




Genetic diversity among selected ESBL and Carbapenem-producing *Klebsiella pneumoniae* isolates from urocultures in a portuguese hospital

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Antibiotic resistance – A public health problem

ANTIBIOTICS: HANDLE WITH CARE 

BEFORE USING ANTIBIOTICS

THINK TWICE


FIRST
Is it necessary?

SECOND
Is it appropriate?

- ✓ Right drug
- ✓ Right time
- ✓ Right dose
- ✓ Right duration

ANTIBIOTIC RESISTANCE can lead to increased DEATHS.
Step into change: THINK TWICE.

antibioticawarenessweek.org
#AntibioticsThinkTwice
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 World Health Organization
Western Pacific Region

ANTIBIOTICS ARE LOSING THEIR POWER BECAUSE OF MISUSE



BE PART OF THE SOLUTION

- ✓ Always take the full prescription, even if you feel better
- ✓ Never share or use leftover antibiotics
- ✓ Never buy antibiotics without a prescription

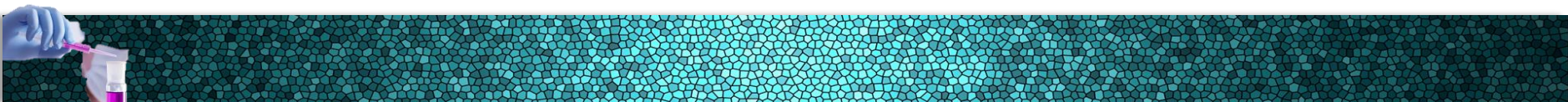
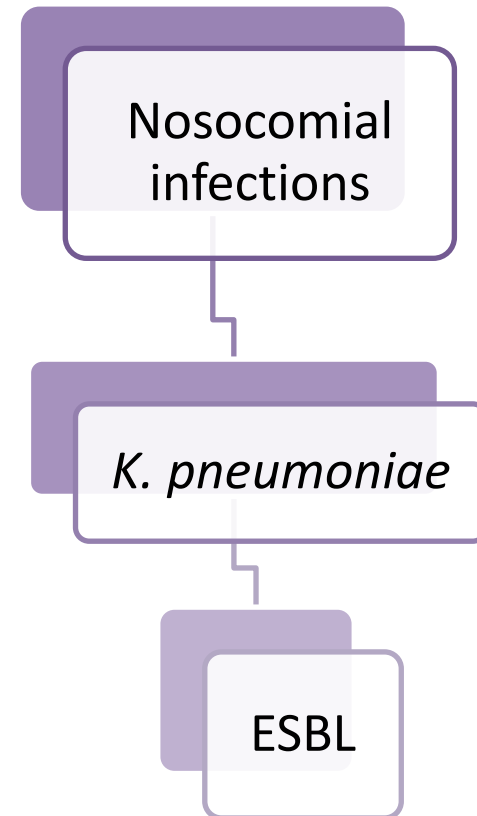
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Fig. 1 and 2 – Campaigns related to the two main factors for antibiotic resistance: overuse and misuse of antimicrobials (WHO, 2018)

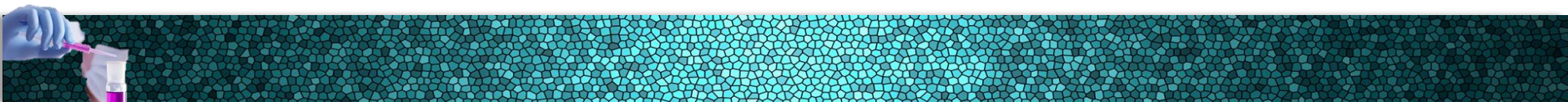


Klebsiella pneumoniae

- Major pathogen implicated in nosocomial infections that is known to spread easily;
- Frequently associated with resistance to the highest-priority critically important antimicrobials.



- ✓ Determine the carriage rate of ESBL-producing *K. pneumoniae* in a hospital in Portugal;
- ✓ Analyze the type of enzymes implicated;
- ✓ Determine the genetic diversity (MLST) among selected carbapenem- and ESBL-producing *K. pneumoniae* isolates from human urinary infections.



Sampling



49 CTX/CAZ-resistant *K. pneumoniae* isolates obtained aleatory from patients' urocultures in a Portuguese hospital; December 2016-September 2018.

Bacteria isolation



MacConkey agar and BHI agar

Maldi-TOF

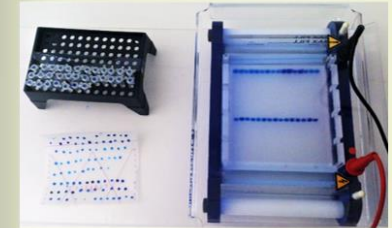


Matrix-Assisted Laser Desorption/Ionization



13 different antibiotics according to CLSI, 2019⁶

PCR and sequencing

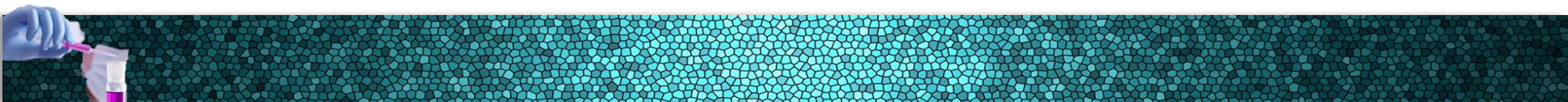


Presence of *bla*_{CTX-M} (different groups), *bla*_{SHV}, *bla*_{TEM}, *bla*_{KPC}, *bla*_{NDM}, *bla*_{VIM}, *tetA* and *tetB* genes



General resistance phenotype

- ESBL-production was detected in 26.5% of the isolates (13/49);
- Most of them carried the gene of CTX-M-15 enzyme (n=10);
- It is important to note that all ESBL-positive and negative isolates carried the $KPC_{2/3}$ gene and showed carbapenem resistance.



Resistance phenotype

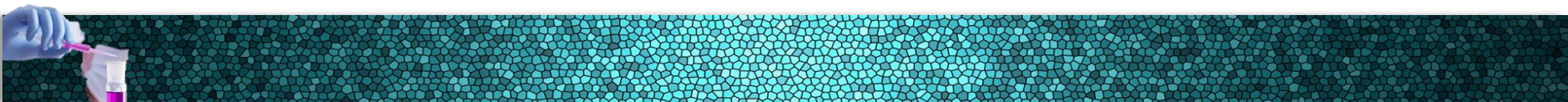
Table 1 – Resistance phenotype and genotype associated with different sequence types (ST) for selected *K. pneumoniae* isolates from urocultures in a Portuguese hospital.

Sample	Date	ESBL ^b	Resistance phenotype ^a	Resistance genotype	MLST ^c
X2142	15/12/2016	P	AMC, FOX, CTX, CAZ, CHL, CIP, CN, SXT, S, IMP, MRP, ERT	KPC-2/3, SHV-12, TEM	ST147
X2143	15/12/2016	P	AMC, FOX, CTX, CAZ, CIP, CN, SXT, S, TET, IMP, MRP, ERT	CTX-M-15, KPC-2/3, SHV-27, TEM, <i>tetA</i>	ST280
X2157	27/04/2017	P	AMC, FOX, CTX, CAZ, CHF, CIP, CN, SXT, S, IMP, MRP, ERT	CTX-M-15, KPC-2/3, SHV-28, TEM	ST15
X2165	25/05/2017	P	AMC, CTX, CAZ, CIP, CN, SXT, IMP, MRP, ERT	KPC-2/3, SHV-28, TEM	ST15
X2175	10/06/2018	P	AMC, CTX, CAZ, CIP, SXT, S, IMP, MRP, ERT	CTX-M-15, KPC-2/3, SHV-12, TEM	ST15
X2232	20/01/2017	P	AMC, CTX, CAZ, CIP, CN, SXT, S, TET, IMP, MRP, ERT	KPC-2/3, SHV-27, TEM, <i>tetA</i>	ST280
X2168	20/05/2018	N	AMC, CTX, CAZ, CN, SXT, S, IMP, MRP, ERT	KPC-2/3, SHV-11, TEM	ST348
X2173	20/05/2018	N	AMC, FOX, CTX, CAZ, IMP, MRP, ERT	KPC-2/3, SHV-26, TEM	ST34

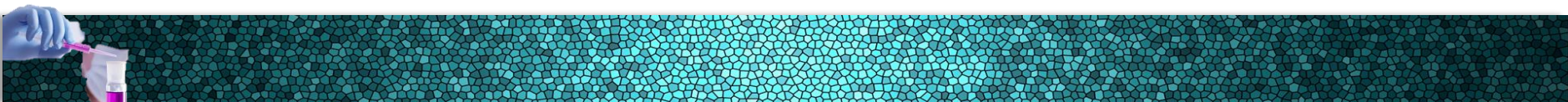
Legend: ^aAMC: amoxicillin+clavulanic acid; FOX: ceftazidime; CTX: cefotaxime; CAZ: ceftazidime; CHL: chloramphenicol; CIP: ciprofloxacin; CN: gentamicin; SXT: trimethoprim + sulfamethoxazole; S: streptomycin; TET: tetracycline; IMP: imipenem; MRP: meropenem; ERT: ertapenem;

^bP – Positive, N- Negative;

^cMLST - MultiLocus Sequence Typing.

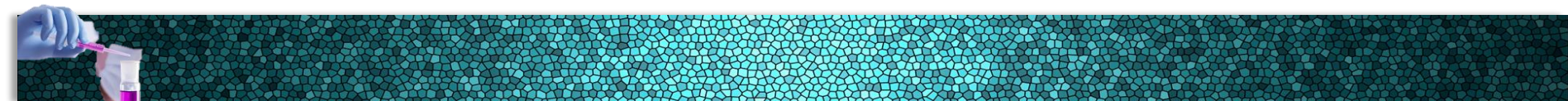


- ✓ These findings indicate the genetic diversity among urinary infections isolates in our hospital.
- ✓ The KPC2/3 is the main mechanism of carbapenem resistance in *K. pneumoniae* isolates in the studied period, frequently detected together with CTX-M-15 gene.
- ✓ Three different ST were detected among ESBL-producing *K. pneumoniae* isolates (ST15, ST147 and ST280).



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Thank you for your attention!



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