Bush-Jacoby-Medeiros (Integron) system	Ambler system	Enzyme activity — Examples
Group 1 cephalosporinases	C	Chromosomal AmpC resistance (usual) to β-lactams, except carbapenems; not inhibited by clavulanic acid (e.g., *Enterobacteriaceae*, except *Salmonella* and *Klebsiella*)
Group 2 penicillinases	A	Staphylococcal penicillinases;TEM-1, TEM-2, SHV-1 (broadspectrum); TEM and SHV variants (e.g., ESBLs,1 cephalosporinase inhibited by clavulanic acid; carbapenemases inhibited by clavulanic acid) CTX-M
	D	Oxacillin-hydrolyzing, OXA; cloxacillin; ESBL cephalosporins; weak activity for carbapenems (e.g,. *A baumannii*)
Group 3 metallo-β-lactamase	B	Zinc-dependent carbapenemases; resistant to inactivation by clavulanic acid, sulbactam, tazobactam, some to aztreonam; (*bla*$_{VIM-1}$, *bla*$_{VIM-2}$) *Pseudomonas aeruginosa* ... IMP (e.g., *Pseudomonas putida, Serratia marcescens, A baumannii, K pneumoniae, Klebsiella oxytoca*, et al. NDM-1 in *E coli, K pneumoniae, Enterobacter cloacae*)

Table 1. Classification of β-lactamase enzymes

Ambler class	Enzyme	Function	Known organisms
A	KPC[1]	Hydrolyzes all β-lactam antibiotics; inhibited by clavulanate	*K pneumoniae*, Enterobacteriaceae
B	MBLs[2] (NDM, IMP, VIM, GIM, SPM)	Hydrolyze all β-lactams except aztreonam; may be inhibited by clavulanate; require zinc for enzymatic activity; inhibited by EDTA	*P aeruginosa*, *Acinetobacter* spp, Enterobacteriaceae
D	OXA	Oxacillin hydrolyzing; less able to hydrolyze carbapenems	*P aeruginosa*, *A baumannii*, Enterobacteriaceae

Table 2. Classification of carbapenemases (metallo-β-Lactamases — MBLs)

MCR-1 (colistin-resistant GN/*E. coli*)

Emergence of plasmid-mediated colistin resistance mechanism MCR-1 in animals and human beings in China: a microbiological and molecular biological study

- Polymyxin resistance has involved chromosomal mutations but has never been reported via horizontal gene transfer
- A routine surveillance project on antimicrobial resistance in commensal *E. coli* from food animals in China → major increase of colistin resistance
- *E. coli* strain SHP45 (processing colistin resistance that could be transferred to another strain) was isolated from a pig
- Further analysis of possible plasmid-mediated polymyxin resistance
- Report the emergence of the first plasmid-mediated polymyxin resistance mechanism, MCR-1, in *Enterobacteriaceae*
- Mcr-1 gene in *E. coli* strain SHP45 was identified by whole plasmid sequencing and subcloning

เชื้อจุลชีพดื้อยาล่าสุด!!!
พบในปศุสัตว์และคนไข้ที่ประเทศจีน
ดื้อต่อยา Colistin (plasmid transferrable) !!!

Lancet Infect Dis 2016; 16: 161-68

International Journal of Infectious Diseases 54 (2017) 77–84

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Review

How should we respond to the emergence of plasmid-mediated colistin resistance in humans and animals?

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mcr-1
- Human isolates
29 countries
- Food animal isolates
28 countries

Middle East /Southeast Asia

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Multi-drug resistant bacteria

SUMMARY

Objective: The widespread use of antibiotics in humans and animals has contributed to growing rates of antibiotic resistance. Previously treatable bacterial infections now require the last line of antibiotics or are untreatable. The current antibiotic of last resort for carbapenem-resistant Gram-negative bacterial infections is often colistin. Evidence for the shifting pattern of colistin resistance and how the international community should respond are discussed in this review.
Methods: The literature on colistin resistance was reviewed.
Results: Plasmid-mediated colistin resistance encoded by *mcr-1* was first documented in China during the routine surveillance of food animals. This has been followed by similar reports across a wide geographic area, in humans, animals, and the environment. The *mcr-1* gene has been reported among human isolates in 29 countries, related to environmental samples in four countries, and in food animals and other animals in 28 countries. More recently, a second gene encoding resistance, *mcr-2*, has been isolated from porcine and bovine *Escherichia coli*.
Conclusion: The emergence and horizontal transmission of colistin resistance highlights the need for heightened stewardship efforts across the One Health platform for this antibiotic of last resort, and indeed for all antibiotics used in animals and humans.

Identification of a novel plasmid-mediated colistin-resistance gene, *mcr-2*, in *Escherichia coli*, Belgium, June 2016

BB Xavier [1,2,3], C Lammens [1,2,3], R Ruhal [1,2,3], S Kumar-Singh [1,3,4], P Butaye [5,6,7], H Goossens [1,2,3], S Malhotra-Kumar [1,2,3]

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6. CODA-CERVA, Brussels, Belgium
7. Ross University School of Veterinary Medicine, Basseterre, Saint Kitts and Nevis

Correspondence: Surbhi Malhotra-Kumar (surbhi.malhotra@uantwerpen.be)

mcr-2
Bovine/ porcine colistin-resistant *E. coli*
In Belgium, prevalence of mcr-2 gene > mcr-1 gene

lasmid-mediated colistin-resistance gene, mcr-
50-7917.ES.2016.21.27.30280

epted on 07 July 2016 / published on 07 July 2016

We identified a novel plasmid-mediated colistin-resistance gene in porcine and bovine colistin-resistant *Escherichia coli* that did not contain *mcr-1*. The gene, termed *mcr-2*, a 1,617 bp phosphoethanolamine transferase harboured on an IncX4 plasmid, has 76.7% nucleotide identity to *mcr-1*. Prevalence of *mcr-2* in porcine colistin-resistant *E. coli* (11/53) in Belgium was higher than that of *mcr-1* (7/53). These data call for an immediate introduction of *mcr-2* screening in ongoing molecular epidemiological surveillance of colistin-resistant Gram-negative pathogens.

Carbapenem-Resistant *Acinetobacter baumannii* and *Enterobacteriaceae* in South and Southeast Asia

Li-Yang Hsu,[a,b,c] Anucha Apisarnthanarak,[d] Erum Khan,[e] Nuntra Suwantarat,[f] Abdul Ghafur,[g] Paul Anantharajah Tambyah[b]

Saw Swee Hock School of Public Health, National University of Singapore, Singapore[a]; Yong Loo Lin School of Medicine, National University of Singapore, Singapore[b]; Tan Tock Seng Hospital, Singapore[c]; Thammasat University Hospital, Pathum Thani, Thailand[d]; Aga Khan University, Karachi, Pakistan[e]; Chulabhorn International College of Medicine, Thammasat University, Pathum Thani, Thailand[f]; Apollo Hospital, Chennai, India[g]

Published 19 October 2016

Citation Hsu L-Y, Apisarnthanarak A, Khan E, Suwantarat N, Ghafur A, Tambyah PA. 2017. Carbapenem-resistant *Acinetobacter baumannii* and *Enterobacteriaceae* in South and Southeast Asia. Clin Microbiol Rev 30:1–22. https://doi.org/10.1128/CMR.00042-16.

In Thailand (limited data)

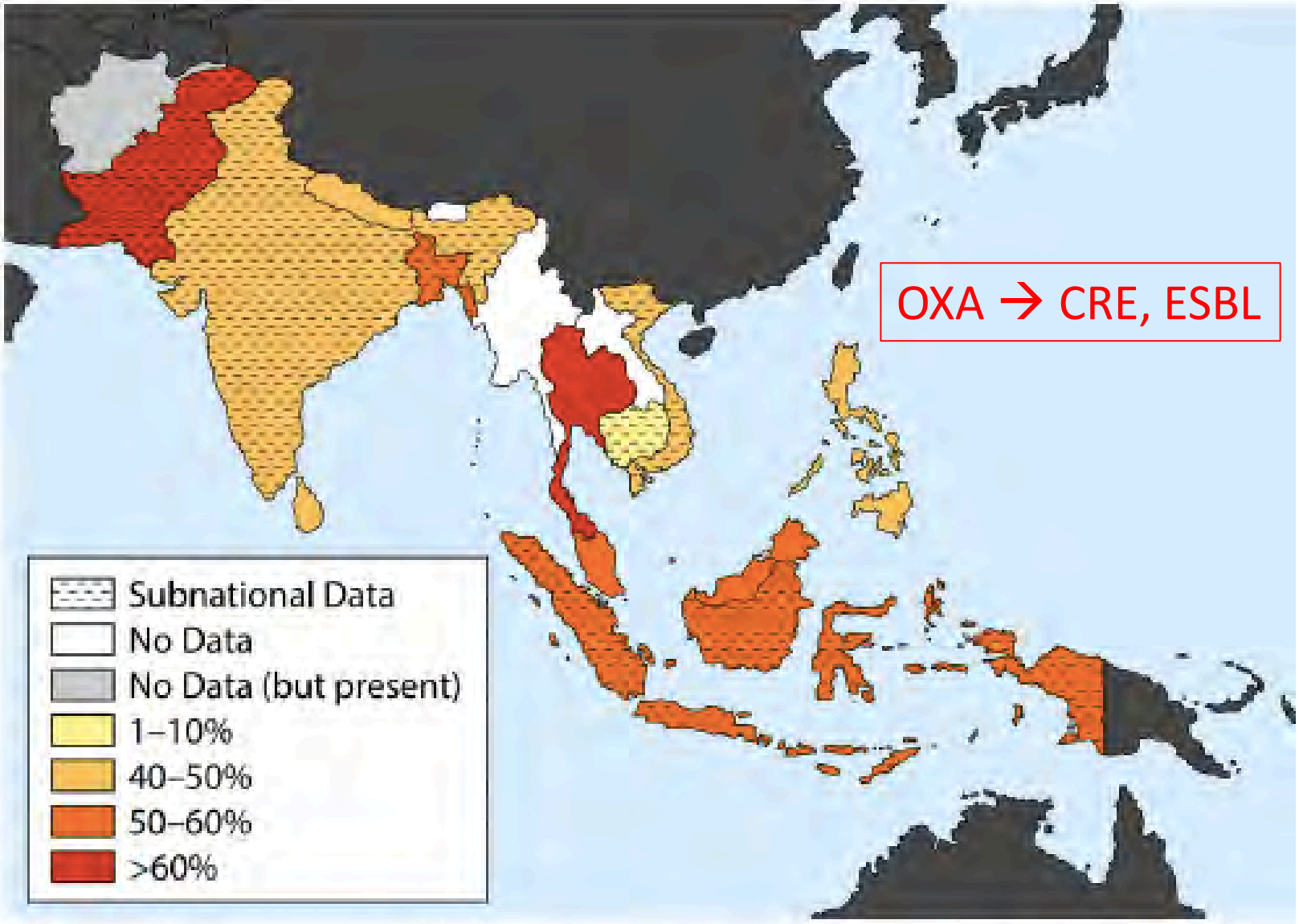
What is the most common MDR-GN Pathogen?

ESBLs

Acinetobacter baumannii

Pseudomonas aeruginosa

CRE (Carbapenem Resistance Enterobacteriaceae)



OXA → CRE, ESBL

FIG 1 Estimated prevalence of carbapenem-resistant *Acinetobacter baumannii* in South and Southeast Asian countries.

OXA in Acinetobacter = intrinsic resistance

OXA in Enterobactericeae = plasmid resistance

AmpC = SPACE bacteria
Chromosomal Resistance
Do not use 3rd gen cephalosporin

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Table 1. Classification of β-lactamase enzymes

Suwantarat and Carroll *Antimicrobial Resistance and Infection Control* (2016) 5:15
DOI 10.1186/s13756-016-0115-6

Antimicrobial Resistance and Infection Control

GUIDELINES ARTICLE Open Access

Epidemiology and molecular characterization of multidrug-resistant Gram-negative bacteria in Southeast Asia

Nuntra Suwantarat[1,2*] and Karen C. Carroll[2,3]

International Journal of Antimicrobial Agents 39 (2012) 311–316

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International Journal of Antimicrobial Agents

journal homepage: http://www.elsevier.com/locate/ijantimicag

Comparative in vitro activity of carbapenems against major Gram-negative pathogens: results of Asia-Pacific surveillance from the COMPACT II study

Pattarachai Kiratisin[a,*], Anan Chongthaleong[b], Thean Yen Tan[c], Evelina Lagamayo[d], Sally Roberts[e], Jemelyn Garcia[f], Todd Davies[g]

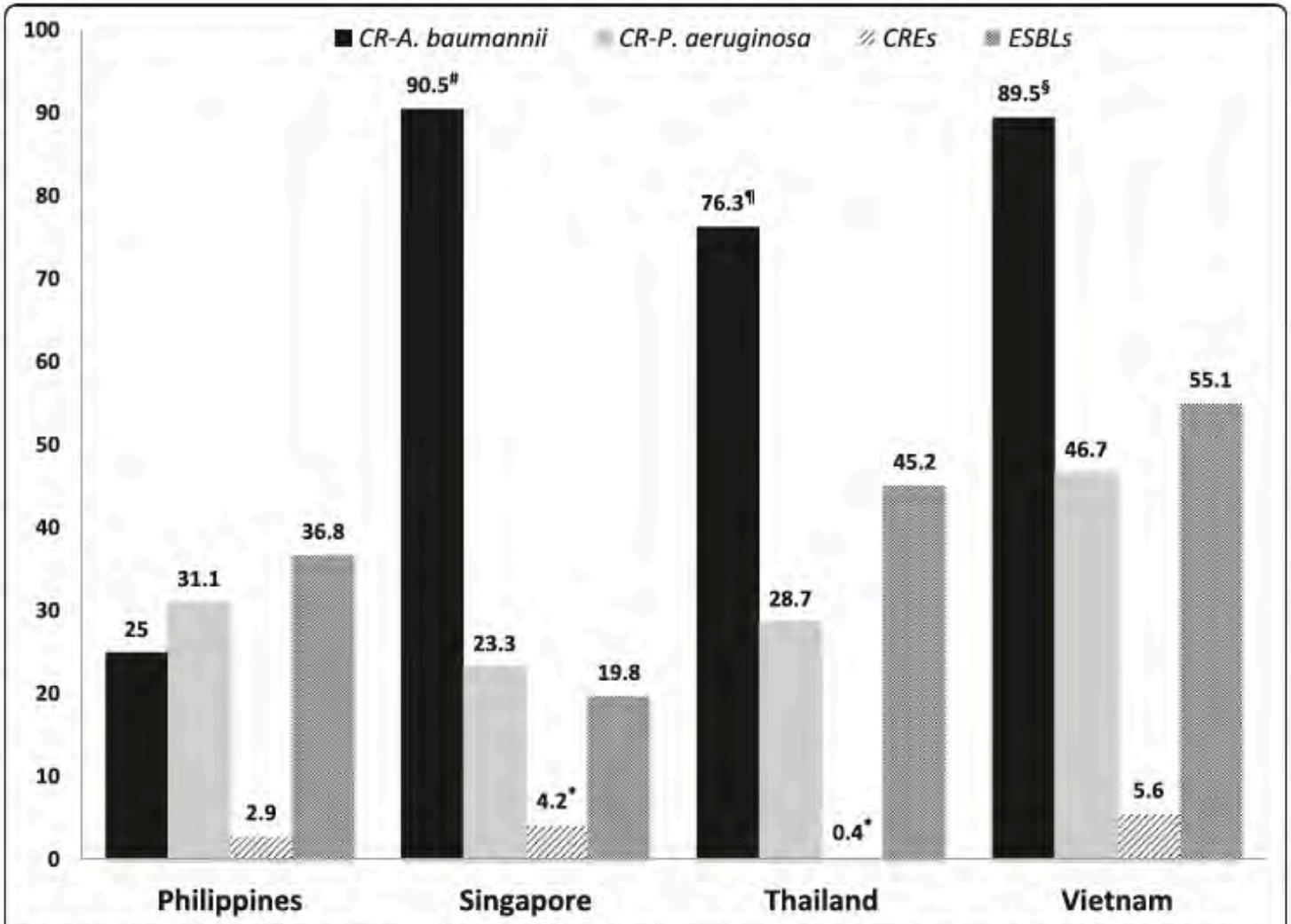


Fig. 1 Prevalence (%) of extended-spectrum β-lactamases (ESBLs) and carbapenem-resistant organisms by country in Southeast Asia, adopted from reference 5 (COMPACT II study). The organisms were obtained during April – July 2010, from 5 Centers in Asia-Pacific countries including New Zealand (data not shown), the Philippines (3 centers, 16 *A. baumannii* isolates, 90 *P. aeruginosa* isolates, 70 *Enterobacteriaceae* isolates), Singapore (3 centers, 21 *A. baumannii* isolates, 120 *P. aeruginosa* isolates, 96 *Enterobacteriaceae* isolates), Thailand (10 centers, 59 *A. baumannii* isolates, 296 *P. aeruginosa* isolates, 239 *Enterobacteriaceae* isolates) and Vietnam (3 centers, 19 *A. baumannii* isolates, 90 *P. aeruginosa* isolates, 71 *Enterobacteriaceae* isolates). There are small numbers of *A. baumannii* isolations tested from reference 5. Prevalence of CRAB in other studies are #70.5-91 % (Singapore) [6, 43, 44], ¶46.7-80 % (Thailand) [29–31] and §more than 90 % (Vietnam) [48]. *Recent studies have been shown the increasing prevalence of CRE in Singapore and Thailand [1, 3, 4, 6, 22]. Abbreviation; CR, carbapenem–resistant; CRE, carbapenem-resistant *Enterobacteriaceae*; ESBLs, extended-spectrum β-lactamases

Table 4
Prevalence (%) of extended-spectrum β-lactamase-producing Enterobacteriaceae.

Country	ICU isolates	Non-ICU isolates	All isolates
New Zealand	0	0	0
Philippines	58.8	27.5	36.8
Singapore	17.2	21.2	19.8
Thailand	44.4	45.3	45.2
Vietnam	81.0	43.8	55.1
Overall	43.8	37.6	39.4

ICU, Intensive Care Unit.

Kiratisin P, COMPACT II study, IJAA, 2012

National data?

Local data?

* Hospital antibiogram

Extended Spectrum Beta-Lactamases (ESBLs)

- Type (group) of plasmid beta-lactamases produced by *E.coli* and *Klebsiella sp*.
- Detect in lab by hydrolysis of BL ring of cefotax., Ceftriaxone, Ceftazidime, and/or Aztreonam
- Do not hydrolyse BL ring of Cephamycins (Cefoxitin or Cefotetan) or carbapenems.

*Cephamycins works only in-vitro test
Associated with treatment failure in clinical trials

No need to perform ESBL confirmation test,
if use new CLSI breakpoints (2010)

Etest®

Cefotaxime/cefotaxime + clavulanic acid

Ceftazidime/ceftazidime + clavulanic acid

Cefepime/cefepime + clavulanic acid

For *in vitro* confirmation of ESBL

Interpretation

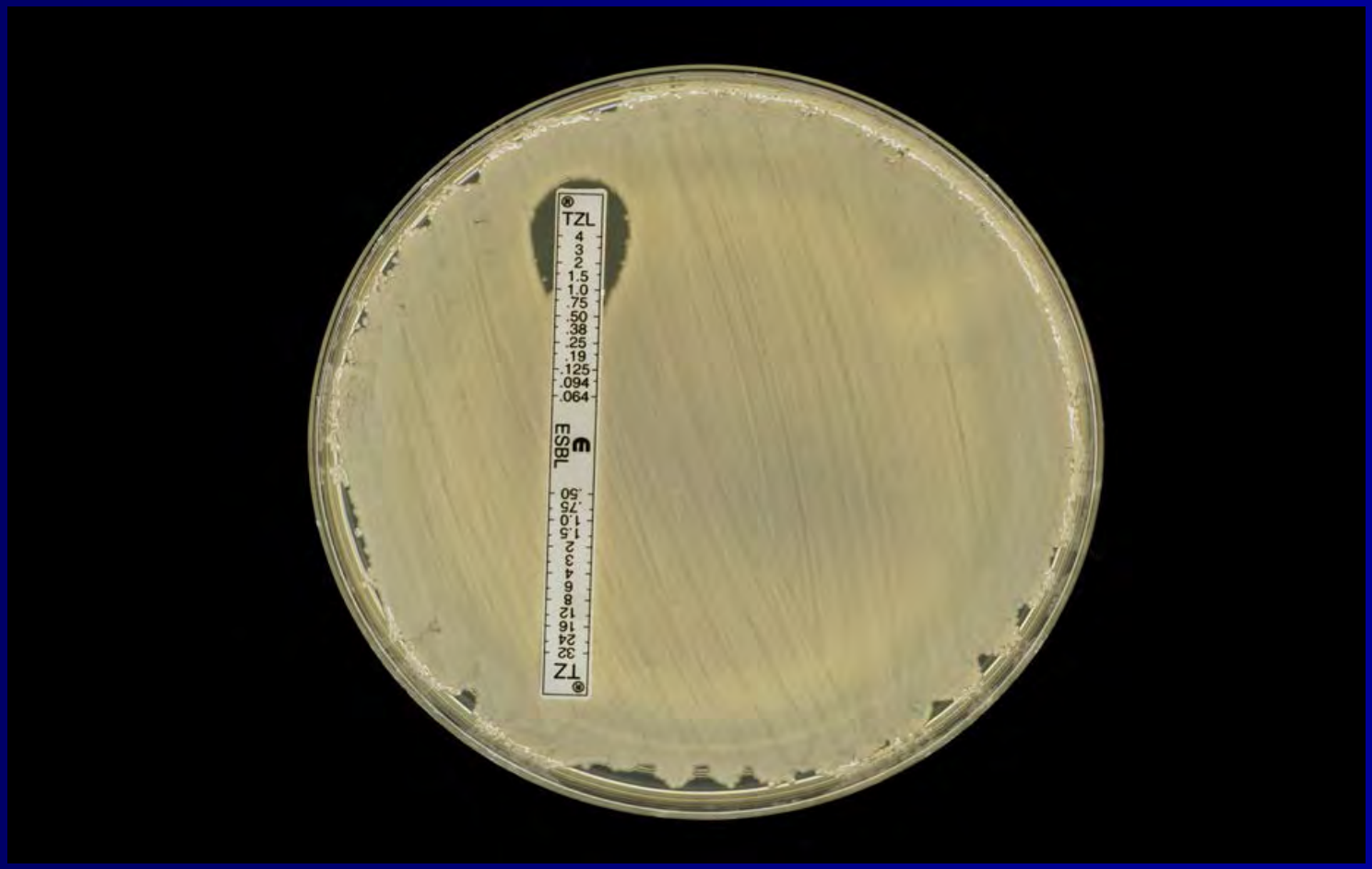
Table 1: Guidelines for interpretation of Etest ESBL.

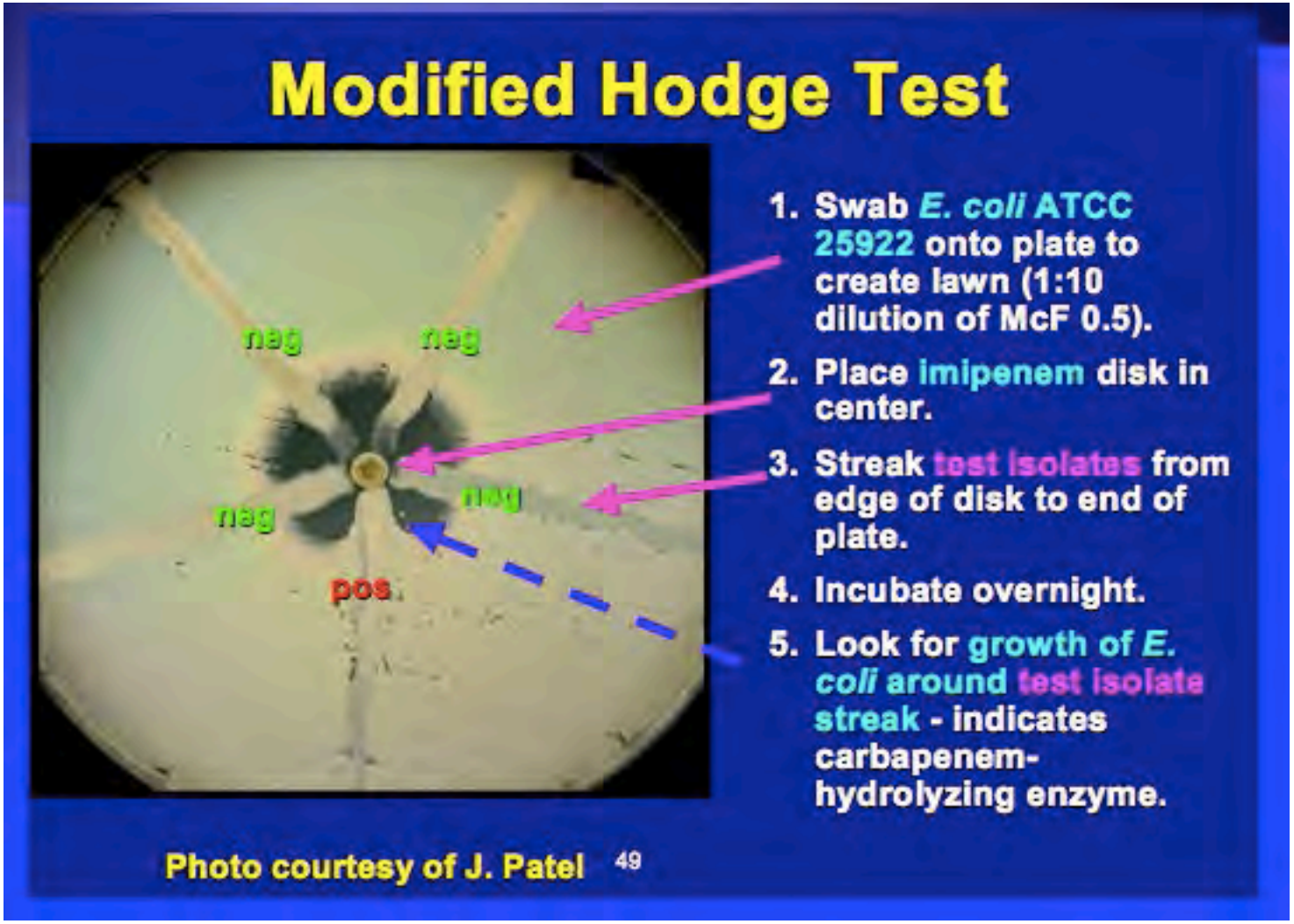
ESBL	MIC Ratio	Reporting
Positive	CT ≥ 0.5 and CT/CTL ≥ 8 OR TZ ≥ 1 and TZ/TZL ≥ 8 OR PM ≥ 0.25 and PM/PML ≥ 8 OR "Phantom" zone or deformation of the CT, TZ or PM ellipse	ESBL producer and resistant to all penicillins, cephalosporins and aztreonam (CLSI M100-S series).
Negative	CT < 0.5 or CT/CTL < 8 AND TZ < 1 or TZ/TZL < 8	ESBL non-producer and report actual MICs of relevant drugs as determined by a MIC method.
Non-determinable (ND)	CT > 16 and CTL > 1 AND TZ > 32 and TZL > 4 AND PM > 16 and PML > 4 OR When one strip is ESBL negative and the other ND	ESBL non-determinable and report actual MICs of relevant drugs as determined by a MIC method. If ESBL is suspected, confirm results with genotyping.

Different growth-inhibition patterns:

Figure 4. Clear cut ESBL positive:
MIC CT/CTL =1.5/0.047 = 32

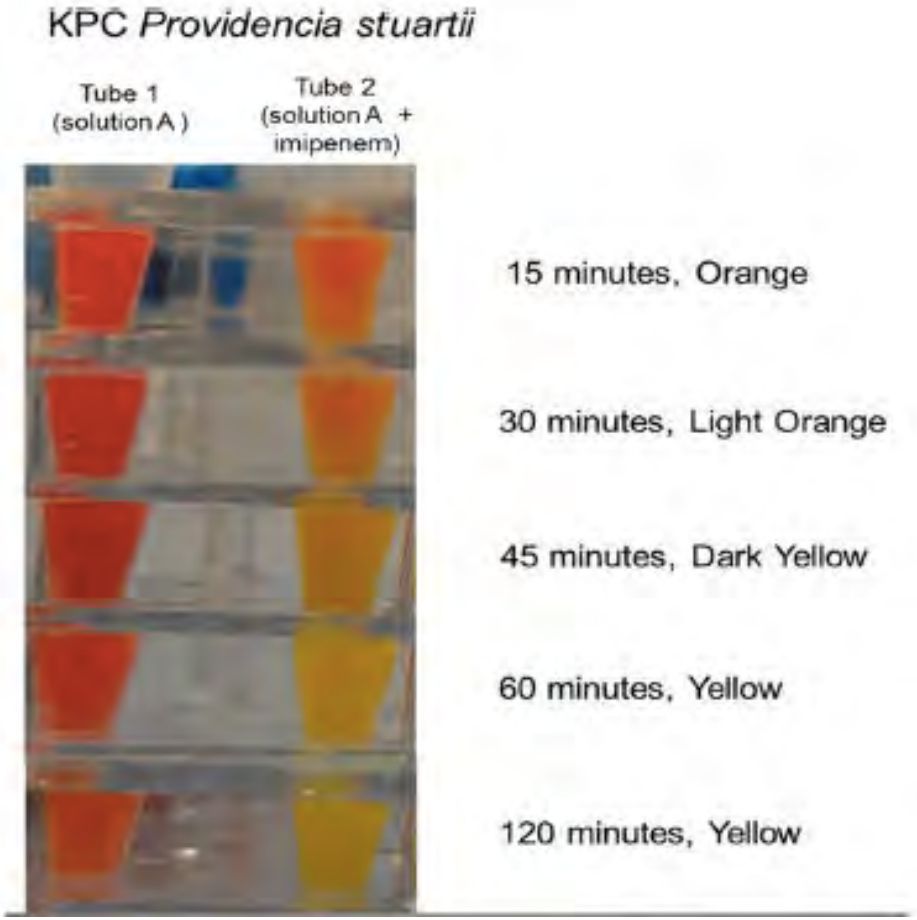
Extended spectrum beta lactamase production test showing an organism which is positive for the enzyme. In this case, the organism is totally resistant to ceftazadime, but shows inhibited growth with ceftazadime+clavulinic acid. The addition of clavulinic acid interferes with the production of the ESBL, so there is some sensitivity to ceftazadime.





CRE confirmation test

No need to perform if use new CLSI breakpoints (2010)



Comparison of a Novel, Rapid Chromogenic Biochemical Assay, the Carba NP Test, with the Modified Hodge Test for Detection of Carbapenemase-Producing Gram-Negative Bacilli

Shawn Vasoo,[a] Scott A. Cunningham,[a] Peggy C. Kohner,[a] Patricia J. Simner,[a] Jayawant N. Mandrekar,[b] Karen Lolans,[c] Mary K. Hayden,[c,d] Robin Patel[a,e]

JCM, Sep 2013

The CIM (Carbapenemase inactivation method) a new phenotypic Test to assess Carbapenemase activity

PLOS ONE | The CIM, a New Phenotypic Test to Assess Carbapenemase Activity

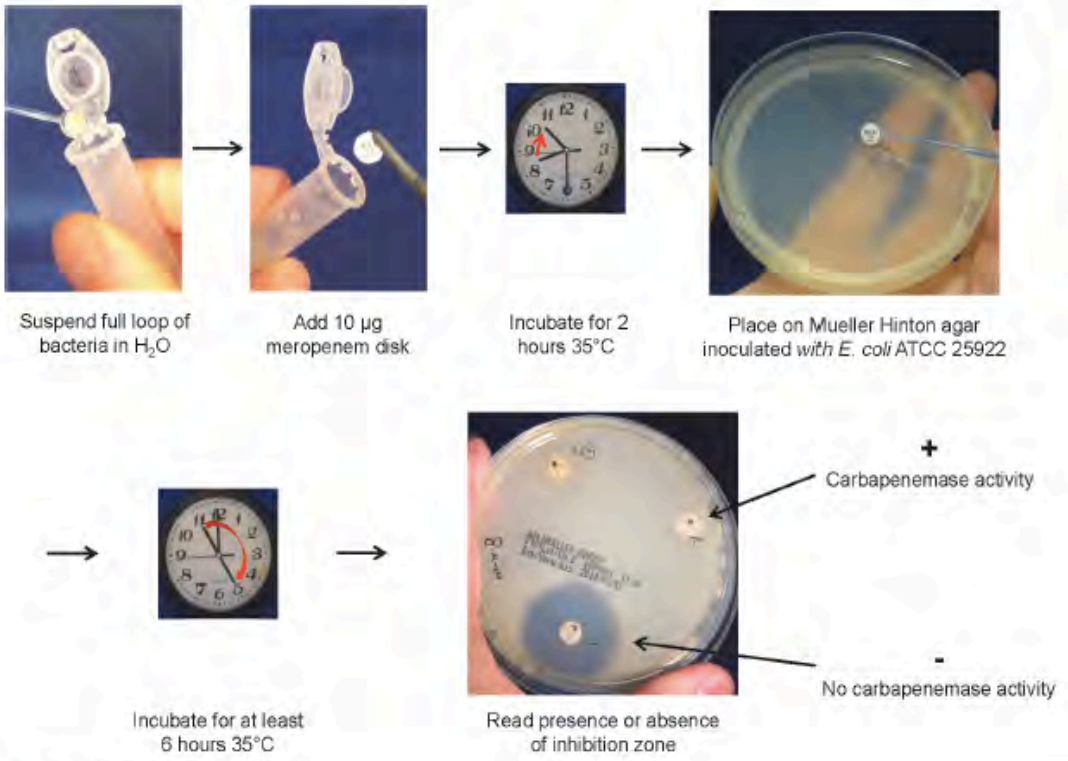


Fig 1. Schematic of the CIM.

doi:10.1371/journal.pone.0123690.g001

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(3): e0123690.