

# Comparison of two immunochromatographic tests for the detection of CTX-M ESBL on clinical isolates at the Belgian National Reference Centre

Pierre Bogaerts<sup>1</sup>, Cyprien Mbundu Lukukula<sup>2</sup>, Warda Bouchahrouf<sup>1</sup>, Catherine Berhin<sup>1</sup>, Isabel Montesinos<sup>1</sup>, Olivier Denis<sup>1</sup>, Jean-Marie Liesse Iyamba<sup>2</sup>, Te-Din Huang<sup>1</sup>

<sup>1</sup>National Reference Center for Antibiotic-resistant Gram-negative bacilli, CHU UCL Namur, Yvoir, Belgium. <sup>2</sup>UNIKIN - Kinshasa (Democratic Republic of Congo)

## Revised abstract

### Background

CTX-M is the most prevalent ESBL in the world causing nosocomial and community acquired enterobacterial infections. Rapid detection is important for the choice of antibiotherapy and for the potential initiation of infection control measures in high-risk acute care units. We compared the performance of two different immunochromatographic tests: NG-Test® CTX-M MULTI (NG Biotech) and Coris BioConcept RESIST CTX-M (Coris BioConcept) on a collection of clinical isolates.

### Methods

167 Enterobacterales strains (82 *Escherichia coli*, 58 *Klebsiella pneumoniae* and 27 other species) characterised in our national reference centre were tested for the assays comparison. Isolates were previously identified to the species by MALDI-TOF MS (Bruker), tested for susceptibility phenotype by disk diffusion and for the presence of CTX-M of group -1, -2 and -9 by ISO15189-certified CR/sequencing. The collection included 99 strains expressing CTX-M-Group1 (G1), 11 CTX-M-G2 and 17 CTX-M-G9, and 1 strain harbouring CTXMG1 and CTX-M-G9, while 40 isolates harboured no CTX-M. NG Biotech is designed to detect all CTX-M groups on a single band and Coris BioConcept detects and identifies G1 and G9 (2 different bands) but not CTX-M-G2. Both assays were performed according to manufacturer's recommendations (15 min. to results).

### Results

Both assays present 100% sensitivity and specificity for their targeted groups. For the strains expressing CTX-M-G1 and G9, as expected two bands are clearly observed with Coris BioConcept and 1 band with NG Biotest. For all CTX-M-G2 (all CTX-M-2), a clear positive band was obtained by NG Biotest, while no positive signal band was observed with Coris BioConcept. Both tests detected CTX-M-1, 3, 15, 55, 135 (G1) and CTX-M-9, 14, 27 and 65 (G9).

### Conclusions

Both NG-Test® CTX-M MULTI (NG Biotech) and RESIST CTX-M (Coris BioConcept) performed well for the detection of CTX-M. Coris BioConcept presents the advantage to differentiate between CTX-M-G1 and G9 but does not target CTX-M-G2. NG Biotest targets all CTX-M groups but on a single band. Intended use and local epidemiology should guide the choice between these two tests.

## Introduction and Objectives

- Rapid detection of multi-drug resistant bacteria is essential for choosing antibiotherapy and the potential initiation of infection control measures in high-risk acute care units.
- Extended-spectrum  $\beta$ -lactamases (ESBLs) are the most frequent resistance mechanism in *Enterobacterales*, and CTX-Ms represent the most prevalent ESBLs worldwide, causing nosocomial and community-acquired enterobacterial infections.
- The immunochromatographic tests have been developed and commercialized to reduce the time to results of antimicrobial susceptibility approaches from 24 hours to a few hours.
- In this study, we compared the performance of two different immunochromatographic assays for rapid detection of CTX-M Enterobacterales strains on a collection of clinical isolates: NG-Test® CTX-M MULTI (NG Biotech) and Coris BioConcept RESIST CTX-M (Coris BioConcept).

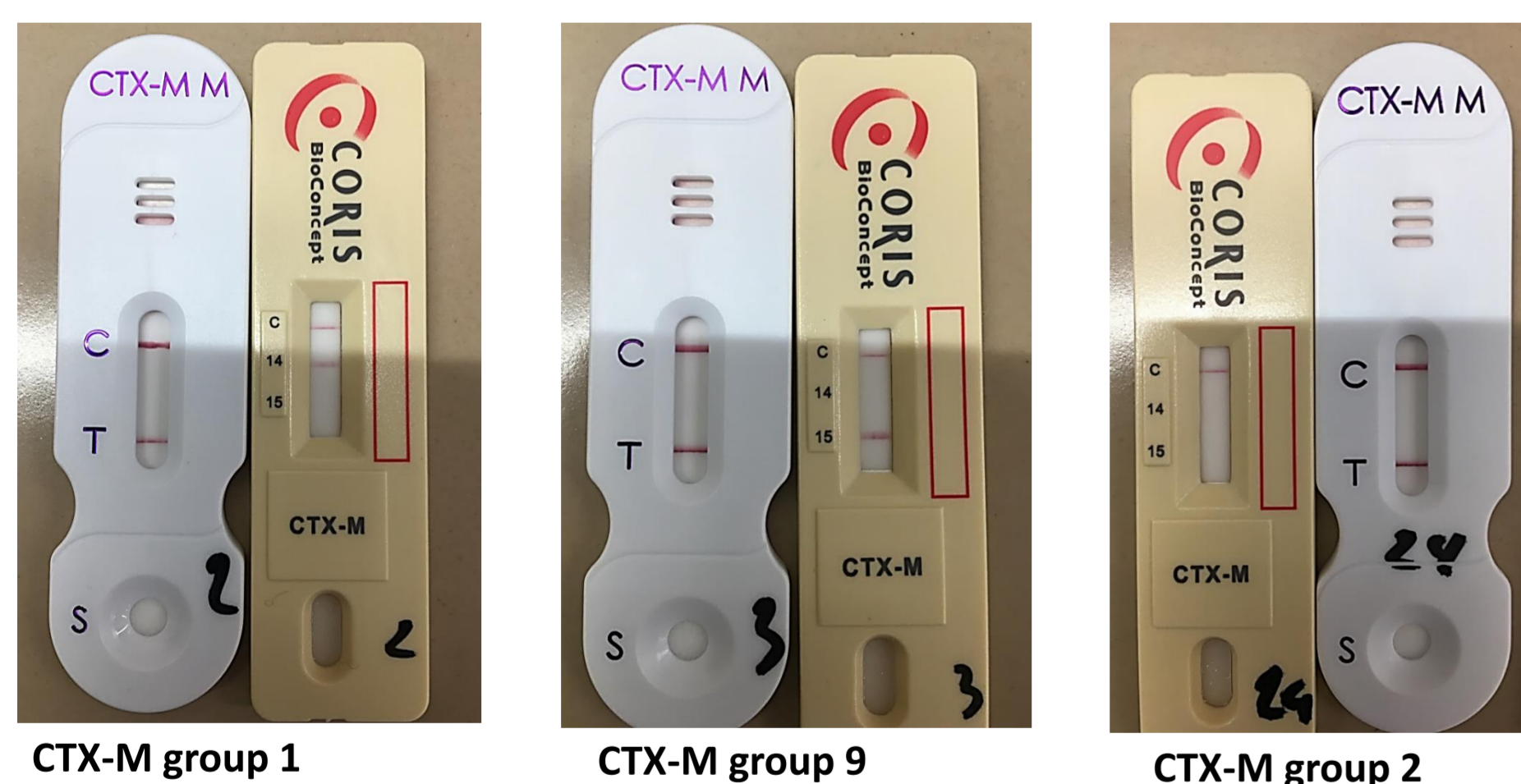
## Methods

- A total of 167 Enterobacterales strains (82 *Escherichia coli*, 58 *Klebsiella pneumoniae* and 27 other species) were tested to compare NG Biotech and Coris BioConcept. Eighty-eight of the strains included in this study are from UNIKIN-Kinshasa (Democratic Republic of Congo).
- All isolates were previously characterized in the Belgian National Reference Center using MALDI-TOF MS (Bruker) for identification at the species level, by disk diffusion for susceptibility phenotype and by ISO15189-certified CR/sequencing for the presence of CTX-M of group -1, -2 and -9:
  - 127 strains harbouring CTX-M group 1 (99), group 2 (11) and group 9 (17). (1 strain CTXMG1 and CTX-M-G9)
  - 40 strains harbouring no CTX-M
- The Coris BioConcept is an immunochromatographic test line (ICT) allowing the identification of CTX-M of groups 1 and 9. Coris BioConcept does not target CTX-M-G2 or CTX-M G8/25.
- NG Biotech is also an immunochromatographic assay for the rapid detection of CTX-M Groups 1, 2, 8, 9 and 25 on a single band
- Both assays were performed according to the manufacturer's recommendations. Results for both tests were available in less than 15 minutes from a bacterial colony. Two independent readers performed reading and interpretation.

## Results

- Coris BioConcept and NG Biotest present 100% sensitivity and specificity for their targeted groups.
- For the strains expressing CTX-M-G1 and G9, two bands are clearly observed with Coris BioConcept and one band with NG Biotest as expected. Both tests detected CTX-M-1, 3, 15, 55, 135 (G1) and CTX-M-9, 14, 27 and 65 (G9).
- For all CTX-M-G2, a clear positive band was obtained by NG Biotest, while no positive signal band was observed with Coris BioConcept.
- The signals were easy to detect by eye and presented no difficulty of interpretation for both tests

Figure 1: NG Biotest and Coris BioConcept tests results



	n of strains	Coris BioConcept <sup>#</sup>	NG Biotech
<i>E. coli</i>	44	100%	100%
<i>K. pneumoniae</i>	43	100%	100%
<i>K. oxytoca</i>	2	100%	100%
<i>E. cloacae</i>	3	100%	100%
<i>C. freundii</i>	2	100%	100%
<i>M. morgani</i>	2	100%	100%
<i>S. marcescens</i>	2	100%	100%
<i>Salmonella spp.</i>	1	100%	100%

<sup>#</sup> The strains tested did not show CTX-M group 9 band

	n of strains	Coris BioConcept <sup>#</sup>	NG Biotech
<i>E. coli</i>	7	100%	100%
<i>K. pneumoniae</i>	3	100%	100%
<i>K. oxytoca</i>	1	100%	100%
<i>E. cloacae</i>	3	100%	100%
<i>E. kobei</i>	1	100%	100%
<i>E. aerogenes</i>	1	100%	100%
<i>C. freundii</i>	1	100%	100%

<sup>#</sup> The strains tested did not show CTX-M group 1 band

	n of strains	Coris BioConcept <sup>#</sup>	NG Biotech
<i>E. coli</i>	6	0%	100%
<i>K. pneumoniae</i>	1	0%	100%
<i>K. oxytoca</i>	1	0%	100%
<i>P. mirabilis</i>	3	0%	100%

	n of strains	Coris BioConcept <sup>#</sup>	NG Biotech
<i>E. coli</i>	25	0%	0%
<i>K. pneumoniae</i>	11	0%	0%
<i>K. oxytoca</i>	2	0%	0%
<i>E. cloacae</i>	1	0%	0%
<i>E. aerogenes</i>	1	0%	0%

## Conclusions

- The intrinsic performances of CTX-M MULTI (NG Biotech) and RESIST CTX-M (Coris BioConcept) are excellent for the detection of CTX-M.
- Coris BioConcept presents the advantage of differentiating between CTX-M-G1 and G9, providing some information at the epidemiological level. The clinical impact of this differentiation remains to be confirmed.
- Coris BioConcept does not target CTX-M-G2, a disadvantage in specific epidemiology where CTX-M G2 is more prevalent than in Belgium, where CTX-M G1 and G9 are far more prevalent.
- Intended use and local epidemiology should guide the choice between these tests.

## References

- Gonzalez C, Moguet C, Chalin A, Oueslati S, Dortet L, Simon S, Volland H, Naas T. Comparison of Three Expanded-Spectrum Cephalosporin Hydrolysis Assays and the NG-Test CTX-M Multi Assay That Detects All CTX-M-Like Enzymes. *Diagnostics* (Basel). 2022 Jan 14;12(1):197. doi: 10.3390/diagnostics12010197. PMID: 35054364; PMCID: PMC8775164.
- Boattini M, Bianco G, Comini S, Iannaccone M, Casale R, Cavallo R, Nordmann P, Costa C. Direct detection of extended-spectrum- $\beta$ -lactamase-producers in Enterobacterales from blood cultures: a comparative analysis. *Eur J Clin Microbiol Infect Dis*. 2022 Mar;41(3):407-413. doi: 10.1007/s10096-021-04385-1. Epub 2021 Nov 25. PMID: 34822029; PMCID: PMC8614078.

## Funding and Acknowledgements

- We acknowledge colleagues from UNIKIN-Kinshasa for collaborating on this study
- The national reference center is supported by the Belgian Ministry of Social Affairs through a fund within the Health Insurance System.

