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699. Relationship Between Klebsiella pneumoniae Antimicrobial Resistance and Biofilm Formation

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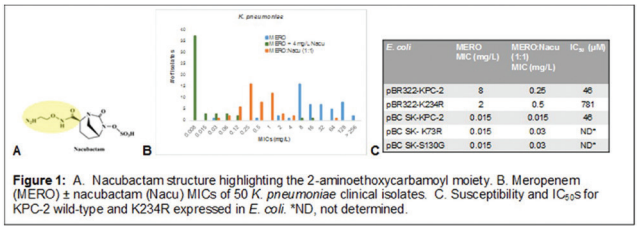
combinations based on the breakpoint of MERO. The strains harboring K73R, S130G, and K234R had slightly elevated MERO-nacubactam MICs relative to wild type but did not have corresponding increases in MERO MICs. Strains with pBC SK-KPC2, K73R or S130G had 0.015 mg/L MERO MICs. The pBR322-K234R strain had a twofold lower MERO MIC than pBR322-KPC-2 (Figure 1C). The IC_{50} of cell extracts containing the K234R variant is 781 μM, which is 12-fold higher than that for KPC-2 (66 μM) (Figure 1C). Extracts containing the S130G variant were not inhibited by nacubactam (IC_{50} > 2.6 mM).

Conclusion. Meropenem-nacubactam is an effective β-lactam β-lactamase inhibitor combination for Enterobacteriaceae with KPC or OXA-48 β-lactamases. The single amino acid substitutions K73R, S130G, and K234R in KPC-2 affect the inactivation mechanism.



E. coli	MERO MIC (mg/L)	MERO:Nacu (1:1) MIC (mg/L)	IC_{50} (μM)
pBR322-KPC-2	8	0.25	46
pBR322-K234R	2	0.5	781
pBC SK-KPC-2	0.015	0.015	46
pBC SK- K73R	0.015	0.03	ND*
pBC SK-S130G	0.015	0.03	ND*

Figure 1: A. Nacubactam structure highlighting the 2-aminoethoxycarbamoyl moiety. B. Meropenem (MERO) ± nacubactam (Nacu) MICs of 50 *K. pneumoniae* clinical isolates. C. Susceptibility and IC_{50}s for KPC-2 wild-type and K234R expressed in *E. coli*. *ND, not determined.

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699. Relationship Between *Klebsiella pneumoniae* Antimicrobial Resistance and Biofilm Formation

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Background. *Klebsiella pneumoniae* is a frequently multidrug-resistant organism with a high propensity to form biofilm. *K. pneumoniae* is the most common carbapenem-resistant *Enterobacteriaceae* (CRE), and labeled an urgent threat by the CDC. The relationship between *K. pneumoniae* biofilm formation and specific antimicrobial resistance patterns has not been well defined.

Methods. *K. pneumoniae* isolates (n = 139) were evaluated for antimicrobial resistance and biofilm formation (CDC, Providence VA Med. Ctr., Rhode Island Hosp., BEI, and ATCC). Susceptibility was based predominantly on 2017 CLSI (Clinical and Laboratory Standards Institute) breakpoints. Isolates were categorized as multidrug-resistant (MDR: resistant to ≥ 1 antimicrobial in ≥ 3 out of 16 antimicrobial categories) or extensively drug-resistant (XDR: resistant to ≥ 1 antimicrobial in all but ≤ 2 out of 16 antimicrobial categories) based on expert consensus criteria for *Enterobacteriaceae* (European CDC (ECDC)/CDC, 2012). We collapsed antimicrobial categories described by the ECDC/CDC consensus group into nine categories: penicillins, cephalosporins, monobactam, carbapenems, protein synthesis inhibitors, fluoroquinolones, folate pathway inhibitors, fosfomycin, and colistin. Biofilm formation was assessed using a modified crystal violet method (OD_{570}) and defined by tertile cut-points. Antimicrobial resistance was compared for weak (n = 47) vs. strong (n = 46) biofilm formation by chi-square or Fisher's exact test. Predictors of strong biofilm formation were identified using logistic regression.

Results. MDR isolates were more common among weak (n = 46/47, 97.9%) vs. strong biofilm formers (n = 35/46, 76.1%; P = 0.002), whereas XDR was similar between groups (n = 12/47, 25.5% vs. n = 13/46, 28.3% P = 0.77). Resistance to penicillins, cephalosporins, monobactams, carbapenems, protein synthesis, or fluoroquinolones was more common among weak biofilm formers (P < 0.05). Carbapenem resistance was inversely associated with strong biofilm formation (odds ratio 0.09; 95% confidence interval 0.02–0.33).

Conclusion. Carbapenem-resistant *K. pneumoniae* was 91% less likely to form strong biofilm. Potential trade-off mechanisms between antimicrobial resistance and biofilm formation require further exploration.

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700. Identification and Whole-Genome Sequencing (WGS) of Meropenem-Vaborbactam (MV) Resistant *Klebsiella pneumoniae* (MVRKP) Among Patients Without Prior Exposure to MV: Collateral Damage

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Background. MV is a newly approved β-lactam/β-lactamase inhibitor combination (BLIC) for the treatment of complicated urinary tract infections (cUTI). Vaborbactam is a cyclic boronic acid BLI that was mainly developed as a potent inhibitor of KPC carbapenemases and other Ambler class A&C enzymes. Vaborbactam is inactive against metallo-β-lactamases (MBL) and certain Class D enzymes (e.g. OXA-2 and OXA-48). We encountered a case of MV-resistant *Klebsiella pneumoniae* (MVRKP)and sought to explore the various mechanisms of MV resistance within KP.

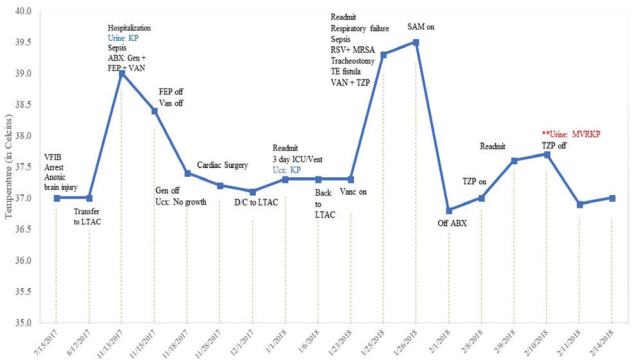
Methods. A 65-year-old nursing home resident with multiple prior hospitalizations and recent exposure to antibiotics (Timeline) developed sepsis secondary to carbapenem-resistant *Klebsiella pneumoniae* (CRKP) cUTI. WGS of the patient's isolate was performed. This was followed by random screening for MV resistance and WGS of other isolates from a historical database.

Results. Results of WGS are seen in the table below. Sequencing of our patient's isolate revealed strain ST258 with a premature stop in aa89 of OmpK35 as well as insertions at Gly134 and Asp135 (i.e., the GD repeat) of OmpK36. Furthermore, the KPC plasmid's copy number was approximately five times higher than the chromosome. No mutations encoding efflux system AcrAB-TolC were found.

Conclusion. Resistance to MV in KP was found in isolates that predate the drug's availability. Notably, resistance occurred in the absence of MBLs and OXAs. The mechanism seems to involve outer membrane porin mutations in OmpK35 and/or OmpK36. WGS is a useful tool in identifying the mechanism of resistance especially for newer agents.

Table: Characterization of MVRKP by WGS

			Typing Enzymes Efflux			Outer Membrane Porin Variant		
Strain	Date	MV MIC (mg/mL)	MLST	β-Lactamase	Multidrug Transporters and Regulators	OmpK35	OmpK36	Notes
1	2-1-12	16	ST258	KPC-2 & SHV-160	Emr, mexk, oqxAB, smeD	FS 121insG		S to CZA and TGC
2	4-3-12	16	ST258	KPC-2 & SHV-160	Emr, mexk, oqxAB, smeD	FS 121insG		S to CZA and TGC
3	2013	**16**	**ST258**	KPC-2 & SHV-160	Emr, mexk, oqxAB, smeD	FS 121insG		S to CZA and TGC
4	2013	4	ST258	KPC-2 & SHV-160	Emr, mexk, oqxAB, smeD	FS 121insG		S to CZA and TGC
5	2017	32	ST258	KPC-2, SHV-11, SHV-12	N/A	STOP aa89	134-135 GD ins	CZA MIC 8



Abbreviations: KP: *Klebsiella pneumoniae*; FEP: Cefepime; VAN: Vancomycin; TZP: Piperacillin/Tazobactam; SAM: Ampicillin/Sulbactam; Gen: Gentamicin

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701. Rapid Detection of Antimicrobial Resistance Determinants with the BioFire® System

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