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Concordance Between Antibiotic Resistance Genes by Multiplex Polymerase Chain Reaction and Antibiotic Susceptibility by Pooled Antibiotic Sensitivity Testing in Symptomatic Patients with Urinary Tract Infection

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detect 24 different bacterial species in the urine specimens. A cutoff of 10 detections was used to distinguish a bacterial consortium from a random association of bacteria in a polymicrobial specimen. Summary statistics and Kruskal-Wallis test were used to compare the mean numbers of clinical findings across different number of bacterial species within a bacterial consortium.

Results: Bacteria were detected in 69% (1710/2493) of patients. Among these, monomicrobial infections were found in 40% (683/1710), and polymicrobial infections were found in 60% (1027/1710). Among the polymicrobial infections, consortia were identified in 433 specimens. Consortia polymicrobial urine specimens had more clinical findings than monomicrobial specimens, 2.84 vs. 2.66, respectively (p=0.022). The more bacteria detected within a consortium, the more clinical findings were reported (p=0.026), **Table 1.** Among consortia polymicrobial specimens, specimens containing Gram-negative bacteria were associated with a higher number of clinical findings compared to specimens without Gram-negative bacteria, 3.21 vs. 2.52, respectively (p<0.0001).

Conclusion: Consortia polymicrobial UTIs were associated with more clinical findings than monomicrobial infections. The number of clinical findings also increased with increasing numbers of bacteria within a consortium. The identification of a consortium within polymicrobial UTIs and the correlation of clinical findings may be important for understanding the treatment of polymicrobial UTIs.

Table 1. Mean number of clinical findings by number of bacteria detected

Bacterial content of urine specimen	Number of specimens	Mean number of clinic findings (standard deviation, range)
Monomicrobial	683	2.66 (1.25, 0-6)
Consortia polymicrobial	433	2.84 (1.25, 0-6)
2 bacteria within consortia	271	2.77 (128, 0-6)
3 bacteria within consortia	144	2.85 (1.15, 1-6)
4 bacteria within consortia	18	3.72 (1.41, 2-6)

Funding: Pathnostics

#BS24 | CONCORDANCE BETWEEN ANTIBIOTIC RESISTANCE GENES BY MULTIPLEX POLYMERASE CHAIN REACTION AND ANTIBIOTIC SUSCEPTIBILITY BY POOLED ANTIBIOTIC SENSITIVITY TESTING IN SYMPTOMATIC PATIENTS WITH URINARY TRACT INFECTION

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Presented By: Annah Vollstedt, MD

Introduction: Studies have shown many genes influence antibiotic resistance, and the relationship between genotypic and phenotypic antibiotic resistance is unclear. We sought to analyze the concordance between the presence of antibiotic resistance (ABR) genes and antibiotic susceptibility results in urine samples collected from symptomatic UTI patients.

Methods: Urine samples were collected from patients presenting with possible UTI to 37 geographically disparate Urology clinics from July 2018 to February 2019. Multiplex polymerase chain reaction (M-PCR) was used to test for 33 different ABR genes. Samples in which at least one organism was identified at a quantity of ≥104 cells per mL, Pooled Antibiotic Susceptibility Testing (P-AST), which involves simultaneously growing all detected bacteria together in the presence of antibiotics and then measuring susceptibility, was performed against 14 different antibiotics. The concordance rate between the ABR genes and the P-AST results was generated for the overall group. The concordance rates for each antibiotic between monomicrobial and polymicrobial infection were compared using chi-square test.

Results: Among the 2,512 patients, bacteria were detected in 1,579. ABR gene genotyping and P-AST analysis was performed for 1,155. ABR genes were detected in 36.3% (419/1155) of specimens. Overall, the presence or absence of ABR genes was 60% concordant with antibiotic susceptibility patterns. Two circumstances accounted for the concordance: ABR gene not present by M-PCR/antibiotic sensitive by P-AST (48.4%) and ABR gene present/antibiotic resistant (11.5%). In the 40% non-concordant cases, 25% were ABR gene not present/antibiotic resistant and 15% were ABR gene present/antibiotic sensitive, **Table 1.** Most antibiotics were associated with similar concordance rates for monomicrobial and polymicrobial infections. However, the concordance rates were significantly lower for polymicrobial for three antibiotics: vancomycin, meropenem,

and piperacillin/tazobactam, with absolute differences of 9.3% (p value=0.002), 13.1% (p value<0.0001), and 19.0% (p value = 0.019), respectively.

Conclusion: The concordance rate of the ABR genes as identified by M-PCR and phenotypic resistance as detected by P-AST was 60%. Thus, in 40% of samples, the reliance on the M-PCR antibiotic resistance gene report without the phenotypic data reported by P-AST data may lead to inappropriate treatment.

Table 1. Overall Concordance between Presence of ABR genes by M-PCR and the Antibiotic Susceptibility by P-AST Testing

CONCORDANCE		DISCORDANCE	
ABR detected + Bacteria Resistant based on P-AST	ABR NOT detected + Bacteria Susceptible based on P-AST	ABR detected but Bacteria Susceptible based on P-AST	ABR NOT detected but Bacteria Resistant based on P-AST
11.5%	48.4%	15%	25%
60%		40%	

Funding: Pathnostics

#BS25 | ROLE OF KLEBSIELLA PNEUMONIAE TYPE 3 FIMBRIAE IN ASSOCIATION AND INVASION OF HUMAN BLADDER EPITHELIAL CELLS

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Presented By: Sundharamani Venkitapathi

Introduction: Recurrent urinary tract infection (rUTI), defined as two UTI episodes within 6 months or 3 within 12 months, poses a major health issue. UPEC accounts for 65-75% of rUTIs and is followed in prevalence by the genus *Klebsiella* (15%-17%). *K. pneumoniae* is one of the most common human pathogens implicated in lung, urinary tract and surgical site infections (1) and are gaining importance due to their antibiotic resistance. Unlike UPEC *UTI89* which expresses type 1 (*fim*) fimbriae, *K. pneumoniae* produces both type 1 (*fim)* and type 3 (*mrk*) fimbriae that play important roles in host cell attachment and invasion. Type 1 fimbriae are unique from type 3 fimbriae in that their association with host cells is through mannosylated host receptors and can therefore be inhibited by mannose, while type 3 fimbriae binding remains unaffected (2, 3). Uropathogenic *K. pneumoniae* expressing type 3 fimbriae may therefore be unaffected by therapies targeting type 1 fimbriae-mediated adhesion. The goal of this study was to analyze the contribution of type 3 fimbriae of uropathogenic *K. pneumoniae* in human urothelial cell attachment and invasion.

Methods: Ten uropathogenic *K. pneumoniae* strains were isolated from urine of postmenopausal women with active, symptomatic rUTI and screened for mannose-insensitive cell association and invasion. *In vitro* cell association and invasion assays were performed using the urinary bladder epithelial 5637 cell line as previously described. Mannose-insensitive isolates PF9-uk1 and PF25-uk1 were selected for gene deletion experiments. Type 1 and type 3 fimbriae knockout strains were generated by markerless deletion of *fimA* or *mrkA* gene, respectively using the Lambda Red system. Gene complementation was performed by L-arabinose-induced ectopic expression of the *fim* and *mrk* operons (pBAD).

Results: Adhesion and invasion assays using type 3 fimbriae knockout strains showed a significant decrease in attachment and invasion of Δ*mrkA* isogenic mutants in comparison to wildtype and Δ*fimA* strains. Also, cell association of Δ*mrkA* isogenic mutants was rescued by expression of the *mrkABCDF* gene cluster.

Conclusion: Type 3 fimbriae are necessary and sufficient for uropathogenic *K. pneumoniae* mediated attachment and invasion of host urothelial cells *in vitro*.

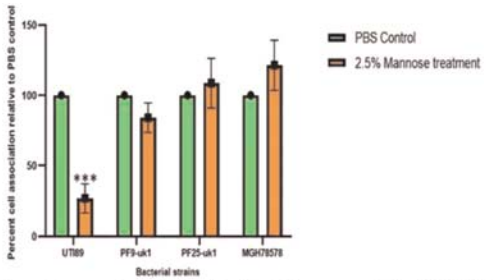

Figure. Mannose insensitive adhesion of *K. pneumoniae* to 5637 bladder epithelial cells

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#BS26 | URINARY CYTOKINES ASSOCIATED WITH RECURRENT URINARY TRACT INFECTION IN POSTMENOPAUSAL WOMEN

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