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Evaluation of screening algorithms to detect rectal colonization with carbapenemase-producing Enterobacterales in a resource-limited setting

Thi Anh Mai Pham [1,2,3,4]†, Tung Xuan Nguyen[5], Troung Nhat My [3]‡, Lan Thi Le[5]‡, Huyen Thi Vu[5]‡, Ngoc Thi Bich Hoang [5], Dien M. Tran[6], Linh Viet Nguyen [7], Phuc D. Pham [7], Dennis Nurjadi [3,4], Flavie Goutard [8], Thirumalaisamy P. Velavan [2,3,9], Van Anh Thi Dinh[10,11], Y.M. Gildas Hounmanou [12], Bent Jörgensen[13,14,15], Le Huu Song [3,16], Nhung T.T. Nguyen[7,10], Etienne Loire [8], Åse Östholm [14], Lennart E. Nilsson [14], Tuyet Hanh T. Tran [7], Phuc H. Phan [6,10], Anders Dalsgaard [12], Mattias Larsson [1,13], Linus Olson [1,13,17]*§ and Håkan Hanberger [13,14]†§

[1]Department of Global Public Health, Karolinska Institutet, Stockholm, Sweden; [2]Institute of Tropical Medicine, Universitätsklinikum Tübingen, Tübingen, Germany; [3]Vietnamese German Center for Medical Research (VG-CARE), Hanoi, Vietnam; [4]Department of Infectious Diseases and Microbiology, University of Lübeck and University Hospital Schleswig-Holstein, Lübeck, Germany; [5]Department of Microbiology, Vietnam National Children's Hospital, Hanoi, Vietnam; [6]Director Board, Vietnam National Children's Hospital, Hanoi, Vietnam; [7]Hanoi University of Public Health, Hanoi, Vietnam; [8]The French Agricultural Research Centre for International Development (CIRAD), Montpellier, France; [9]Faculty of Medicine, Duy Tan University, Da Nang, Vietnam; [10]Training and Research Institute for Child Health, Vietnam National Children's Hospital, Hanoi, Vietnam; [11]Department of Infection Control, Vietnam National Children's Hospital, Hanoi, Vietnam; [12]Department of Veterinary and Animal Sciences, University of Copenhagen, Copenhagen, Denmark; [13]Training and Research Academic Collaboration (TRAC), Sweden, Vietnam; [14]Department of Biomedical and Clinical Sciences (BKV), Linköping University, Linköping, Sweden; [15]Department of Global Studies, Göteborg University, Gothenburg, Sweden; [16]Director Board, 108 Military Central Hospital, Hanoi, Vietnam; [17]Department of Women's and Children's Health, Karolinska Institutet, Tomtebodavägen 18 A, 8 fl, Stockholm 17176, Sweden

*Corresponding author. E-mail: Linus.Olson@ki.se
†Equal first author contribution.
‡Equal contribution.
§Shared last authorship.

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Objectives: To improve and rationalize the detection of carbapenemase-producing Enterobacterales (CPE) in rectal swabs in a high-prevalence and resource-constrained setting, addressing surveillance challenges typically encountered in laboratories with limited resources.

Methods: A point prevalence survey (PPS) was conducted on 15 August 2022, in a provincial children's hospital in northern Vietnam. Rectal swab samples of all admitted children were collected and plated on a selective medium for carbapenem-resistant Enterobacterales (CRE). Species identification and antimicrobial susceptibility testing (AST) were performed by MALDI-TOF, and VITEK2 XL and interpreted according to CLSI breakpoints (2022). Carbapenemases were detected by the carbapenem inactivation method (CIM) and quantitative real-time PCR (qRT-PCR).

Results: Rectal swab samples were obtained from 376 patients. Of 178 isolates growing on the CRE screening agar, 140 isolates were confirmed as Enterobacterales of which 118 (84.3%) isolates were resistant to meropenem and/or ertapenem. CIM and PCR showed that 90/118 (76.3%) were carbapenemase producers. Overall, 83/367 (22.6%) were colonized by CPE. *Klebsiella pneumoniae*, *Escherichia coli* and *Enterobacter cloacae* complex were the most common CPE detected, with NDM as the predominant carbapenemase (78/90; 86.7%). Phenotypic resistance to meropenem was the best predictor of CPE production (sensitivity 85.6%, specificity 100%) compared with ertapenem resistance (95.6% sensitivity, 36% specificity). CIM was 100% concordant with PCR in detecting carbapenemases.

Conclusions: These findings underscore the effectiveness of meropenem resistance as a robust indicator of the production of carbapenemases and the reliability of the CIM method to detect such carbapenemases in resource-limited settings where the performance of molecular methods is not possible.

Introduction

Carbapenem-resistant Enterobacterales (CRE) have been classified as critical priority pathogens by the WHO, signifying their substantial impact on global public health.[1] CRE infections are often associated with poor clinical outcomes, high morbidity and mortality rates, prolonged hospital stays and a high economic burden.[2,3] Within the global landscape, Southeast Asia and especially Vietnam is recognized as a prominent hotspot for CRE, highlighting the region's vulnerability to and the urgent need for addressing the challenges posed by CRE.[2,4]

The upsurge in CRE is predominantly fuelled by the rise and dissemination of carbapenemases, a specific subgroup of β-lactamases proficient in hydrolysing carbapenems. Carbapenem resistance can also be mediated by mutations in target genes, alterations in membrane permeability, or overexpression of efflux pumps.[5] Although CRE as a whole pose treatment challenges, carbapenemase-producing Enterobactererales (CPE) stand out as a more pressing concern for infection prevention and treatment.[6] This is because carbapenemase genes are predominantly carried on plasmids, facilitating their transfer between bacterial species.[7] As a result, CPE outbreaks are frequently reported.[8,9]

Screening for CPE colonization and carbapenemase characterization is invaluable in monitoring the dynamics of regional CPE spreads.[3] CPE colonization has been identified as a risk factor for acquiring infection with CPE.[10] In addition, prolonged rectal carriage may promote onward transmission in local communities as well as globally.[11] Therefore, we undertook a point prevalence survey (PPS) to evaluate the performance of a CRE selective growth medium combined with species identification and antimicrobial susceptibility testing (AST) to obtain a snapshot of CPE epidemiology in a Vietnamese paediatric hospital in the Red River Delta. This study aimed to contribute insights into the epidemiology of CPE to better understand and manage the escalating challenges posed by CPE in the region.

Methods

Study design and participants

A PPS was conducted on 15 August 2022 at a Director Board hospital in the Red River Delta in Vietnam. Paediatric inpatients of all ages who were admitted to one of the 13 departments were included in the study after informed consent.

Study setting

Thai Binh province is located in the Red River Delta region of northern Vietnam with a population of 1.9 million inhabitants, with an estimated 2500 physicians in 2021 registered for practice in the study hospital. It is subdivided into seven rural districts and one provincial capital, Thai Binh.

Data collection and bacterial culture on study site

Rectal swab samples were collected using sterile cotton swabs. Samples were cultured on selective chromogenic carba agar (CHROMagar™ mSuperCARBA™; Melab/Lavitec, Vietnam) to screen for CRE and incubated at 35 ± 2°C for 18–24 h. In the case of bacterial growth, species identification of suspected CRE and AST for ertapenem and meropenem were performed using VITEK MS (bioMérieux) and VITEK 2 XL (bioMérieux), respectively. AST results were interpreted according to the clinical breakpoints suggested by CLSI 2022. Bacterial isolates were cryopreserved until further use.

External validation and detection of carbapenemase production

Cryopreserved CRE isolates were recultured on selective medium for CRE, CHROMagar™ mSuperCARBA™ (MAST Group, Germany). Isolates growing on mSuperCARBA plates were identified by MALDI-TOF MS (Bruker Daltonics). All confirmed CRE isolates underwent carbapenemase detection by *in vitro* carbapenem inactivation assay (CIM)[12] and molecular detection of carbapenemase genes by quantitative real-time PCR (qRT-PCR) ($bla_{\text{OXA-48-like}}$, bla_{VIM}, bla_{NDM} and bla_{KPC}), using previously published and validated primers[13] (Tables S1 and S2, available as Supplementary data at *JAC-AMR* Online). The strains *Klebsiella pneumoniae* ATCC BAA 1705 and *K. pneumoniae* ATCC BAA 1706 were used as positive and negative controls for the CIM assay, respectively.

Statistical analysis

Descriptive statistics analysis and diagnostic test performance were performed using Stata18 (StataCorp, USA).

Results

Evaluation of procedures and methods to detect CPE

On 15 August 2022, 376 patients were screened for CRE rectal colonization using chromogenic CRE selective media. Of the 376 rectal swabs, 150 (39.9%) yielded positive culture results with 178 isolates (one species in each of 122 samples and two different species in each of 28 samples). VITEK MS confirmed 140/178 isolates (78.7%) as Enterobacterales, but 38 (21.3%) isolates were non-fermenters and therefore excluded from further analysis. Phenotypic AST using VITEK 2 XL confirmed that 118/140 (84.3%) were CRE, with 77/140 isolates being resistant to both ertapenem and meropenem and 41/140 (29.3%) isolates resistant to ertapenem only. Twenty-two (15.7%) suspected CRE isolates were resistant neither to meropenem nor ertapenem (Table 1).

All 178 isolates recovered on the CRE selective medium were further characterized in an external laboratory. All 178 isolates grew on the CHROMagar™ mSuperCARBA™ plates, revealing a minor discordant result (140 versus 141 Enterobacterales). One isolate previously identified as *Aeromonas sobria* was identified as *Citrobacter freundii* in the validation but was phenotypically susceptible to meropenem and was therefore not processed further. The carbapenemase activity (CIM assay) test and qRT-PCR corroborated and showed that 90/140 isolates (63.8%) were carbapenemase producers. Carbapenemase genes were detected in 90/140 (63.8%), with 89 (98.9%) isolates harbouring a single

Table 1. Detection of carbapenemase-producing Enterobacterales in a point prevalence study, Vietnam 2022; data presented are the number of isolates obtained from 376 patient samples

	CPE[a]		Ertapenem resistance[b]		Meropenem resistance[c]		Meropenem inactivation (CIM)		Carbapenemase gene detection[d]	
Species	Yes, *n* (%)	No, *n* (%)	Yes, *n* (%)	No, *n* (%)	Yes, *n* (%)	No, *n* (%)	Yes, *n* (%)	No, *n* (%)	Yes, *n* (%)	No, *n* (%)
Klebsiella pneumoniae	35 (70.0)	15 (30.0)	43 (86.0)	7 (14.0)	33 (66.0)	17 (34.0)	35 (70.0)	15	35 (70.0)	15 (30.0)
Escherichia coli	33 (49.3)	34 (50.7)	52 (77.6)	15 (22.4)	22 (32.8)	45 (67.2)	33 (49.3)	34	33 (49.3)	34 (50.7)
Enterobacter cloacae	20 (95.2)	1 (4.8)	21 (100)	0 (0)	20 (95.2)	1 (4.8)	20 (95.2)	1	20 (95.2)	1 (4.8)
Citrobacter freundii	1 (100)	0 (0)	1 (100)	0 (0)	1 (100)	0 (0)	1 (100)	0	1 (100)	0 (0)
Klebsiella oxytoca	1 (100)	0 (0)	1 (100)	0 (0)	1 (100)	0 (0)	1 (100)	0	1 (100)	0 (0)
Total	90	50	118	22	77	63	90	50	90	50

CIM, carbapenem inactivation assay; CPE, carbapenemase-producing Enterobacterales; I, intermediate; MIC, minimum inhibitory concentration; R, resistant.
[a]CPE is defined as isolates belonging to the order Enterobacterales with positivity for carbapenem inactivation *in vitro* and/or detection of carbapenemase genes.
[b]Ertapenem susceptibility was determined using VITEK2 XL, interpreted according to CLSI guidelines (MIC interpretation: S ≤ 0.5 mg/L, I = 1 mg/L, R ≥ 2 mg/L).
[c]Meropenem susceptibility was determined using VITEK2 XL, interpreted according to CLSI guidelines(MIC interpretation: S ≤ 1 mg/L, I = 2 mg/L, R ≥ 4 mg/L).
[d]Molecular detection of carbapenemase genes were performed by qRT-PCR. Genes included in the panel: bla_{NDM}, bla_{VIM}, $bla_{OXA\text{-}48\text{-}like}$, bla_{KPC}.

gene, and 1 isolate (1.1%) harbouring both $bla_{OXA\text{-}48}$ and bla_{NDM} (Figure 1, Table S3).

Characteristics of carbapenemase-producing Enterobacterales

Rectal colonization with CPE was detected in 83 of 376 (22.1%) screened patients with 7 patients (7/83; 8.4%) colonized by more than one CPE. Almost all *Enterobacter cloacae* complex (20/21; 95%) growing on selective CRE agar were carbapenemase producers (NDM). In contrast, only 70% (35/50) *Klebsiella pneumoniae* isolated from CRE agar were carbapenemase producers, and only (33/34; 49%) of *Escherichia coli* growing on CRE agar were CPEs. NDM was the predominant carbapenemase encountered (78/90; 86.7%) and was the predominant carbapenemase in both *K. pneumoniae* and the *E. cloacae* complex (70% and 95%, respectively). OXA-48-like was more common in *E. coli*, with 11/13 isolates harbouring the $bla_{OXA\text{-}48\text{-}like}$ gene (Figure 1a and b). NDM-type carbapenemases were typically found in isolates exhibiting high ertapenem and/or meropenem MICs, whereas OXA-48-like–producing Enterobacterales could be phenotypically susceptible to ertapenem and/or meropenem. Seven of 12 (58.3%) OXA-48-like–producing Enterobacterales were ertapenem-resistant but meropenem-susceptible, and 4 of 12 (33.3%) isolates were both ertapenem-susceptible and meropenem-susceptible (Figure 1c and d).

Phenotypic resistance to meropenem was the best predictor for carbapenemase production, with a sensitivity of 85.6% (95% CI: 76.6%–92.1%), 100% specificity (95% CI: 92.9%–100%), 100% positive predictive value (PPV) and 79.4% negative predictive value (NPV). Although ertapenem AST had a higher sensitivity of 95.6% (95% CI: 89%–98.8%), the specificity was significantly lower than meropenem AST, at only 36% (95% CI: 22.9%–50.8%), with 72.9 PPV and 81.8% NPV. Thus, meropenem resistance was the best predictor of carbapenemase genes and carbapenemase activity in our study. Ertapenem AST was more sensitive in detecting OXA-48-like producers than meropenem AST (8/12 isolates producing OXA-48-like only were ertapenem resistant versus 1/12 meropenem resistant).

Discussion

Our evaluation of the screening algorithm indicated that culturing of rectal swabs on selective CRE agar lacked specificity to detect CPE. The incorporation of meropenem AST exhibited a higher specificity than ertapenem AST in detecting CPE isolates. However, relying solely on meropenem AST may lead to the oversight of OXA-48-like producers.[5] Performing an additional assay on ertapenem- and/or meropenem-resistant isolates to detect meropenem hydrolysing activity, such as the CIM assay, can significantly increase the specificity while retaining the highest sensitivity, especially in resource-limited settings like ours in Vietnam where access to PCR is not feasible. It has been reported that strains with low expression of carbapenemases may be missed due to the sensitivity of the mSuperCARBA plates. However, compared with other plates, mSuperCARBA has been reported to have the highest sensitivity for detection of carbapenemase producers.[14,15] Our PPS study revealed that 22.1% of the screened patients exhibited colonization with CPE, and 8.4% were colonized by more than one species. The predominant carbapenemase genes identified were bla_{NDM} and $bla_{OXA\text{-}48\text{-}like}$. Interestingly, our analysis did not reveal the presence of bla_{KPC} in the isolates from this PPS, which contrasts somewhat with expectations. Previous studies in Vietnam had indicated a high prevalence of bla_{KPC} and bla_{NDM}.[16–18]

Supporting our findings, a recent study by Yen *et al.*[19] investigating the prevalence of CRE in rural Vietnam identified both bla_{NDM} and bla_{OXA} as the predominant carbapenemase genes. It is worth noting that many studies are conducted in hospital settings, often involving critically ill patients, particularly those in ICUs, or are focused on specific bacterial species.[16,17,20] Such study settings may introduce selection bias, and prevalence studies should consider sampling diverse populations, settings and regional variations to understand the landscape of CPE.[21] The rise

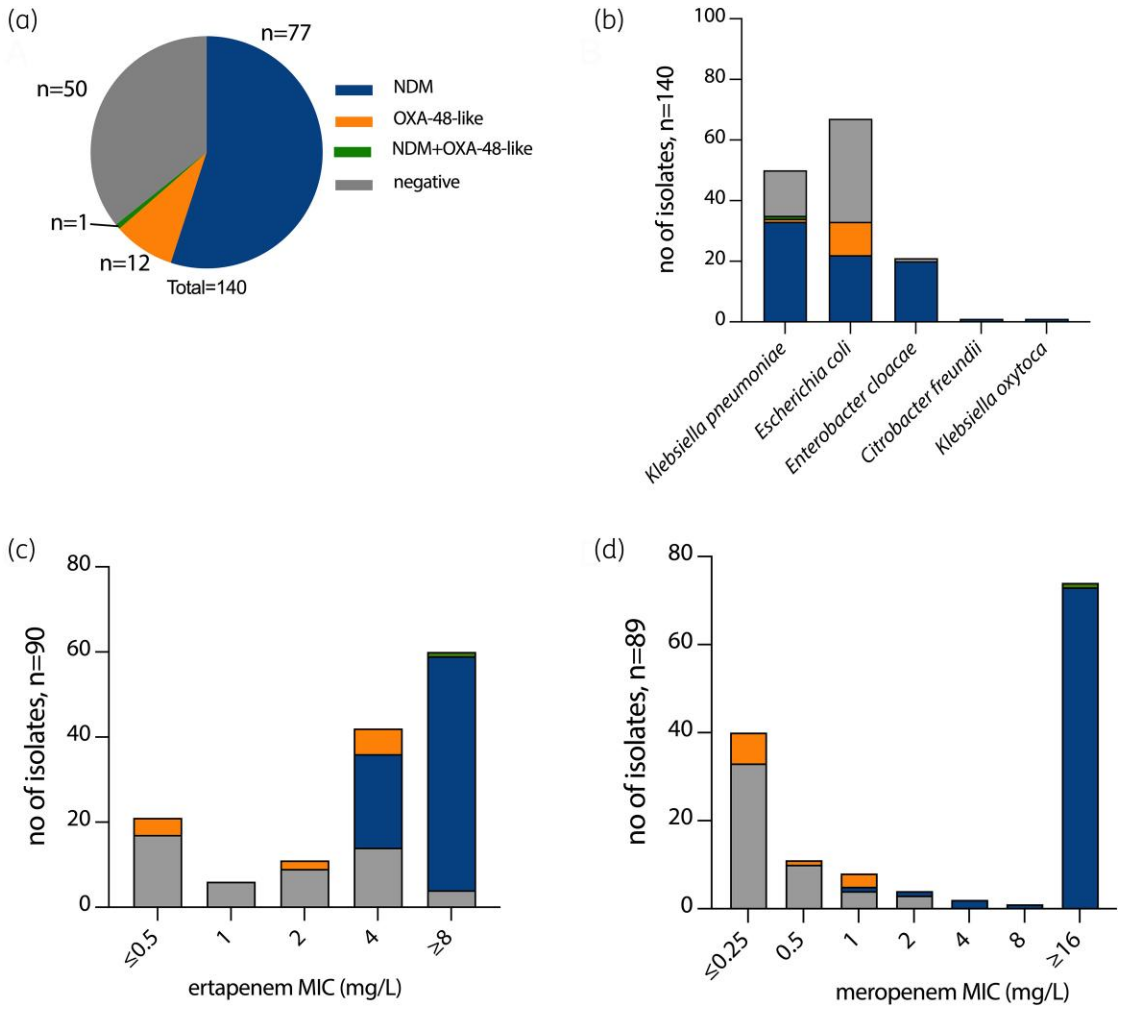

Figure 1. Detection of carbapenemase genes in carbapenem-resistant Enterobacterales (CRE) from rectal swabs in Vietnam. (a) Frequency (%) of carbapenemase gene detection in CRE and (b) by species ($n=140$). (c) Distribution of carbapenemase genes by ertapenem MIC. (d) Distribution of carbapenemase genes by meropenem MIC, one missing value for meropenem MIC (total $n=89$).

of NDM as a prevalent carbapenemase is indeed alarming.[19,22] NDM-type β-lactamase is one of the most potent β-lactamases, which cannot be inhibited by any β-lactamase inhibitors approved for clinical use, highlighting the urgent need for heightened surveillance and effective containment measures.[23]

The accuracy of detection of carbapenemase-encoding genes is determined by the targets included in the PCR panel. Our panel included only the five most common carbapenemases, so some rare carbapenemases may have been missed. However, by including the meropenem disc hydrolysis assay, we would have been able to detect carbapenemases not included in our PCR panel and overcome this limitation. We did not find any discrepancy between CIM and qRT-PCR, suggesting that most carbapenemases would have been detected with our chosen targets. Our results suggest that colony growth on mSuperCARBA selective agar combined with phenotypic resistance to meropenem is a good predictor for detecting CPE and underscores the significance of incorporating molecular methods such as PCR to monitor the dynamics of carbapenemase gene spread in high-prevalence regions such as Vietnam. Although our PCR assay may not have characterized the specific carbapenemase gene subtypes, we believe that our study contributes valuable data to the collective efforts in combating AMR in the region. We strongly advocate for the inclusion of genome surveillance as part of a comprehensive approach to surveillance measures. Integrating WGS analysis can provide a deeper understanding of the genetic variations and transmission patterns, enabling more targeted and effective strategies to curb the spread of resistant strains and enhance overall antimicrobial stewardship.

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The I-CRECT team

[A]Håkan Hanberger[1,2], Bent Jörgensen[2,3], Åse Östholm Balkhed[1], Lennart E. Nilsson[1]
[B]Mattias Larsson[2,4], Linus Olson[2,4,5],
[C]Thirumalaisamy P. Velavan[6,7], Le Huu Song[7,8], Nhat My Truong[7], Bui Tien Sy[7,8], Alexa Purgreth[6]
[D]Flavie Goutard[9], Etienne Loire[9]
[E]Y.M. Gildas Hounmanou[10], Anders Dalsgaard[10]
[F]Phuc D. Pham[11], Linh Viet. Nguyen[11], Tuyet Hanh T. Tran[12], Minh V. Hoang[13]
[G]Dien M.Tran[14,15,16], Phuc H. Phan[2,14], Nhung T. Nguyen[14], Ngoc T.B Hoang[17], Lien T.L Pham[14], Nhung Hong[18]

A. Linköping University, Linköping, Sweden.
B. Karolinska Institutet, Stockholm, Sweden.
C. University of Tübingen, Tübingen, Germany.
D. The French Agricultural Research Centre for International Development (CIRAD), Montpellier, France.
E. University of Copenhagen, Copenhagen, Denmark.
F. Hanoi University of Public Health, Hanoi, Vietnam.
G. Vietnam National Children's Hospital, Hanoi, Vietnam.

Author details

1. Department of Clinical Microbiology and Department of Clinical and Experimental Medicine, Linköping University, Linkoping, Sweden.
2. Training and Research Academic Collaboration (TRAC)—Sweden—Vietnam.
3. Department of Global Studies Göteborg University, Gothenburg, Sweden.
4. Department of Global Public Health, Karolinska Institutet, Stockholm, Sweden.
5. Department of Women's and Children's Health, Karolinska Institutet, Sweden.
6. Institute of Tropical Medicine, University of Tübingen, Tübingen, Germany.
7. Vietnamese-German Center for Medical Research, Hanoi, Vietnam.
8. Director Board, 108 Military Central Hospital, Hanoi, Vietnam.
9. Department of Vietnam, The French Agricultural Research Centre for International Development (CIRAD), Montpellier, France.
10. Department of Veterinary and Animal Sciences, University of Copenhagen, Copenhagen, Denmark.
11. Vietnam One Health University Network (VOHUN), Center for Public Health and Ecosystem Research (CENPHER)RFER, Hanoi University of Public Health, Hanoi, Vietnam.
12. Department of Research Management and Cooperation, Hanoi University of Public Health, Hanoi, Vietnam.
13. Director Board, Hanoi University of Public Health, Hanoi, Vietnam.
14. Director Board, Vietnam National Children's Hospital, Hanoi, Vietnam.
15. Training and Research Institute for Child Health, Vietnam National Children's Hospital, Hanoi, Vietnam.
16. University of Medicine and Pharmacy—Vietnam National University, Hanoi, Vietnam.
17. Department of Microbiology, Vietnam National Children's Hospital, Hanoi, Vietnam.
18. Department of Infection control, Vietnam National Children's Hospital, Hanoi, Vietnam.

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Transparency declarations

None of the authors have any conflicts of interest to declare. All authors of this research have approved this manuscript and agreed to submission of the manuscript.

Ethical approval

The study is part of the JPIAMR-funded I-CRECT project titled 'Intervention to decrease CRE colonization and transmission between hospitals, households, communities and domesticated animals.' This study was approved by the Ethical Review Board of the Ministry of Health, Vietnam, and performed after ethical approval from the Ethical Review Board of Vietnam National Children's Hospital with operating code VNCH-TRICH-2022-87 dated 30 September 2022 issued by the Vietnam Ministry of Health, and the Ethical Review Board of Hanoi University of Public Health (HUPH) with operating code 022-350/DD-YTCC dated 25 July 2022, issued by HUPH.

Supplementary data

Tables S1 to S3 are available as Supplementary data at *JAC-AMR* Online.

References

1 Tacconelli E, Carrara E, Savoldi A *et al*. Discovery, research, and development of new antibiotics: the WHO priority list of antibiotic-resistant bacteria and tuberculosis. *Lancet Infect Dis* 2018; **18**: 318–27. https://doi.org/10.1016/S1473-3099(17)30753-3

2 Antimicrobial Resistance Collaborators. Global burden of bacterial antimicrobial resistance in 2019: a systematic analysis. *Lancet* 2022; **399**: 629–55. https://doi.org/10.1016/S0140-6736(21)02724-0

3 Sihombing B, Bhatia R, Srivastava R *et al*. Response to antimicrobial resistance in South-East Asia region. *Lancet Reg Health Southeast Asia* 2023; **18**: 100306. https://doi.org/10.1016/j.lansea.2023.100306

4 Torumkuney D, Kundu S, Vu GV *et al*. Country data on AMR in Vietnam in the context of community-acquired respiratory tract infections: links between antibiotic susceptibility, local and international antibiotic prescribing guidelines, access to medicines and clinical outcome. *J Antimicrob Chemother* 2022; **77**: i26–34. https://doi.org/10.1093/jac/dkac214

5 Nordmann P, Poirel L. Epidemiology and diagnostics of carbapenem resistance in gram-negative bacteria. *Clin Infect Dis* 2019; **69**: S521–8. https://doi.org/10.1093/cid/ciz824

6 Morrill HJ, Pogue JM, Kaye KS *et al*. Treatment options for carbapenem-resistant Enterobacteriaceae infections. *Open Forum Infect Dis* 2015; **2**: ofv050. https://doi.org/10.1093/ofid/ofv050

7 Kocer K, Boutin S, Dalpke AH *et al*. Comparative genomic analysis reveals a high prevalence of inter-species in vivo transfer of carbapenem-resistance plasmids in patients with haematological malignancies. *Clin Microbiol Infect* 2020; **26**: 780.e1–e8. https://doi.org/10.1016/j.cmi.2019.10.014

8 Iovleva A, Doi Y. Carbapenem-resistant Enterobacteriaceae. *Clin Lab Med* 2017; **37**: 303–15. https://doi.org/10.1016/j.cll.2017.01.005

9 Tamma PD, Goodman KE, Harris AD *et al*. Comparing the outcomes of patients with carbapenemase-producing and non-carbapenemase-producing carbapenem-resistant Enterobacteriaceae bacteremia. *Clin Infect Dis* 2017; **64**: 257–64. https://doi.org/10.1093/cid/ciw741

10 Hoellinger B, Deboscker S, Danion F *et al*. Incidence and time-to-onset of carbapenemase-producing Enterobacterales (CPE) infections in CPE carriers: a retrospective cohort study. *Microbiol Spectr* 2022; **10**: e0186822. https://doi.org/10.1128/spectrum.01868-22

11 van Hattem JM, Arcilla MS, Bootsma MC *et al*. Prolonged carriage and potential onward transmission of carbapenemase-producing Enterobacteriaceae in Dutch travelers. *Future Microbiol* 2016; **11**: 857–64. https://doi.org/10.2217/fmb.16.18

12 van der Zwaluw K, de Haan A, Pluister GN *et al*. The carbapenem inactivation method (CIM), a simple and low-cost alternative for the carba NP test to assess phenotypic carbapenemase activity in gram-negative rods. *PLoS One* 2015; **10**: e0123690. https://doi.org/10.1371/journal.pone.0123690

13 Probst K, Boutin S, Bandilla M *et al*. Fast and automated detection of common carbapenemase genes using multiplex real-time PCR on the BD MAX system. *J Microbiol Methods* 2021; **185**: 106224. https://doi.org/10.1016/j.mimet.2021.106224

14 Amar M, Shalom O, Adler A. Comparative evaluation of a new commercial media, the CHROMAgar mSuperCARBA, for the detection of carbapenemase-producing Enterobacteriaceae. *Diagn Microbiol Infect Dis* 2017; **88**: 20–2. https://doi.org/10.1016/j.diagmicrobio.2017.02.004

15 Garcia-Fernandez S, Hernandez-Garcia M, Valverde A *et al*. CHROMagar mSuperCARBA performance in carbapenem-resistant Enterobacteriaceae isolates characterized at molecular level and routine surveillance rectal swab specimens. *Diagn Microbiol Infect Dis* 2017; **87**: 207–9. https://doi.org/10.1016/j.diagmicrobio.2016.11.014

16 Tran DM, Larsson M, Olson L *et al*. High prevalence of colonisation with carbapenem-resistant Enterobacteriaceae among patients admitted to Vietnamese hospitals: risk factors and burden of disease. *J Infect* 2019; **79**: 115–22. https://doi.org/10.1016/j.jinf.2019.05.013

17 Pham MH, Hoi LT, Beale MA *et al*. Evidence of widespread endemic populations of highly multidrug resistant *Klebsiella pneumoniae* in hospital settings in Hanoi, Vietnam: a prospective cohort study. *Lancet Microbe* 2023; **4**: e255–63. https://doi.org/10.1016/S2666-5247(22)00338-X

18 Tada T, Tsuchiya M, Shimada K *et al*. Dissemination of carbapenem-resistant *Klebsiella pneumoniae* clinical isolates with various combinations of carbapenemases (KPC-2, NDM-1, NDM-4, and OXA-48) and 16S rRNA methylases (RmtB and RmtC) in Vietnam. *BMC Infect Dis* 2017; **17**: 467. https://doi.org/10.1186/s12879-017-2570-y

19 Yen NTP, Nhung NT, Phu DH *et al*. Prevalence of carbapenem resistance and its potential association with antimicrobial use in humans and animals in rural communities in Vietnam. *JAC Antimicrob Resist* 2022; **4**: dlac038. https://doi.org/10.1093/jacamr/dlac038

20 Roberts LW, Hoi LT, Khokhar FA *et al*. Genomic characterisation of multidrug-resistant *Escherichia coli*, *Klebsiella pneumoniae*, and *Acinetobacter baumannii* in two intensive care units in Hanoi, Viet Nam: a prospective observational cohort study. *Lancet Microbe* 2022; **3**: e857–66. https://doi.org/10.1016/S2666-5247(22)00181-1

21 van Duin D, Doi Y. The global epidemiology of carbapenemase-producing Enterobacteriaceae. *Virulence* 2017; **8**: 460–9. https://doi.org/10.1080/21505594.2016.1222343

22 Hoang CQ, Nguyen HD, Vu HQ *et al*. Emergence of New Delhi metallo-beta-lactamase (NDM) and *Klebsiella pneumoniae* carbapenemase (KPC) production by *Escherichia coli* and *Klebsiella pneumoniae* in Southern Vietnam and appropriate methods of detection: a cross-sectional study. *Biomed Res Int* 2019; **2019**: 9757625. https://doi.org/10.1155/2019/9757625

23 Mojica MF, Rossi MA, Vila AJ *et al*. The urgent need for metallo-beta-lactamase inhibitors: an unattended global threat. *Lancet Infect Dis* 2022; **22**: e28–34. https://doi.org/10.1016/S1473-3099(20)30868-9