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# Comparison of four methods for the determination of tigecycline susceptibility in multi-resistant *E. coli* and *K. pneumoniae*

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## Introduction

- The incidence of infections with multidrug resistant (MDR) Enterobacteriaceae has risen considerably in recent years, associated mostly with ESBL production.
- Tigecycline is a glycylcycline antibiotic that shows good activity against most MDR Enterobacteriaceae and is therefore frequently used in infections with ESBL or carbapenemase producers [1].
- Conflicting data exist on the validity of susceptibility data generated by different susceptibility testing methods [2, 3].
- We therefore compared three different commercial methods to the current gold standard using a set of molecularly characterized *E. coli* and *K. pneumoniae* isolates from invasive infections.

## Objectives

- To assess the validity of three different commercial methods for susceptibility testing of tigecycline.

## Methods

- Isolates: 89 molecularly characterized *E. coli* (N=66) and *K. pneumoniae* (N=23) isolates showing resistance to 3rd generation cephalosporins (92.4% and 95.7% ESBL).
- All isolates were obtained from blood stream infections.
- Tigecycline MICs were determined by Vitek N214 card, Etest (both bioMérieux, Marcy-l'Étoile, France) and MIC Test Strip (Liofilchem, Roseto Degli Abruzzi, Italy), and compared to the current gold standard (broth microdilution testing).
- The rate of essential agreement (MIC within ± one 2-fold dilution compared with gold standard) and categorical agreement using FDA and EUCAST breakpoints was determined for the three commercial systems. For categorical agreement, the following definitions were used:
  - minor discrepancy: result of gold standard intermediate (I), commercial test resistant (R) or susceptible (S)
  - major discrepancy: result of gold standard S, commercial test R
  - very major discrepancy: result of gold standard R, commercial test S

## Results

Tigecycline MIC50/MIC90s determined by the reference method were 0.5/1.0 mg/L for *E. coli* and 1.0/2.0 mg/L for *K. pneumoniae* (Table 1). In *E. coli*, the Vitek2 N214 card reached the highest essential agreement, in *K. pneumoniae* the MIC Test Strip. The categorical agreement for all methods depends on the interpretation criteria used (FDA or EUCAST); it was better in *E. coli* and when FDA breakpoints were applied. In *K. pneumoniae*, MIC Test Strip performed better than the N214 card and Etest, which both showed less than 50% categorical agreement when EUCAST interpretation criteria were used. No very major errors were observed for any of the commercial systems.

***Table 1: Results of the susceptibility tests using four different methods***

| *E. coli* | microdilut. | Etest | MIC Strip | Vitek N214 |
|---|---|---|---|---|
| **MIC 50 (mg/L)** | 0.5 | 0.75 | 0.38 | ≤ 0.5 |
| **MIC 90 (mg/L)** | 1 | 1 | 0.5 | ≤ 0.5 |
| **Essential agreement (%)** | | 74.2 | 87.9 | 100 |
| **Categorical agreement FDA (%)** | | 100 | 100 | 100 |
| Minor discrepancy (%) | | 0 | 0 | 0 |
| Major discrepancy (%) | | 0 | 0 | 0 |
| **Categorical agreement EUCAST(%)** | | 95.5 | 100 | 100 |
| Minor discrepancy (%) | | 4.5 | 0 | 0 |
| Major discrepancy (%) | | 0 | 0 | 0 |

| *K. pneumoniae* | microdilut. | Etest | MIC Strip | Vitek N214 |
|---|---|---|---|---|
| **MIC 50 (mg/L)** | 1 | 2 | 1 | 2 |
| **MIC 90 (mg/L)** | 2 | 4 | 2 | 4 |
| **Essential agreement (%)** | | 78.3 | 82.6 | 60.9 |
| **Categorical agreement FDA (%)** | | 87.0 | 95.7 | 69.6 |
| Minor discrepancy (%) | | 13.0 | 4.3 | 30.4 |
| Major discrepancy (%) | | 0 | 0 | 0 |
| **Categorical agreement EUCAST(%)** | | 47.8 | 60.9 | 34.8 |
| Minor discrepancy (%) | | 47.8 | 39.1 | 43.5 |
| Major discrepancy (%) | | 4.3 | 0 | 21.7 |

## Conclusion

Tigecycline MICs can be reliably determined in *E. coli* using Etest, N214 card and MIC Test Strip; all methods showed categorical agreements of 95.5-100%. In *K. pneumoniae*, tigecycline MICs were more widely dispersed and the rates of agreement were lower for all methods, especially when EUCAST breakpoints were used. Among the antibiotic gradient testing methods, MIC Test Strip performed better than Etest. The Vitek2 N214 card showed the highest number of minor (43.5%) and major discrepancies (21.7%), which most of the time resulted in an interpretation that was falsely resistant. Tigecycline non-susceptibility in *K. pneumoniae* is therefore most likely overreported when this system is used.

## Acknowledgements

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## References

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