In vitro activity of temocillin, piperacillin-tazobactam, and meropenem against third generation cephalosporin-resistant *Enterobacterales*

L. Stenzel[1], S. Schröder[1], A. Fischer[1], T. Hattwig[1], K. Seefeldt[1], A. Gehnen[1], K. Grimm[2], R. Geisel[1], B. R. Thoma[1]

Medizinische Laboratorien DÜSSELDORF

[1] Medizinische Laboratorien Düsseldorf, Düsseldorf, Germany
[2] EUMEDICA SA, Manage, Belgium

Introduction

Infections caused by third generation cephalosporin-resistant *Enterobacterales* (3GCREB) pose a serious threat to public health[1]. Previous studies have reported inferiority of piperacillin-tazobactam (PTZ) when treating infections due to 3GCREB leading to an overuse of reserve antibiotics like carbapenems[2]. Alternatively, the beta-lactam antibiotic temocillin (TEM) features resistance against most beta-lactamases, including AmpC and extended spectrum beta-lactamases (ESBL) typically found in 3GCREB[3]. In this study, we performed comparative minimal inhibitory concentration (MIC) determination of TEM, PTZ, and meropenem (MER) in *Enterobacterales*, collected from urological wards and in the majority phenotypically harboring AmpC and/or ESBL. Further, we compared MIC values of PTZ and MER with different MIC methods.

Methods

205 isolates, classified as 3GCREB, were collected from urine (n=197) and blood culture samples (n=8) between September 2020 and January 2021 (*Escherichia* (*E.*) *coli* n=116, *Klebsiella* (*K.*) spp. n=40 [*K. pneumoniae* n=30, *K. aerogenes* n=8, *K. oxytoca* n=2], *Proteus* (*P.*) *mirabilis* n=7, *Providencia* (*P.*) *rettgeri* n=1, *Enterobacter* (*E.*) spp. n=20 [*E. cloacae* n=13, *E. cloacae complex* n=7], *Citrobacter* (*C.*) spp. n=5 [*C. freundii* n=4, *C. braakii* n=1], *Serratia* (*S.*) *marcescens* n=2, *Morganella* (*M.*) *morganii* n=14) and MICs determined by gradient diffusion method (GDM) [bioMérieux, France; for TEM, PTZ, MER], broth microdilution (BM) [MERLIN (MICRONAUT–S MDR MRGN-Sreening), Germany; for PTZ, MER], and VITEK® 2 [bioMérieux (AST-371; AST-223), France; for PTZ, MER], respectively. MICs of TEM, PTZ, and MER were assessed using EUCAST (version 11.0) breakpoints. The isolates were phenotypically differentiated for AmpC and/or ESBL using disc diffusion method (zone diameter cut-off at ≥4 mm)[4].

Results

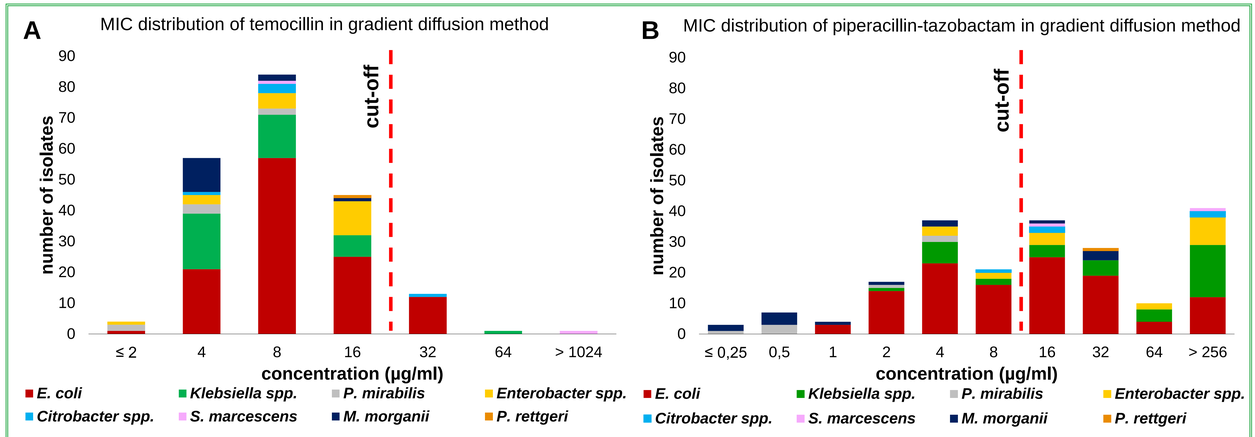


Figure 1 MIC distributions of 205 isolates of TEM and PTZ in GDM. (A) MIC distribution of TEM divided into species and genera. Total MIC_{50}=8 µg/ml and MIC_{90}=16 µg/ml. **(B)** MIC distribution of PTZ divided into species and genera. Total MIC_{50}=16 µg/ml and MIC_{90}>256 µg/ml. For illustrative reasons breakpoints given by EUCAST are applied to all strains.

Results

All isolates were sensitive to MER and the MIC distribution pattern was equal in either MIC method. MIC distribution (GDM) of all strains showed a shift towards higher values of PTZ (MIC_{50}=16 µg/ml, MIC_{90}>256 µg/ml) as compared to TEM (MIC_{50}=8 µg/ml, MIC_{90}=16 µg/ml) (Table 1). In comparison, 89.66% (104/116) *E. coli*, 96.67% (29/30) *K. pneumoniae*, 100% (2/2) *K. oxytoca,* and 100% (7/7) *P. mirabilis* were susceptible to TEM and 48.28% (56/116) *E. coli*, 33.33% (10/30) *K. pneumoniae*, 0% (0/2) *K. oxytoca,* and 100% (7/7) *P. mirabilis* to PTZ in GDM, respectively (Figure 1). For PTZ, MICs derived from GDM (MIC_{50}=16 µg/ml, MIC_{90}>256 µg/ml) were higher compared to VITEK® 2 (MIC_{50}=8 µg/ml, MIC_{90}>128 µg/ml) and BM (MIC_{50}≤4 µg/ml, MIC_{90}>64 µg/ml). Qualitatively, 43.42%, 62.44%, and 68.29% of all isolates were sensitive to PTZ determined by GDM, VITEK® 2, and BM, respectively (Table 1). Phenotypically, 132 ESBL, 38 AmpC, 16 ESBL/AmpC, and 19 negative isolates were determined.

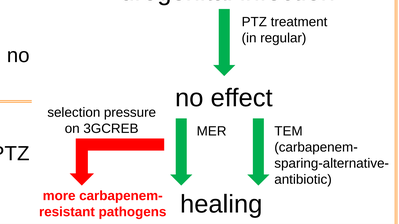
Table 1 MIC distributions of PTZ, TEM, and MER in 205 isolates derived by three methods (GDM, VITEK® 2 and BM). For illustrative reasons breakpoints given by EUCAST are applied to all strains. **I** = breakpoint; green = **≤**; orange = **>**; **N/A** = not applicable and **S** = susceptibility.

Antibiotics (µg/ml)	0.125	0.25	0.5	1	2	4	8	16	32	64	128	256	1024	MIC_{50}	MIC_{90}	S (%)
PTZ (GDM)	-	3	7	4	17	36	22	37	28	11	0	40	-	**16**	**> 256**	**43.42**
PTZ (VITEK® 2)	-	-	-	-	-	91	37	18	6	14	39	-	-	**8**	**> 128**	**62.44**
PTZ (BM)	-	-	-	-	-	125	15	13	16	36	-	-	-	**≤ 4**	**> 64**	**68.29**
TEM (GDM)	-	-	-	-	4	57	84	45	13	1	0	0	1	**8**	**16**	**92.68**
TEM (BM)	-	-	-	-	-	-	-	-	203	2	-	-	-	**N/A**	**N/A**	**N/A**
MER (GDM)	192	13	0	0	0	0	0	0	0	-	-	-	-	**≤ 0.125**	**≤ 0.125**	**100.00**
MER (VITEK® 2)	-	190	15	0	0	0	0	0	-	-	-	-	-	**≤ 0.25**	**≤ 0.25**	**100.00**
MER (BM)	205	0	0	0	0	0	0	0	0	0	0	-	-	**≤ 0.125**	**≤ 0.125**	**100.00**

Conclusion

- In the clinical setting PTZ is always escalated with MER and our data highlight, that TEM could be a better carbapenem-sparing-alternative-antibiotic for the treatment of infections due to 3GCREB than PTZ [e.g., susceptibility TEM (92.68%) vs. PTZ (43.42%) in GDM];
- PTZ, however, has antipseudomonal activity;
- Advantageously, TEM renders a minimal risk of *Clostridium difficile* infections and has no significant inoculum effect[5].

- VITEK® 2 shows near concordant results with the gold-standard-method (BM) for PTZ;
- In routine laboratories GDM is widely used as a MIC-confirmatory-method: in the case of PTZ our results show, that confirmation should be done with the ”gold-standard-method” (BM);
- For comparative reasons MICs for TEM should also be determined by BM and VITEK® 2.



Acknowledgements

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References

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Correspondence

Lisa Stenzel
Phone: +49-211-4978190
Mail: stenzel@labor-duesseldorf.de