Prevalence of *bla*$_{SHV}$, *bla*$_{TEM}$ and *bla*$_{CTX-M}$ antibiotic resistance genes in selected bacterial pathogens from the Pretoria Academic Hospital

Veldsman C, Kock MM, Makgotlho EP, Hoosen AA, Dove MG and Ehlers MM

Department of Medical Microbiology, Faculty of Health Sciences, University of Pretoria/NHLS

INTRODUCTION

- Extended-spectrum β-lactamases (ESBLs) were first identified in the early 1980s in Germany and has since been identified worldwide
- Bacteria producing β-lactamases are increasingly reported as the cause of severe infections in intensive care- and surgical units
- Mortality rates varying from 42% to 100% have been reported in patients infected by ESBL-producing bacteria
- Most ESBL-producing bacteria can be divided into three groups: TEM, SHV and CTX-M types
- Gram-negative β-lactamases are often mediated by *bla*$_{SHV}$, *bla*$_{CTX-M}$ and *bla*$_{TEM}$
- Disk diffusion interpretive criteria is used for the detection of bacteria producing ESBLs
- The choice of drugs for the treatment is limited to carbapenems for example imipenem, fluoroquinolones and aminoglycosides

AIM

The aim of this study was to determine the prevalence of ESBLs in selected Gram-negative clinical bacterial isolates

MATERIALS AND METHODS

- Fifty six (56) selected clinical bacterial isolates were obtained from clinical specimens sent from an academic hospital for analysis
- The prevalence of *bla*$_{SHV}$, *bla*$_{CTX-M}$ and *bla*$_{TEM}$ genes were determined in the following isolates (Table 1)
- Identification and antibiotic resistance was determined using the Vitek System (Vitek 2, bioMérieux, France)

Table 1: Clinical bacterial isolates obtained from Pretoria Academic Hospital (n=56)

Bacteria	Amount of isolates (n=56)
Klebsiella pneumoniae	33
Escherichia coli	14
Enterobacter cloacae	4
Morganella morganii ssp morganii	3
Citrobacter freundii	1
Proteus penneri	1

- The MagNaPure LC Compact (Roche Applied Science, Germany) was used for the automated extraction of total DNA according to the manufacturer's protocol
- The Multiplex PCR assay was performed using the Qiagen Multiplex PCR Kit and a PX2 Thermal cycler (Thermo Electron Corporation, MA distributed by Scientific Group, SA) for the amplification of the DNA templates

RESULTS AND DISCUSSION

- Multiplex PCR successfully detected the presence of *bla*$_{SHV}$ (747 bp), *bla*$_{CTX-M}$ (593 bp) and *bla*$_{TEM}$ (445 bp) genes (Figure 1)
- Multiple *bla*-genes were detected in 63% of all selected bacterial pathogens while 30% of the isolates only had a single *bla*-gene (Figure 2)
- In the remaining isolates no ESBL genes were detected

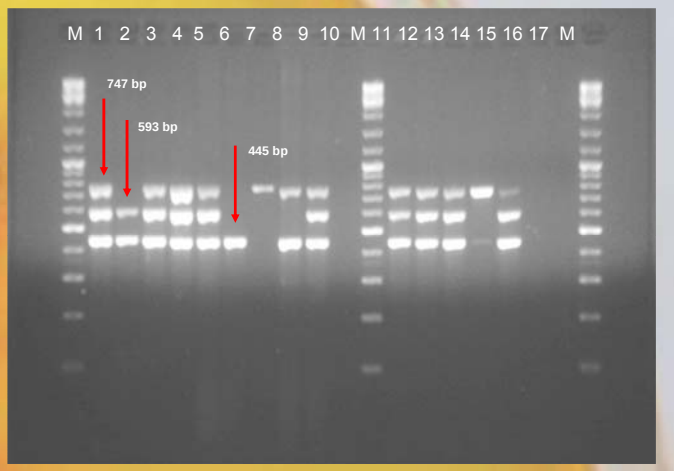

Figure 1 **Gel electrophoresis displaying the amplified *bla*$_{SHV}$ (747 bp), *bla*$_{CTX-M}$ (593 bp) and *bla*$_{TEM}$ (445 bp) genes. Lanes 1, 3-5, 9, 11-13 and 15 are *bla*$_{SHV}$, *bla*$_{TEM}$ and *bla*$_{CTX-M}$ positive. Lane 2 is positive for *bla*$_{TEM}$ and *bla*$_{CTX-M}$. Lane 8-14 are positive for *bla*$_{SHV}$ and *bla*$_{TEM}$. Lane 7 is positive for the *bla*$_{SHV}$ gene and lane 6 is only positive for the *bla*$_{TEM}$ gene. Lane 16 is the only isolate which was negative for any one of the three genes. A negative control was included in lane 17. Lanes M represent the molecular weight marker (50 bp DNA ladder, Promega, Madison, USA)**

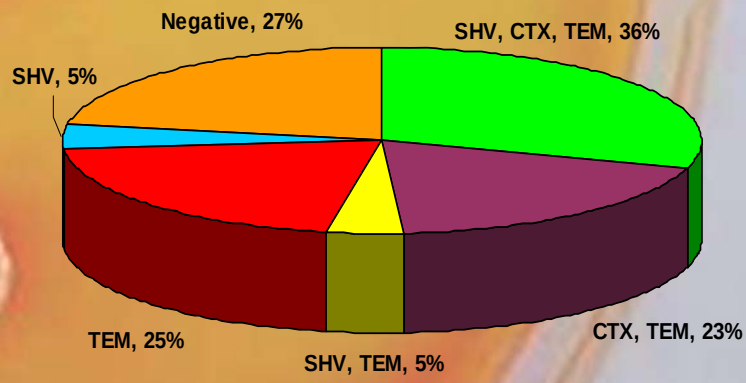

Figure 2 **Summary of final multiplex PCR results for each of the three genes: *bla*$_{SHV}$, *bla*$_{CTX-M}$ and *bla*$_{TEM}$ detected in the selected clinical bacterial isolates**

CONCLUSION

Knowledge of the presence and prevalence of ESBL genes might assist in improved monitoring of these bacterial pathogens in hospital settings and to advice clinicians on possible treatment regimens

REFERENCES

- Colodner R (2005) Extended-spectrum β-lactamases: a challenge for clinical microbiologists and infection control specialists. American Journal of Infection Control 33: 104-107.
- Kim S, Hu J, Gautom R, Kim J, Lee B and Boyle DS (2007) CTX-M extended-spectrum β-lactamases, Washington State. *Emerging Infectious Diseases* **13**: 513-514.