Confirmation tests carbapenemases

- Combination disks or tablets (18 h)
  - Lateral flow tests
  - Colorimetric testing
  - Carbapenem inactivation method (8-10 h)
  - Molecular tests (PCR, LAMP)
- 
- Several tests are mostly performing adequately
  - Practical consideration, price, and time consumption are the main factors determining which tests are suitable to use



**Christian Giske**  
Sweden



Show carbapenemase activity;  
some tests also type; 30min-2h

CIM: tested isolate  
incubated in TSB +  
MEM disc for 2-4 h.  
Disc removed and  
placed on a plate  
inoculated with wt-  
*E. coli*

Lateral flow tests,  
≈20 min, ≈20€

**FIG 2** Phenotypic and immunochromatographic assays for the detection of carbapenemase-producing carbapenem-resistant organisms. (A) Modified Hodge Test; 1. *Klebsiella pneumoniae* ATCC BAA-1705, positive result, 2. *K. pneumoniae* ATCC BAA-1706, negative result, 3. Clinical isolate, positive result. (B) CLSI Carba NP positive result, Tube A. No imipenem added, red, Tube B. Imipenem added, yellow. (C) RAPIDEC CARBA NP (bioMérieux, Inc) positive result, Well d. No imipenem added, red, Well e. Imipenem added, yellow. (D) Neo-Rapid carba screen (Rosco Diagnostica) positive result, Tube 1a no imipenem added, red, Tube 1b imipenem added, yellow (293). (E) Rapid CARB Blue Screen® (Rosco Diagnostica), Tube a, no imipenem added, blue, Tube b, imipenem added, yellow. (F) Modified Carbapenem Inactivation Method (mCIM) positive result; G. mCIM, negative result. (H) mCIM and EDTA-mCIM (eCIM) results; positive for a serine carbapenemase producer as there is no inhibition of carbapenemase activity in the presence of EDTA. (I) mCIM and EDTA-mCIM (eCIM) results; positive for a metallo-beta-lactamase producer as there is inhibition of carbapenemase activity in the presence of EDTA. (J) RESIST-5 O.O.K.N.V. (Coris, Gembloux, Belgium) immunochromatographic lateral flow assay results for a OXA-48-like-producing *Escherichia coli*. (K) CARBA5 (NG Biotech) immunochromatographic results for a triple carbapenemase-producing (KPC, OXA-48-like and NDM) *K. pneumoniae*. The mCIM and eCIM results from panel I were used to demonstrate a positive mCIM (panel F) and a negative mCIM result (panel G). Panels A through I are reproduced from reference (121) [panel A originally from reference (122)] and panels B and F through I from reference (123), reproduced with permission of the publisher; panel D originally from reference (124)].

# CE-marked molecular tests for isolates (& rectal swabs)

Test	TAT	KPC	VIM	NDM	OXA-48	IMP	Others
Cepheid Xpert Carba-R	1 h	x	x	x	x	x	
Check MDR CT103XL (Check-Points)	6 h	x	x	x	x	x	GES, OXA-23-like, OXA-24-like, OXA-48-like, OXA-58-like, SPM, GIM
Revogene (GenePOC) Carba C	70 min	x	x	x	x	x	
Amplex Eazyplex Superbug	30 min	x	x	x	x		CTX-M
Hologic (Mobidiag) CarbaR+	80 min	x	x	x	x	x	OXA-23, OXA-24, OXA-58

Also many syndromic panels include gene tests for the most common carbapenemase groups.

# II. Screening for carriage of OXA-244

Emeraud et al. 2020

JAC

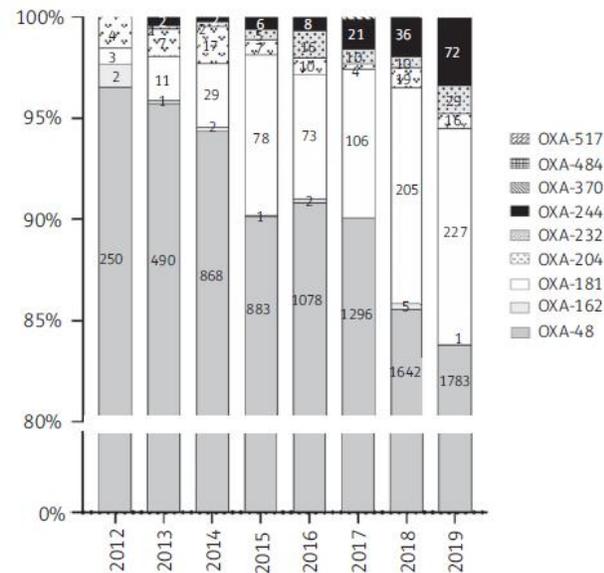


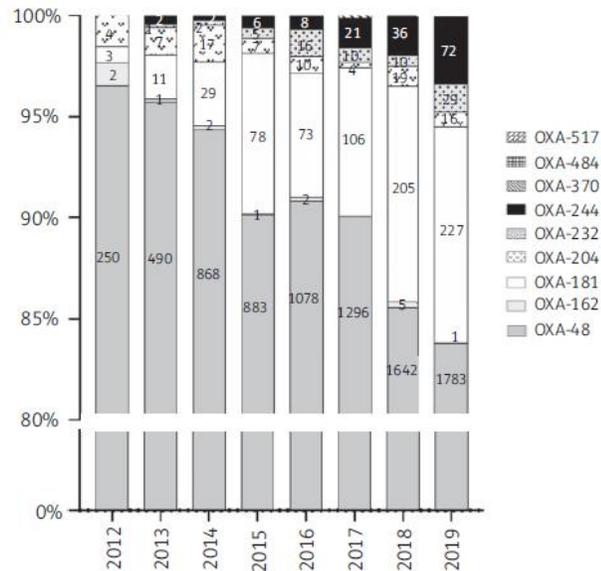
Figure 1. Relative abundance of OXA-48 variants in Enterobacteriales collected at the F-NRC from 2012 to 2019. The numbers within the bars correspond to the number of isolates of each variant.

- Among the 101 OXA-244 producers, 81 concomitantly produced an ESBL or an acquired AmpC
  - Of these, 77-78 (95-96%) grew on ESBL media
- Sensitivity (CI) of CPE media in detecting OXA-244 strains:
  - ChromID Carba Smart 14% (8.1-22.5%)
  - Brilliance CRE 54% (43.3-63.4%)
  - mSuperCARBA 99% (93.9-100%)

# Screening for OXA-244

Emeraud et al. 2020

JAC



**Figure 1.** Relative abundance of OXA-48 variants in Enterobacteriales collected at the F-NRC from 2012 to 2019. The numbers within the bars correspond to the number of isolates of each variant.

- Among the 101 OXA-244 producers, 81 concomitantly produced an ESBL or an acquired AmpC

- Of these, 77-78 (95-96%)

- Sensitivity (CI) of CPE m OXA-244 strains:

- |                       |     |              |
|-----------------------|-----|--------------|
| • ChromID Carba Smart | 14% | (8.1-22.5%)  |
| • Brilliance CRE      | 54% | (43.3-63.4%) |
| • mSuperCARBA         | 99% | (93.9-100%)  |

Concentrations of carbapenem and temocillin in media higher than the strains' MICs

Growth on plate						
Strain β-lactamase MIC temocillin	ChromID® CARBA SMART		ChromID® ESBL	Brilliance™		mSuper CARBA™
	CARBA Side	OXA Side		CRE Side	ESBL Side	
245J1 <i>E. coli</i> OXA-48 MIC >512mg/L	-	+	-	-	-	+
228D2 <i>E. coli</i> OXA-48 + ESBL MIC > 512 mg/L	-	+	+	+	+	+
221C7 <i>E. coli</i> OXA-244 MIC = 32 mg/L	-	-	-	-	-	+
226 I3 <i>E. coli</i> OXA-244 + ESBL MIC = 32 mg/L	-	-	+	-	+	+
225 E1 <i>E. coli</i> OXA-244 + ESBL MIC = 512 mg/L	-	+	+	+	+	+
226 C5 <i>K. pneumoniae</i> OXA-232 MIC = 512 mg/L	+	+	+	+	+	+

Figure S1, Emerald 2020

# Points for selecting the CPE screening media

For optimal sensitivity:

- ertapenem or meropenem at the ECOFF-levels
- $Zn^{2+}$  to promote growth of MBL
- cloxacillin to inhibit AmpC
- potentially vancomycin and amphotericin B (for gram-positives and fungi)
- A more selective agar with higher concentration of carbapenems may help finding minority variants
- A combination of two chromogenic media (plus an ESBL agar) may thus be necessary for optimal sensitivity



# Part III

## **CPE**

Which antibiotics and methods should be available in AST?

*Arnfinn Sundsfjord*

UiT The Arctic University of Norway  
K-res, University Hospital of North-Norway

NordicAST workshop 27-28 May 2025

# Agenda

- **Relevant novel antibiotics**
  - $\beta$ -lactamase inhibitors – how do they work and what is new?
  - Relevant  $\beta$ -lactamase inhibitors (BLIs) for CPE
  - Relevant  $\beta$ -lactam/ $\beta$ -lactamase inhibitor (BLBLIs) combinations for CPE
- **Antibiotics and methods for extended AST in CPE**
  - Relevant BLBLIs + cefiderocol
  - Colistin
  - Tigecycline
  - Additional aminoglycosides (amikacin)
  - Fosfomycin, (rifampicin)
- **How to report AST results?**
  - What about meropenem susceptible CPE?

# β-lactamase inhibitors – how do they work and what is new?

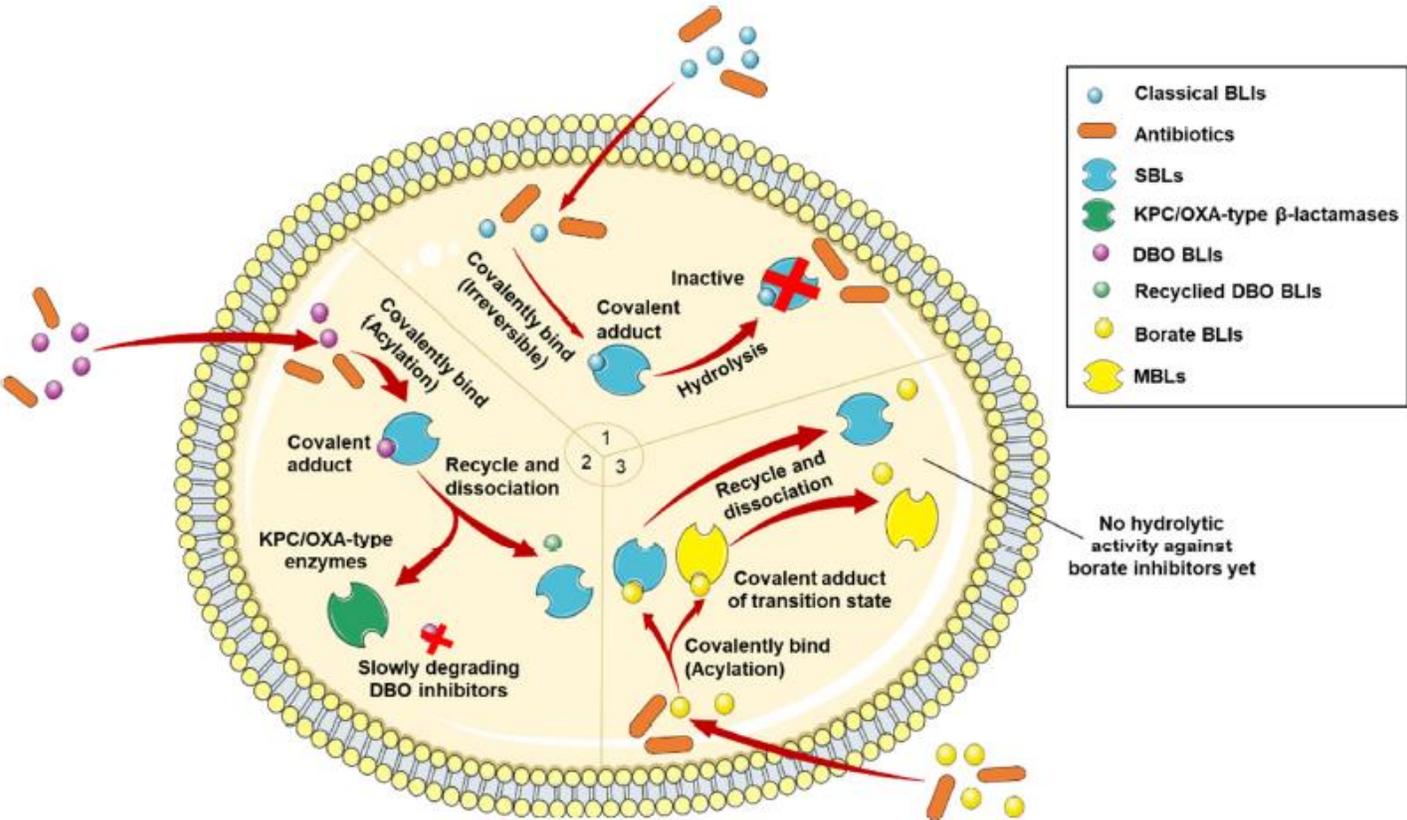


Figure 3. Main inhibitory mechanisms of β-lactamase inhibitors (BLIs). 1: The classical BLIs are

# $\beta$ -lactamase inhibitors (BLIs) - 1

- BLIs target the active sites of beta-lactamases and inhibit the formation of complexes between  $\beta$ -lactams (BLAs) and  $\beta$ -lactamases
- **Early BLIs – classical penicillin-based BLIs**
  - Clavulanic acid, sulbactam, and tazobactam
  - Involve the competitive, irreversible binding of serine residues at the  $\beta$ -lactamase active site resulting in enzyme inactivation
  - Suicidal activity
  - Targets Ambler class A  $\beta$ -lactamases (TEM, SHV, and CTX-M..)
  - Enmetazobactam
    - Inhibitor activity against class A  $\beta$ -lactamases

# $\beta$ -lactamase inhibitors (BLIs) - 2

- **First generation non-BLIs – diazabicyclooctanes (DBOs)**
  - Avibactam, relebactam, zidebactam, durlobactam
  - Non-betalactam compounds that reversibly acylate the active site of serine residues in  $\beta$ -lactamases
  - Deactivate class A, C, and D (except relebactam)  $\beta$ -lactamases
  - Some (zidebactam and durlobactam) have an additional antibacterial activity (anti-PBP2) – *betalactam enhancer*
- **In combination with various  $\beta$ -lactams**
  - Avibactam-ceftazidime/aztreonam
  - Relebactam-imipenem
  - Zidebactam-cefepime
  - Durlobactam-sulbactam

# $\beta$ -lactamase inhibitors (BLIs) - 3

- **Second generation non-BLIs – boronic acid derivatives**
  - Vaborbactam, taniborbactam
  - Transition state analogs that can reversibly bind to the active sites and competitively inhibit  $\beta$ -lactamases
- *Vaborbactam*
  - Deactivate class A and C  $\beta$ -lactamases
  - Combination drug: meropenem-vaborbactam
- *Taniborbactam*
  - Deactivate class A, B (exceptions), C, and D  $\beta$ -lactamases
  - Combination drug: cefepime-taniborbactam

# Relevant BLI and their inhibitor activities

## 1.1 *Enterobacterales* – inhibitor activity of relevant BLIs on clinically relevant $\beta$ -lactamases<sup>1</sup>

$\beta$ -lactamase inhibitor	$\beta$ -lactam partner	ESBL ( <i>bla</i> <sub>CTX-M</sub> )	Chromosomal/plasmid-mediated AmpC ( <i>bla</i> <sub>CMY</sub> , <i>bla</i> <sub>DHA</sub> )	Class A carbapenemases ( <i>bla</i> <sub>KPC</sub> , <i>bla</i> <sub>IMP</sub> )	Class B carbapenemases ( <i>bla</i> <sub>NDM</sub> , <i>bla</i> <sub>VIM</sub> , <i>bla</i> <sub>IMP</sub> )	Class D carbapenemases <sup>2</sup> ( <i>bla</i> <sub>OXA-48</sub> , <i>bla</i> <sub>OXA-181</sub> , <i>bla</i> <sub>OXA-244</sub> )
Clavulanic acid <sup>3</sup>	Amoxicillin	+	-	-	-	-
Tazobactam	Piperacillin	+	-	-	-	-
Tazobactam	Ceftolozane	+	-	-	-	-
Enmetazobactam	Cefepime	+	-	+/-	-	-
Avibactam	Ceftazidime	+	+	+	-	+
Avibactam	Aztreonam	+	+	+	- <sup>4</sup>	+
Relebactam <sup>5</sup>	Imipenem	+	+	+	-	-
Vaborbactam	Meropenem	+	+	+	-	-
Taniborbactam <sup>6</sup>	Cefepime	+	+	+	+ <sup>7</sup>	+
Zidebactam <sup>6</sup>	Cefepime	+	+	+	+	+

<sup>1</sup> +: inhibitory activity; -: No clinically useful inhibitory activity. Note that the activity of inhibitor can vary within and between variants in each  $\beta$ -lactamase group.

<sup>2</sup> Class D carbapenemases mainly associated with *A. baumannii* (e.g. OXA-23-like, OXA-24/40-like and OXA-58-like) are rare among *Enterobacterales*, except for some reports mainly in *P. mirabilis*. The clinical effect of avibactam and taniborbactam is uncertain against these  $\beta$ -lactamases.

<sup>3</sup>  $\beta$ -lactam- $\beta$ -lactamase inhibitor combination relevant for *E. coli*, *Klebsiella* spp. (except *K. aerogenes*), *Raoultella* spp. and *P. mirabilis*.

<sup>4</sup> Avibactam has not inhibitory activity against MBLs. However, aztreonam is not a substrate for MBLs.

<sup>5</sup> BLBLI relevant for *Enterobacterales* except *Morganellaceae* which have reduced susceptibility to imipenem.

<sup>6</sup> BLBLIs in phase 3 clinical trials.

<sup>7</sup> Limited in vitro inhibitory activity observed against some subclass B1  $\beta$ -lactamases such as IMP and SIM, subclass B2 (PFM-like) and B3 (AIM-like). NDM- (e.g. NDM-9 and NDM-30) and VIM-variants (e.g. VIM-83) resistant to the inhibitory activity is described.

# Relevant BLBLIs and their potential clinical efficacy

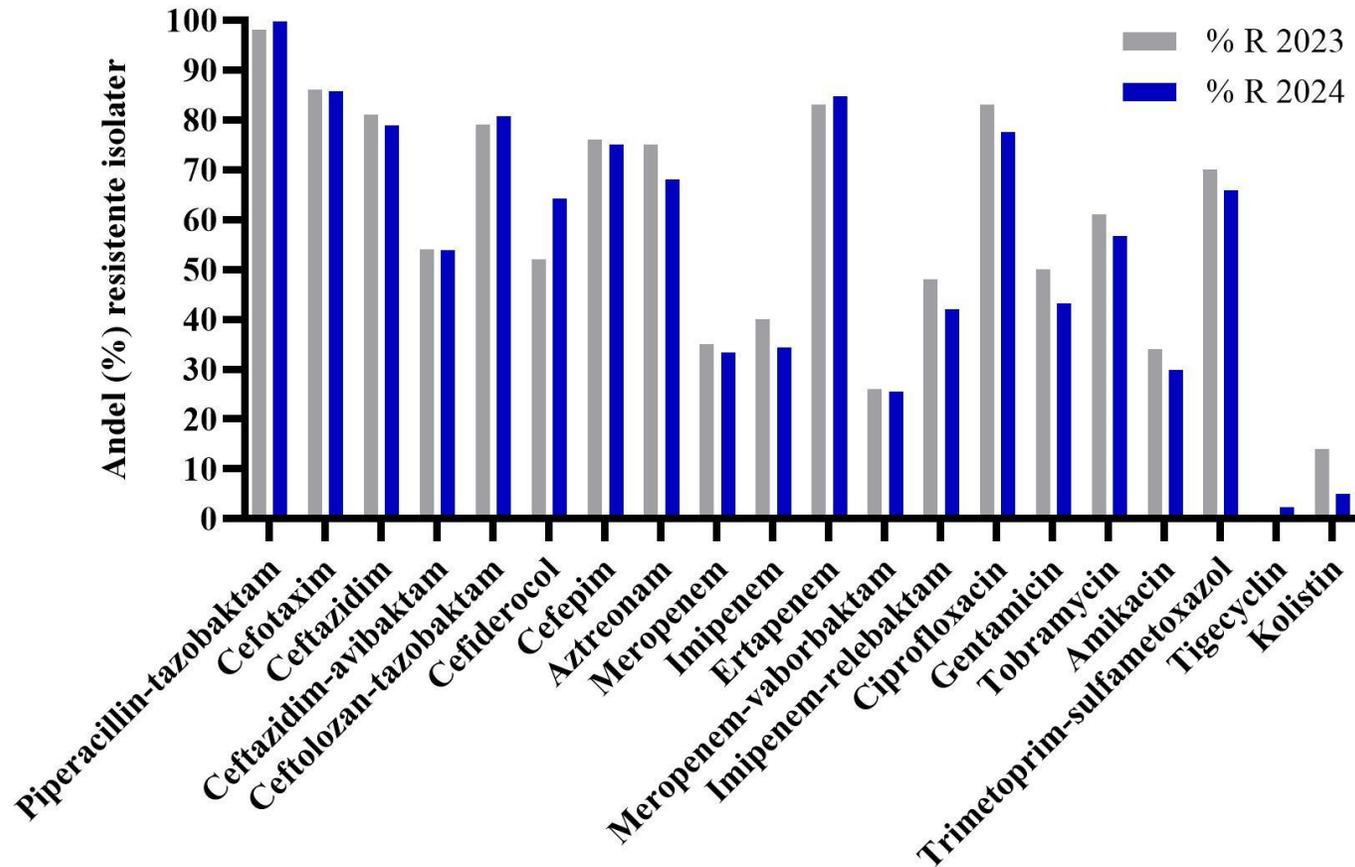
## 2.1 Enterobacterales – potential clinical efficacy of cefiderocol and relevant approved BLBLIs against clinically important resistance mechanisms

Antimicrobial	ESBL	Chromosomal-/ plasmid-AmpC	Class A carbapenemases	Class B carbapenemases	Class D carbapenemases ( <i>bla</i> <sub>OXA-48-like</sub> )	Class A+B/A+D/B+D carbapenemases
Amoxicillin - clavulanic acid <sup>1</sup>	+	-	-	-	-	-
Piperacillin - tazobactam	+	-	-	-	-	-
Ceftolozane - tazobactam	+	-	-	-	-	-
Ceftazidime - avibactam	+	+	+	-	+	-
Meropenem - vaborbactam	+	-	+	-	-	-
Imipenem - relebactam	+	+	+	-	-	-
Aztreonam - avibactam	+	+	+	+	+	+
Cefepime - enmetazobactam	+	+	-	-	+	-
Cefiderocol	+	+	+	(+)	+	+

<sup>1</sup> relevant for *E. coli* and *K. pneumoniae*. Not relevant for AmpC-producing Enterobacterales.

# CPE – co-resistance profile

*K-res data 2023 and 2024 (n=237 and 265)*



**Co-resistance profile is strongly linked to carbapenemase class**

	Andel (%) R		
	OXA-48-lik	NDM/NDM+cpase	KPC
Cefotaxim	65	100	100
Ceftazidim	49	100	100
<b>Ceftazidim-avibaktam</b>	<b>0</b>	<b>99</b>	<b>0</b>
<b>Cefiderocol</b>	<b>13</b>	<b>65</b>	<b>29</b>
Cefepim	44	99	93
Aztreonam	54	78	100
Meropenem	13	48	71
<b>Meropenem-vaborbaktam</b>	<b>9,4</b>	<b>42</b>	<b>0</b>
Imipenem	7,1	53	71
<b>Imipenem-relebaktam</b>	<b>7,9</b>	<b>76</b>	<b>0</b>
Ertapenem	69	98	93
Ciprofloxacin	47	93	79
Gentamicin	30	55	50
Tobramycin	34	73	71
Amikacin	11	45	43
Trimetoprim-sulfametoxazol	49	83	43
Kolistin	1,6	6,8	14

Source: Ørjan Samuelsen, K-res.

\*Cefiderocol: 37 % ATU (21-23 mm, R <23 mm) – interpreted as R

# Antibiotics and methods for extended AST in CPE

- Relevant BLBLIs and cefiderocol
- Colistin
  - Broth microdilution
- Tigecycline
  - Breakpoints only for *Escherichia coli*
- Alternative aminoglycosides (amikacin)
- Fosfomycin
  - Oral versus intravenous
  - Breakpoints only for *Escherichia coli*
- (Rifampicin)

# How to report AST-results?

- **What about meropenem susceptible CPE?**
  - S - report as tested
  - S - with a warning
  - R

**Thanks!**



