Carbapenemases in *Enterobacteriaceae*

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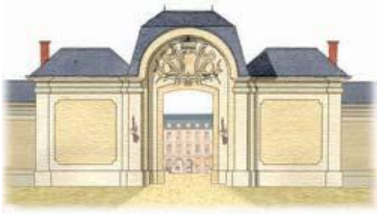
Inserm U914

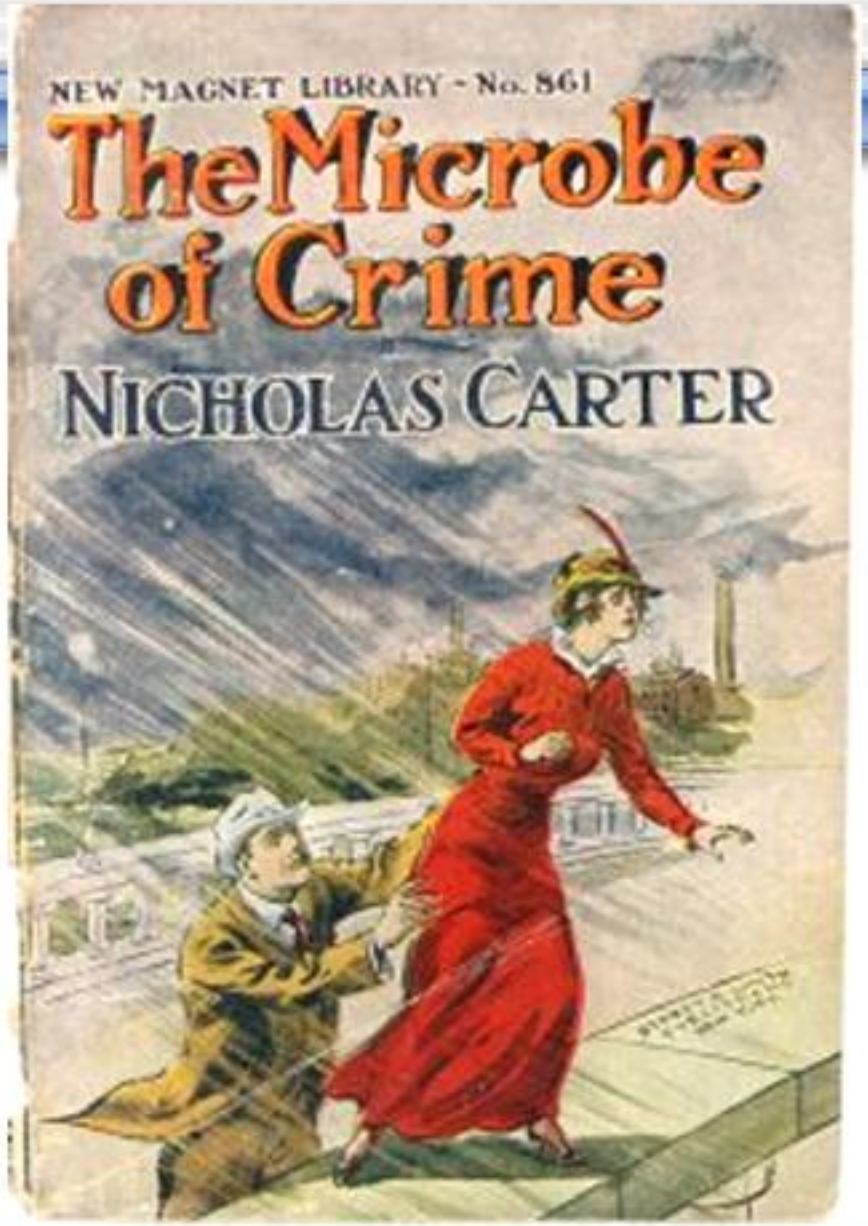
Résistances Emergentes aux Antibiotiques



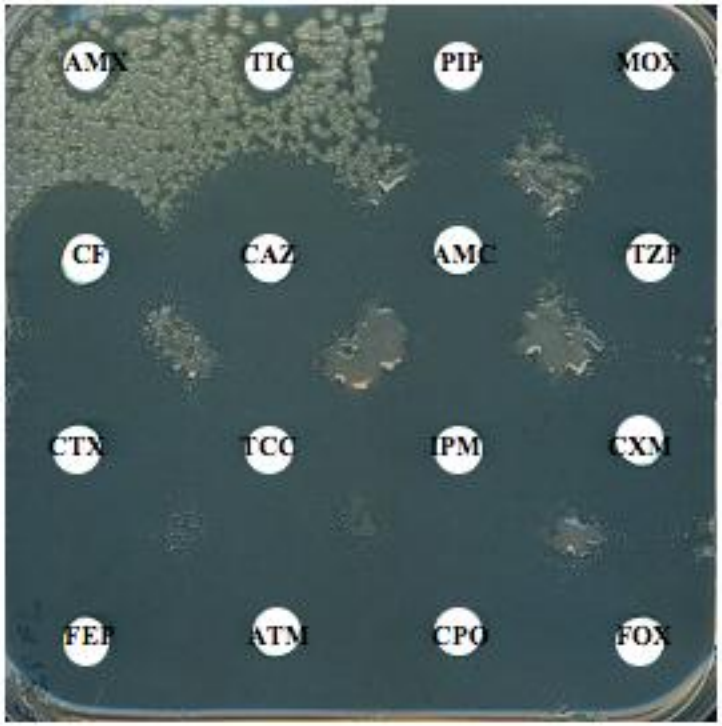
Prof P. Nordmann

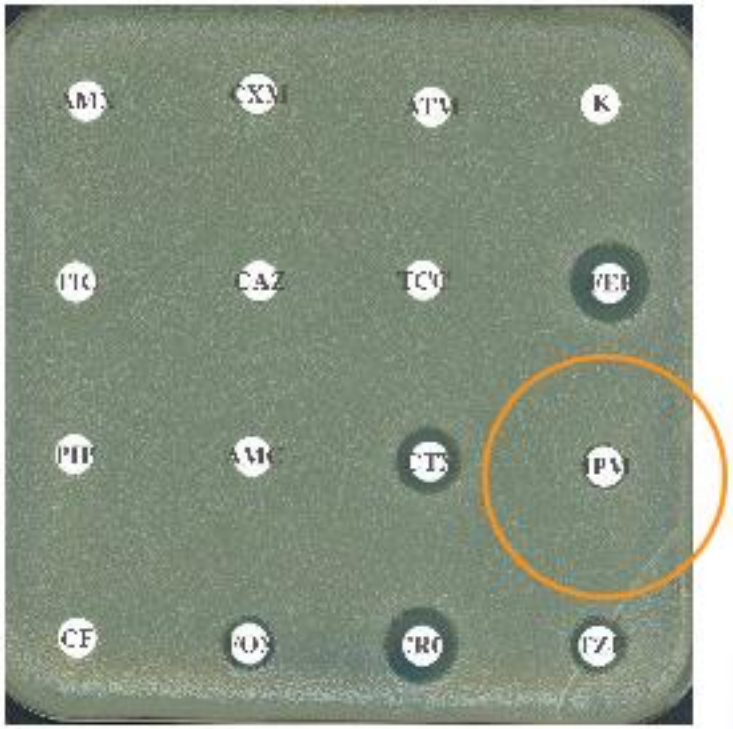
Hôpital de Bicêtre, INSERM 914, Faculté de Médecine

et Université Paris Sud

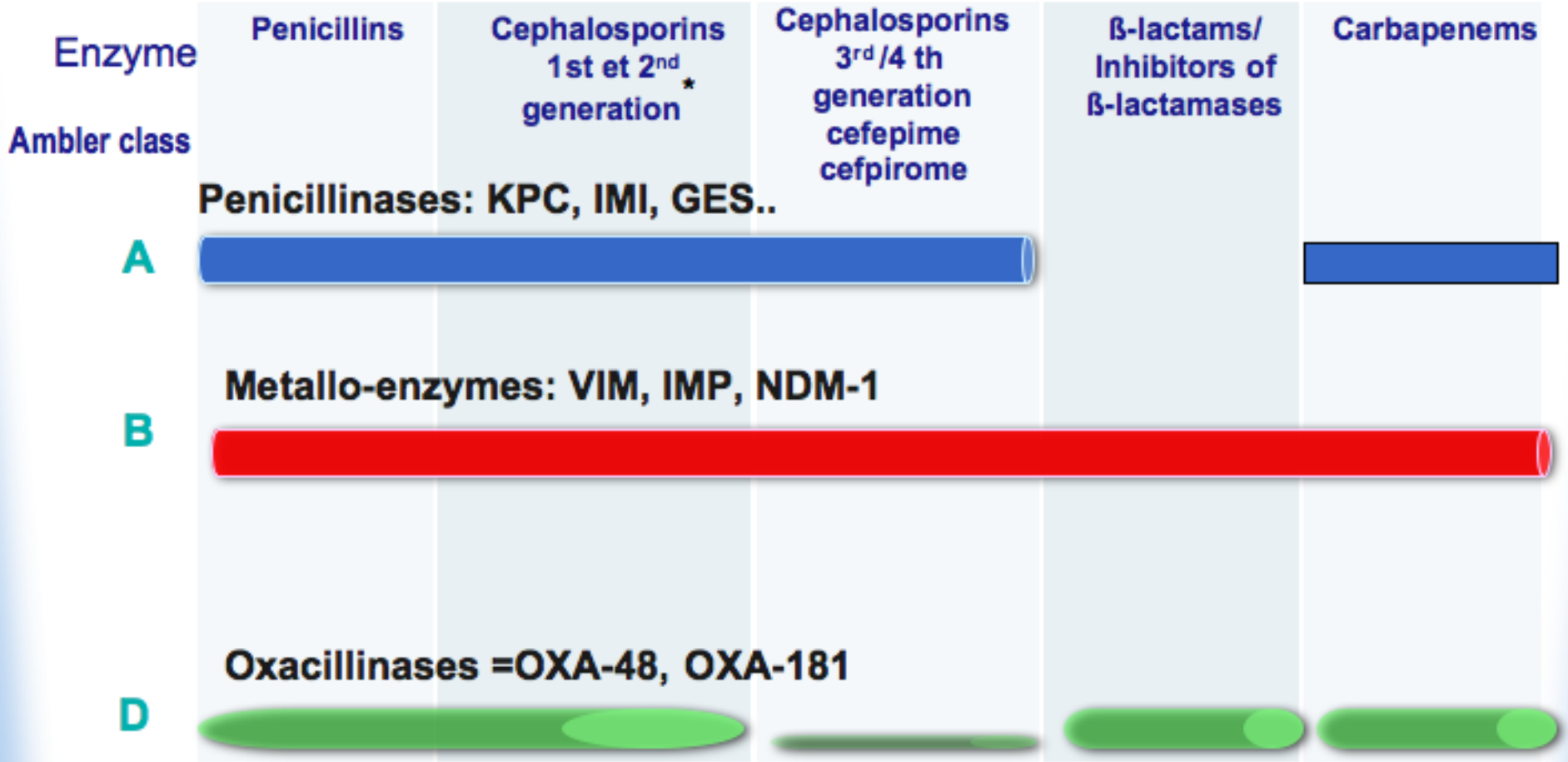

NEW MAGNET LIBRARY - No. 861
The Microbe of Crime
NICHOLAS CARTER

MDR in Gram negatives 2010-2020 = carbapenem-resistance in *Enterobacteriaceae*



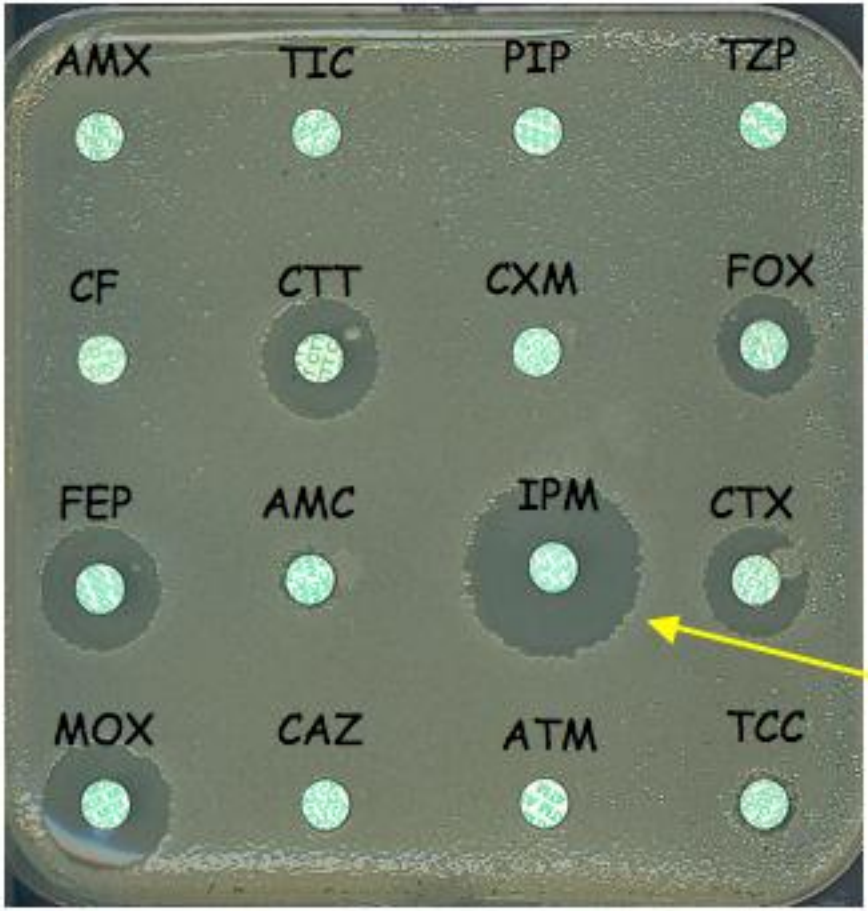


The carbapenemases in *Enterobacteriaceae*



* Cephamycins excluded for most class As

The KPCs



ANTIMICROBIAL AGENTS AND CHEMOTHERAPY, Apr. 2001, p. 1151–1161 Vol. 45, No. 4
0066-4804/01/$04.00+0 DOI: 10.1128/AAC.45.4.1151–1161.2001

Novel Carbapenem-Hydrolyzing β-Lactamase, KPC-1, from a Carbapenem-Resistant Strain of *Klebsiella pneumoniae*

HESNA YIGIT,[1] ANNE MARIE QUEENAN,[2] GREGORY J. ANDERSON,[1] ANTONIO DOMENECH-SANCHEZ,[3] JAMES W. BIDDLE,[1] CHRISTINE D. STEWARD,[1] SEBASTIAN ALBERTI,[4] KAREN BUSH,[2] AND FRED C. TENOVER[1]*

Hospital Infections Program, National Center for Infectious Diseases, Centers for Disease Control and Prevention, Atlanta, Georgia 30333[1]; The R. W. Johnson Pharmaceutical Research Institute, Raritan, New Jersey 08869[2]; and Unidad de Investigacion, Hospital Son Dureta, Andreu Doria, Palma de Mallorca, 07014,[4] and Área de Microbiología, Universidad de las Islas Baleares, Crta. Valldemosa, Palma de Mallorca, 07071,[3] Spain

Received 19 September 2000/Returned for modification 21 November 2000/Accepted 23 January 2001

✓KPC-1:
- plasmid encoded (50-kb)

✓Imipenem resistance or decreased susceptibility, inhibited by clavulanic acid

The KPC enzymes

ORIGINAL INVESTIGATION

Rapid Spread of Carbapenem-Resistant *Klebsiella pneumoniae* in New York City

A New Threat to Our Antibiotic Armamentarium

Simona Bratu, MD; David Landman, MD; Robin Haag, RN; Rose Recco, MD; Antonella Eramo, RN; Maqsood Alam, MD; John Quale, MD

Background: Carbapenem antibiotics are used to treat serious infections caused by extended-spectrum β-lactamase–carrying pathogens. Carbapenem resistance has been unusual in isolates of *Klebsiella pneumoniae*. In this study, the prevalence and molecular epidemiologic characteristics of carbapenem-resistant *K pneumoniae* are analyzed, and the experience involving 2 hospital outbreaks is described.

Methods: A citywide surveillance study was conducted in hospitals in Brooklyn. An observational study involving subsequent outbreaks at 2 hospitals was undertaken. Isolates were genetically fingerprinted by ribotyping and were examined for the presence of KPC-type carbapenem-hydrolyzing β-lactamases.

Results: Of 602 isolates of *K pneumoniae* collected during the citywide surveillance study, 45% had extended-spectrum β-lactamases. Of the extended-spectrum β-lactamase–producing isolates, 3.3% carried the carbapenem-hydrolyzing β-lactamase KPC-2. Several isolates were reported by the clinical microbiology laboratories as being susceptible to imipenem. Although all the isolates were resistant using agar diffusion methods, minimal inhibitory concentrations of imipenem were substantially lower for several isolates using standard broth microdilution tests and were highly dependent on the inoculum used. Two hospitals experienced the rapid spread of carbapenem-resistant isolates involving 5[illegible] 14-day mortality for bacteremic patie[illegible] isolates belonged to a single ribotype

Conclusions: Carbapenem-resistant[illegible] lates are rapidly emerging in New Yor[illegible] of a strain that possesses a carbapenem[illegible] tamase has occurred in regional hospt[illegible] Isolates are resistant to virtually all c[illegible] tibiotics, control of their spread is cru[illegible] tomated systems used for susceptibil[illegible] accurately identify all these isolates, w[illegible] hamper control efforts.

Arch Intern Med. 2005;165:1430-1435

K. pneumoniae KPC+: 33%: ST-type 258

Journal of Antimicrobial Chemotherapy (2007) **60**, 78–82
doi:10.1093/jac/dkm129
Advance Access publication 9 May 2007

JAC

Evolution of antimicrobial resistance among *Pseudomonas aeruginosa*, *Acinetobacter baumannii* and *Klebsiella pneumoniae* in Brooklyn, NY

David Landman[1], Simona Bratu[1], Sandeep Kochar[1], Monica Panwar[1], Manoj Trehan[1], Mehmet Doymaz[2] and John Quale[1]*

[1]*State University of New York – Downstate, Brooklyn, NY, USA;* [2]*Beth Israel Medical Center, New York, NY, USA*

Received 16 February 2007; returned 22 March 2007; revised 2 April 2007; accepted 5 April 2007

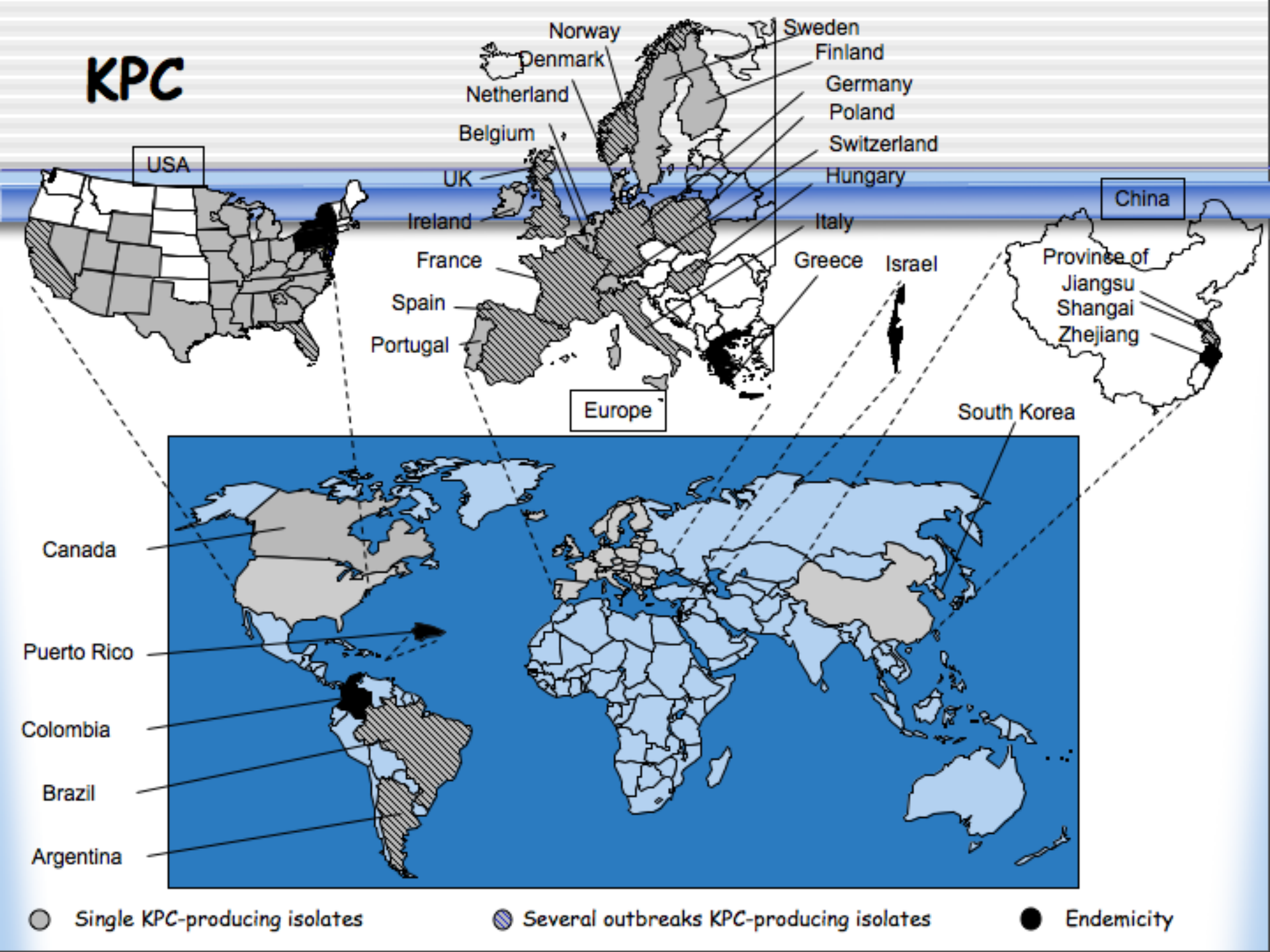
Objectives: To document resistance patterns of three important nosocomial pathogens, *Pseudomonas aeruginosa*, *Acinetobacter baumannii* and *Klebsiella pneumoniae*, present in hospitals in Brooklyn, NY.

Methods: Susceptibility profiles of pathogens gathered during a surveillance study in 2006 were analysed and compared with similar surveys performed in 1999 and 2001. MICs were determined according to CLSI standards, and selected isolates were screened by PCR for the presence of VIM, IMP and KPC β-lactamases.

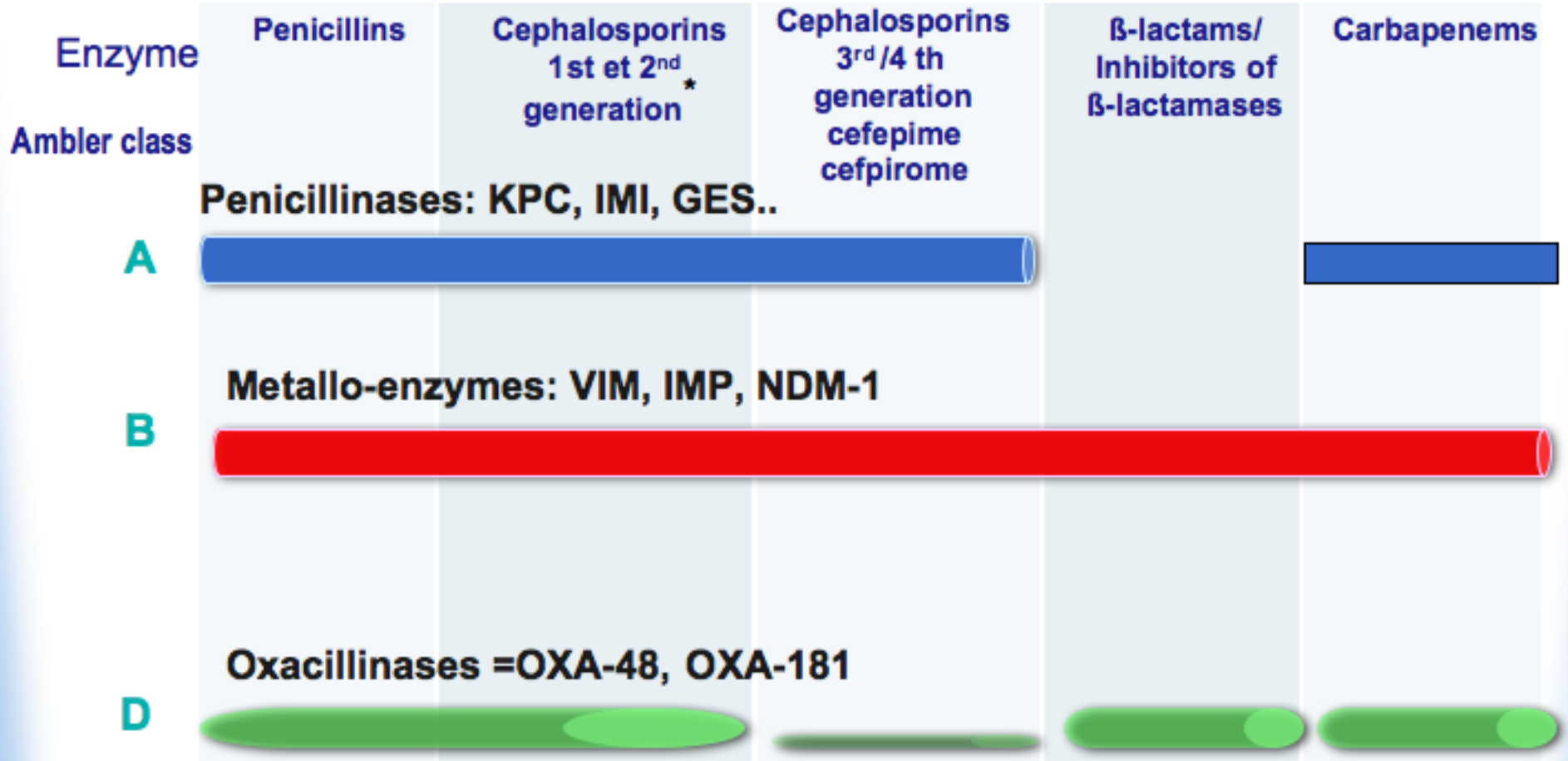
Results: For *P. aeruginosa*, susceptibility to most antimicrobials fell in 2001 and then reached a plateau. However, there was a progressive decrease in the number of patients with *P. aeruginosa* during the three surveys. While the total number of isolates of *A. baumannii* remained steady, there was a progressive decrease in susceptibility to most classes of antimicrobial agents, and approximately one-third had combined resistance to carbapenems, fluoroquinolones and aminoglycosides. There was a noticeable rise in the number of isolates of *K. pneumoniae* over the surveillance periods, suggesting that this has become the predominant pathogen in many medical centres. Over one-third of *K. pneumoniae* collected in 2006 carried the carbapenemase KPC, and 22% were resistant to all three classes of antimicrobial agents.

Conclusions: Hospitals in our region have been beset with antimicrobial-resistant Gram-negative bacteria. *K. pneumoniae* has rapidly emerged as the most common multidrug-resistant pathogen. Improved therapeutic agents and methods of detection are needed to reduce transmission of these bacteria.

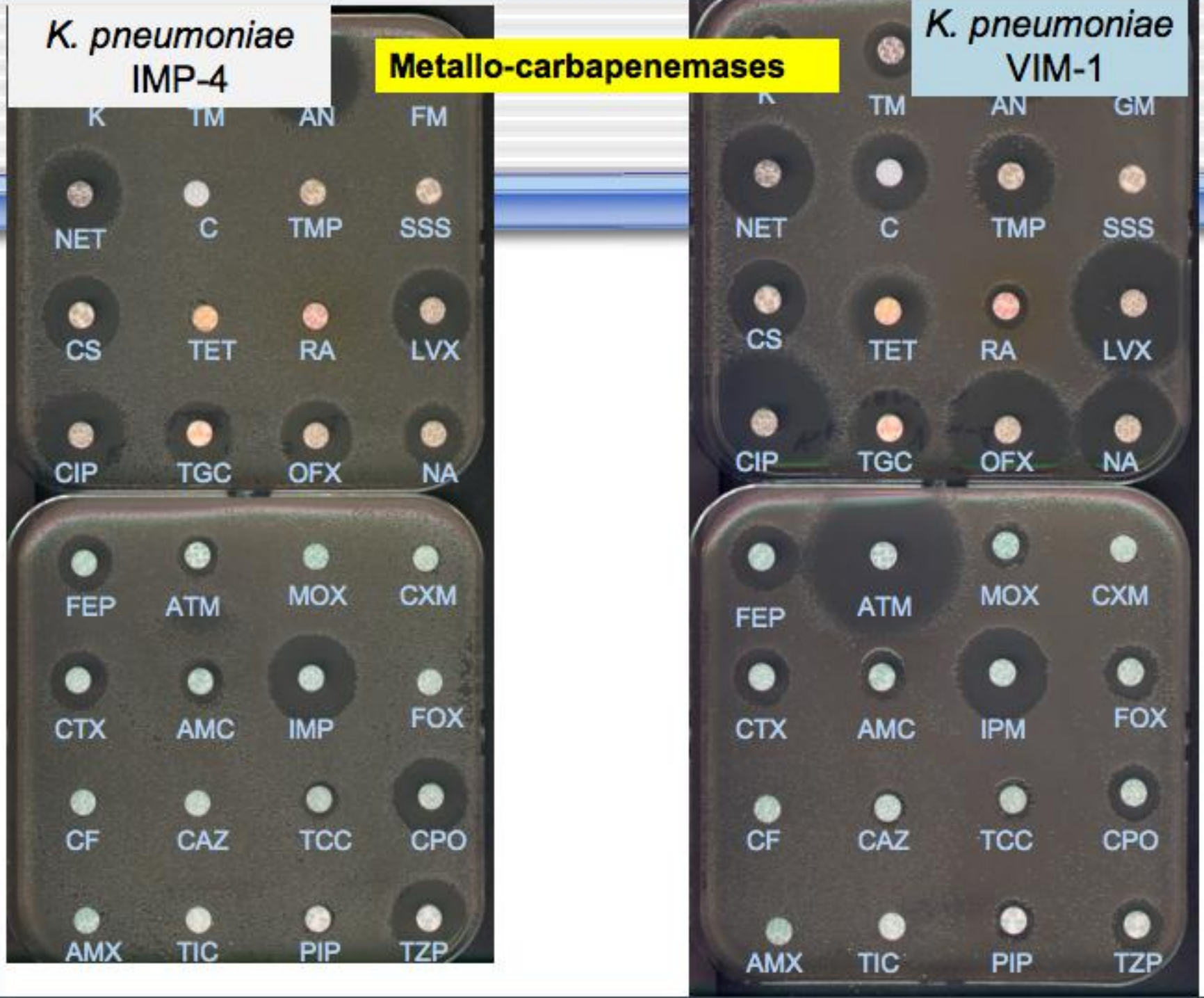
KPC


USA
Norway
Sweden
Denmark
Finland
Netherland
Germany
Poland
Belgium
Switzerland
UK
Hungary
Ireland
Italy
France
Greece
Israel
Spain
Portugal
Europe
China
Province of
Jiangsu
Shangai
Zhejiang
South Korea
Canada
Puerto Rico
Colombia
Brazil
Argentina
Single KPC-producing isolates
Several outbreaks KPC-producing isolates
Endemicity

The carbapenemases in *Enterobacteriaceae*



* Cephamycins excluded for most class As


K. pneumoniae
IMP-4
Metallo-carbapenemases
K. pneumoniae
VIM-1
K
TM
AN
FM
NET
C
TMP
SSS
CS
TET
RA
LVX
CIP
TGC
OFX
NA
FEP
ATM
MOX
CXM
CTX
AMC
IMP
FOX
CF
CAZ
TCC
CPO
AMX
TIC
PIP
TZP
K
TM
AN
GM
NET
C
TMP
SSS
CS
TET
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LVX
CIP
TGC
OFX
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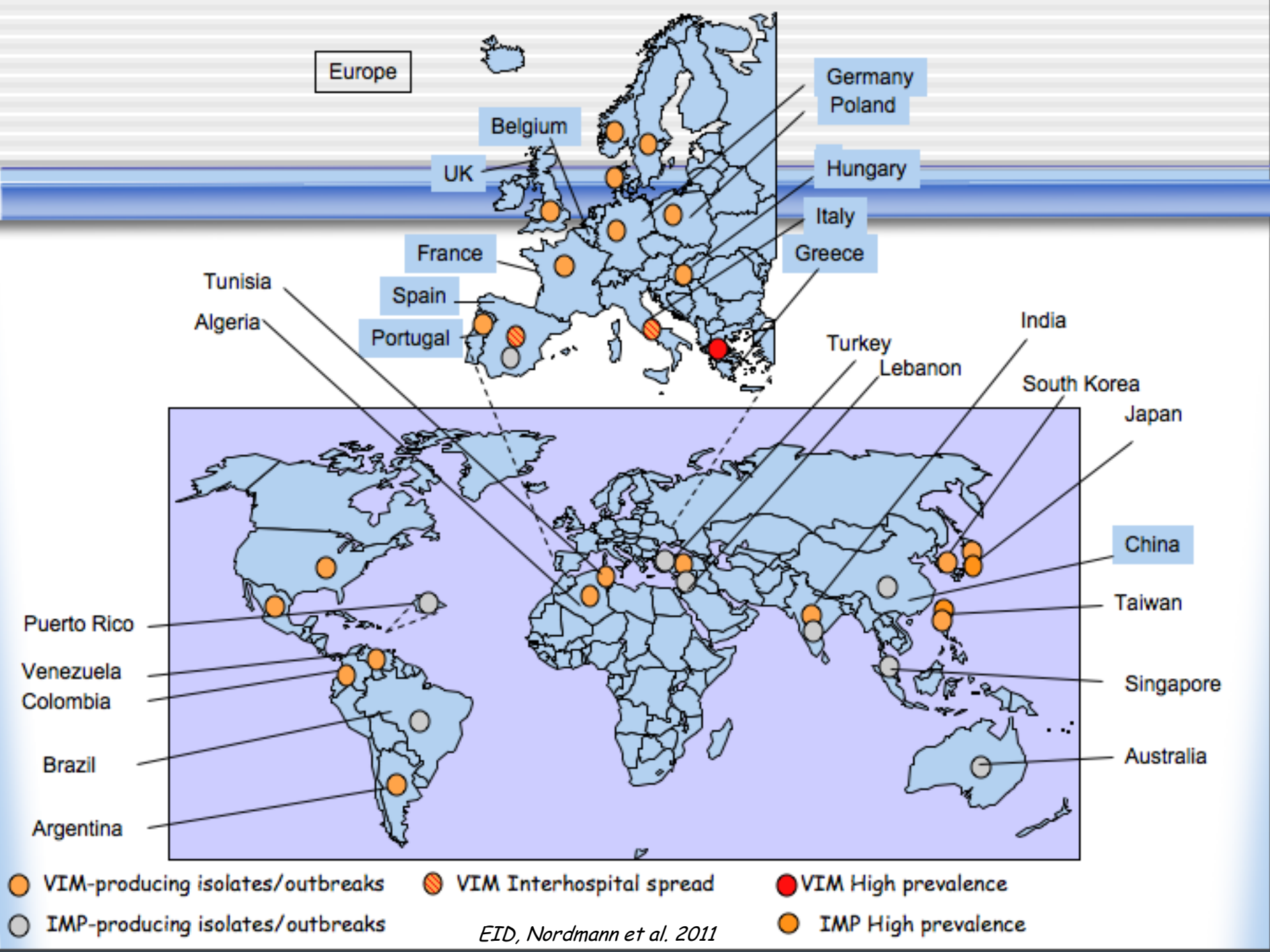

EID, Nordmann et al. 2011


THE LANCET Infectious Diseases
Impact Factor
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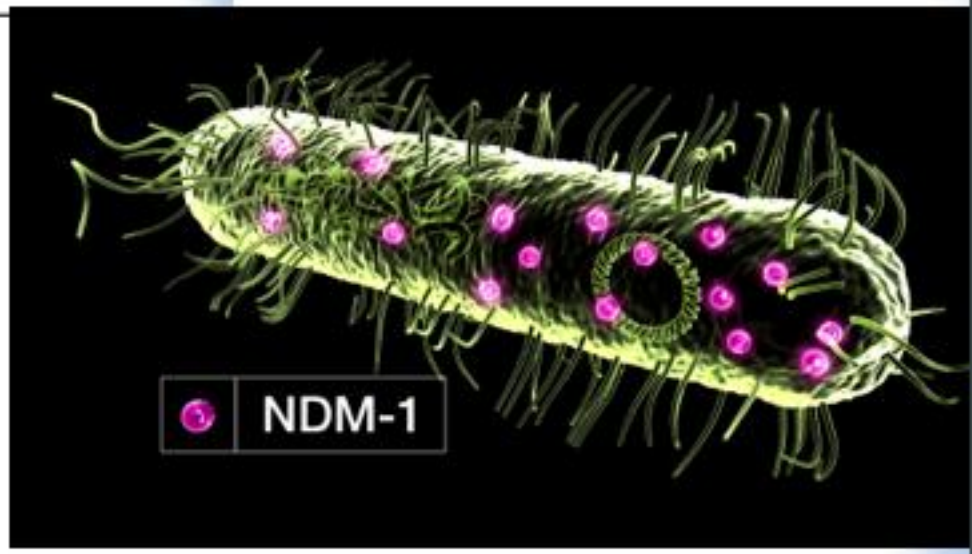
CARBAPENEM
Article
Article
Review


NDM-1

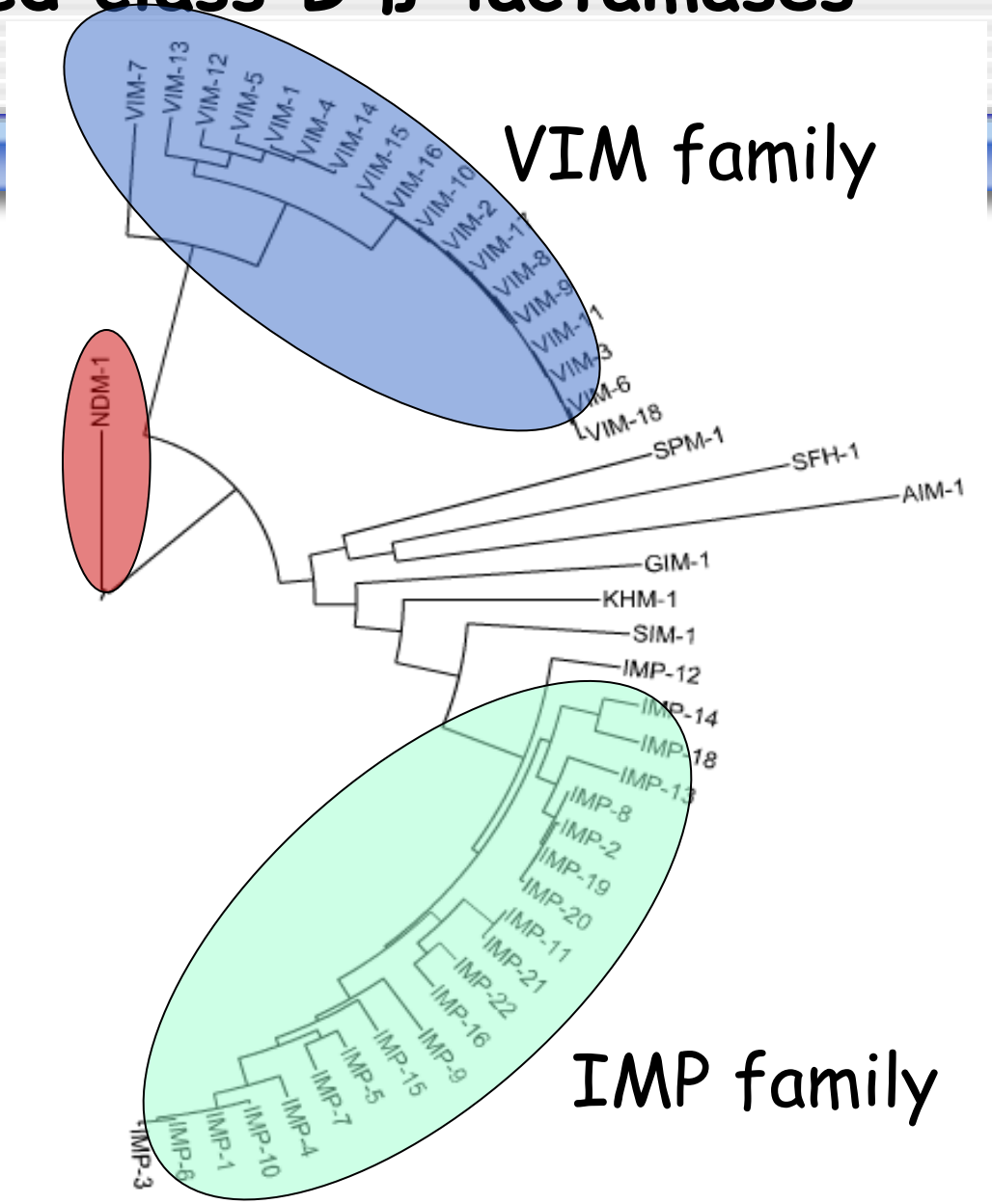
AAC,2009; 53 5046-5054

Characterization of a New Metallo-β-Lactamase Gene, *bla*$_{NDM-1}$, and a Novel Erythromycin Esterase Gene Carried on a Unique Genetic Structure in *Klebsiella pneumoniae* Sequence Type 14 from India[▽]

Dongeun Yong,[1,2] Mark A. Toleman,[2] Christian G. Giske,[3] Hyun S. Cho,[4] Kristina Sundman,[5] Kyungwon Lee,[1] and Timothy R. Walsh[2]*

Yonsei University College of Medicine, Research Institute of Antimicrobial Resistance, Seoul, Republic of Korea[1]; Department of Medical Microbiology, Cardiff University, Cardiff, United Kingdom[2]; Clinical Microbiology, MTC—Karolinska Institutet, Karolinska University Hospital, Stockholm, Sweden[3]; Yonsei University College of Life Science and Biotechnology, Seoul, Republic of Korea[4]; and Department of Clinical Microbiology, Örebro University Hospital, Örebro, Sweden[5]

Aquired class B ß-lactamases



Emergence of a new antibiotic resistance mechanism in India, Pakistan, and the UK: a molecular, biological, and epidemiological study.

Kumarasamy KK, Toleman MA, Walsh TR, Bagaria J, Butt F, Balakrishnan R, Chaudhary U, Doumith M, Giske CG, Irfan S, Krishnan P, Kumar AV, Maharjan S, Mushtaq S, Noorie T, Paterson DL, Pearson A, Perry C, Pike R, Rao B, Ray U, Sarma JB, Sharma M, Sheridan E, Thirunarayan MA, Turton J, Upadhyay S, Warner M, Welfare W, Livermore DM, Woodford N.

Department of Microbiology, Dr ALM PG IBMS, University of Madras, Chennai, India.

Abstract

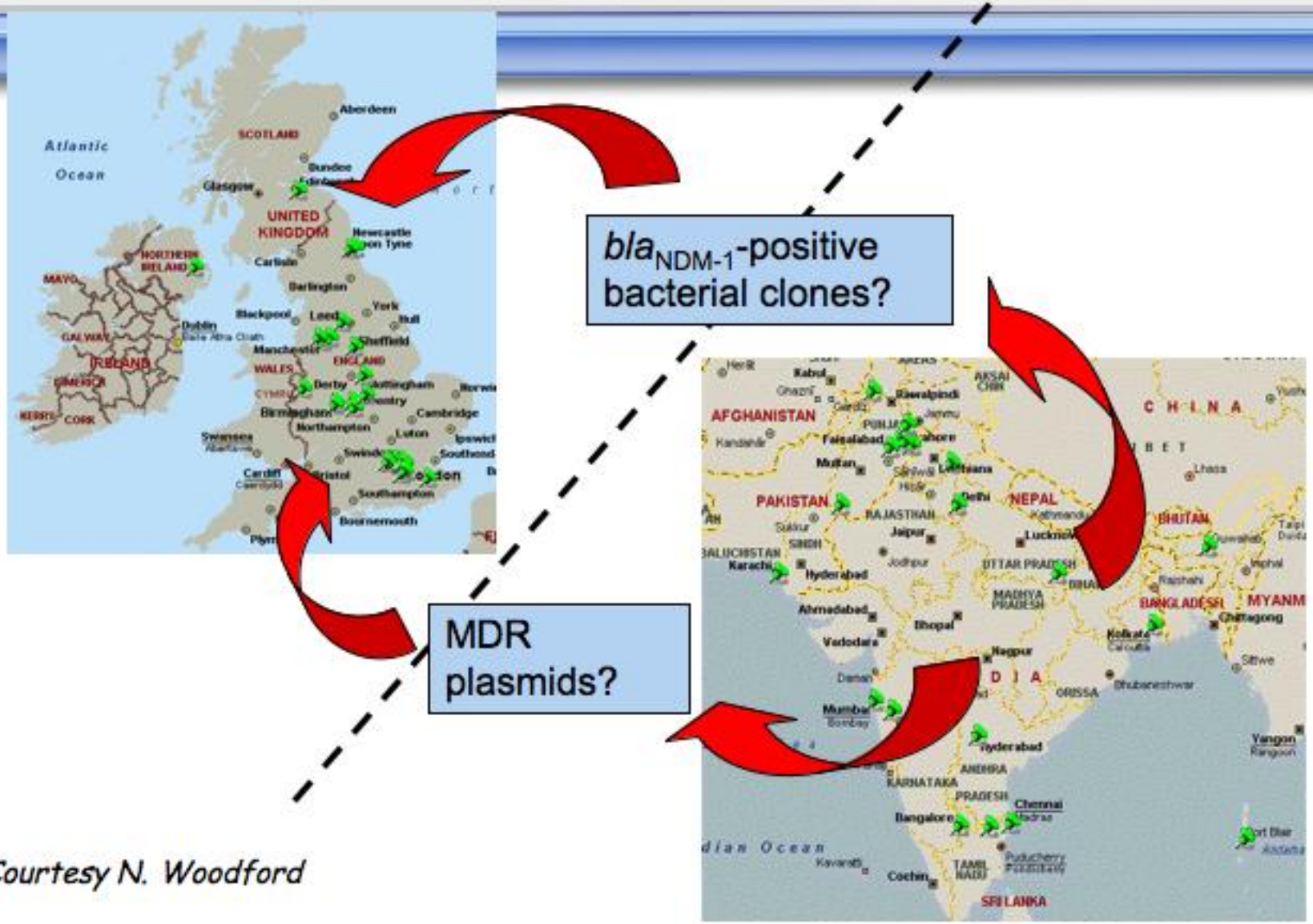
BACKGROUND: Gram-negative Enterobacteriaceae with resistance to carbapenem conferred by New Delhi metallo-beta-lactamase 1 (NDM-1) are potentially a major global health problem. We investigated the prevalence of NDM-1, in multidrug-resistant Enterobacteriaceae in India, Pakistan, and the UK.

METHODS: Enterobacteriaceae isolates were studied from two major centres in India-Chennai (south India), Haryana (north India)-and those referred to the UK's national reference laboratory. Antibiotic susceptibilities were assessed, and the presence of the carbapenem resistance gene bla(NDM-1) was established by PCR. Isolates were typed by pulsed-field gel electrophoresis of XbaI-restricted genomic DNA. Plasmids were analysed by S1 nuclease digestion and PCR typing. Case data for UK patients were reviewed for evidence of travel and recent admission to hospitals in India or Pakistan.

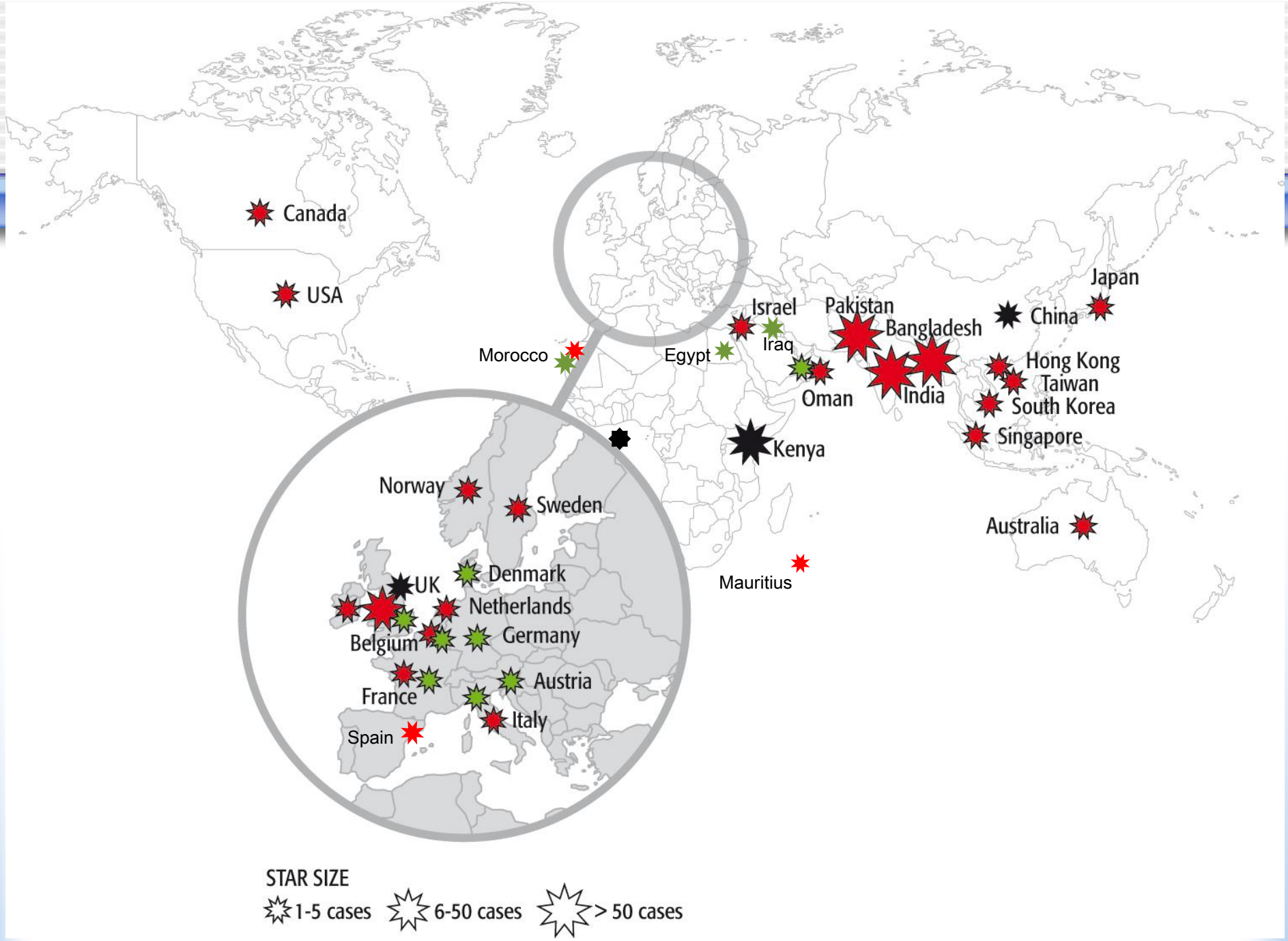
FINDINGS: We identified 44 isolates with NDM-1 in Chennai, 26 in Haryana, 37 in the UK, and 73 in other sites in India and Pakistan. NDM-1 was mostly found among Escherichia coli (36) and Klebsiella pneumoniae (111), which were highly resistant to all antibiotics except to tigecycline and colistin. K pneumoniae isolates from Haryana were clonal but NDM-1 producers from the UK and Chennai were clonally diverse. Most isolates carried the NDM-1 gene on plasmids: those from UK and Chennai were readily transferable whereas those from Haryana were not conjugative. Many of the UK NDM-1 positive patients had travelled to India or Pakistan within the past year, or had links with these countries.

INTERPRETATION: The potential of NDM-1 to be a worldwide public health problem is great, and co-ordinated international surveillance is needed.

Spread of NDM-1 from India/Pakistan to the UK

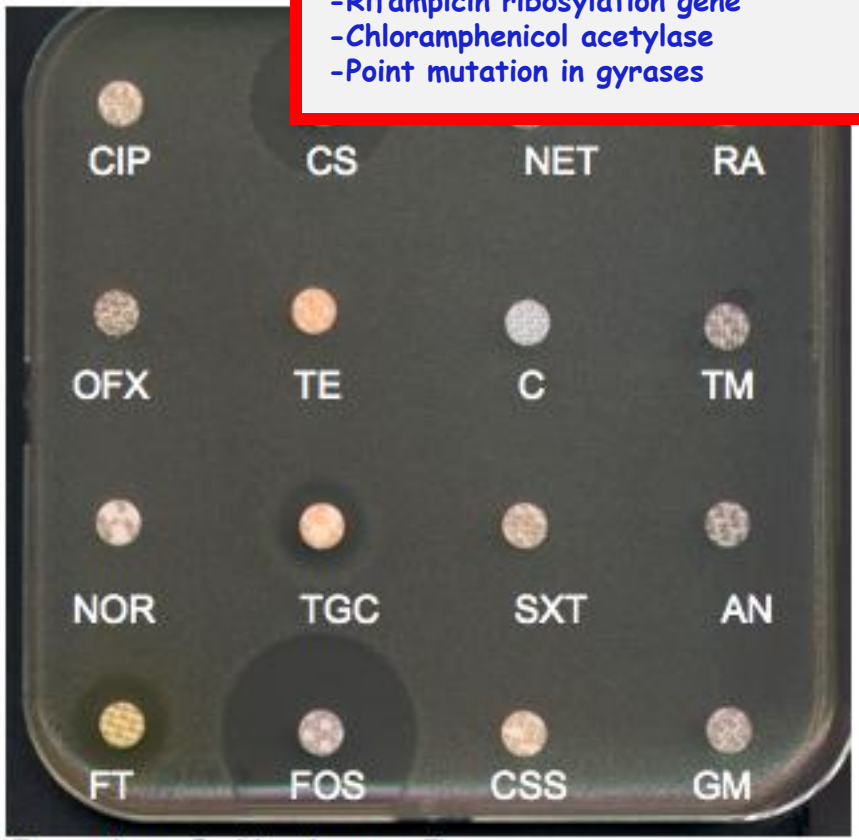


Courtesy N. Woodford


Canada
USA
Morocco
Egypt
Israel
Iraq
Pakistan
Bangladesh
India
China
Japan
Hong Kong
Taiwan
South Korea
Singapore
Oman
Kenya
Mauritius
Australia
Norway
Sweden
UK
Denmark
Netherlands
Belgium
Germany
France
Austria
Italy
Spain
STAR SIZE
1-5 cases
6-50 cases
> 50 cases

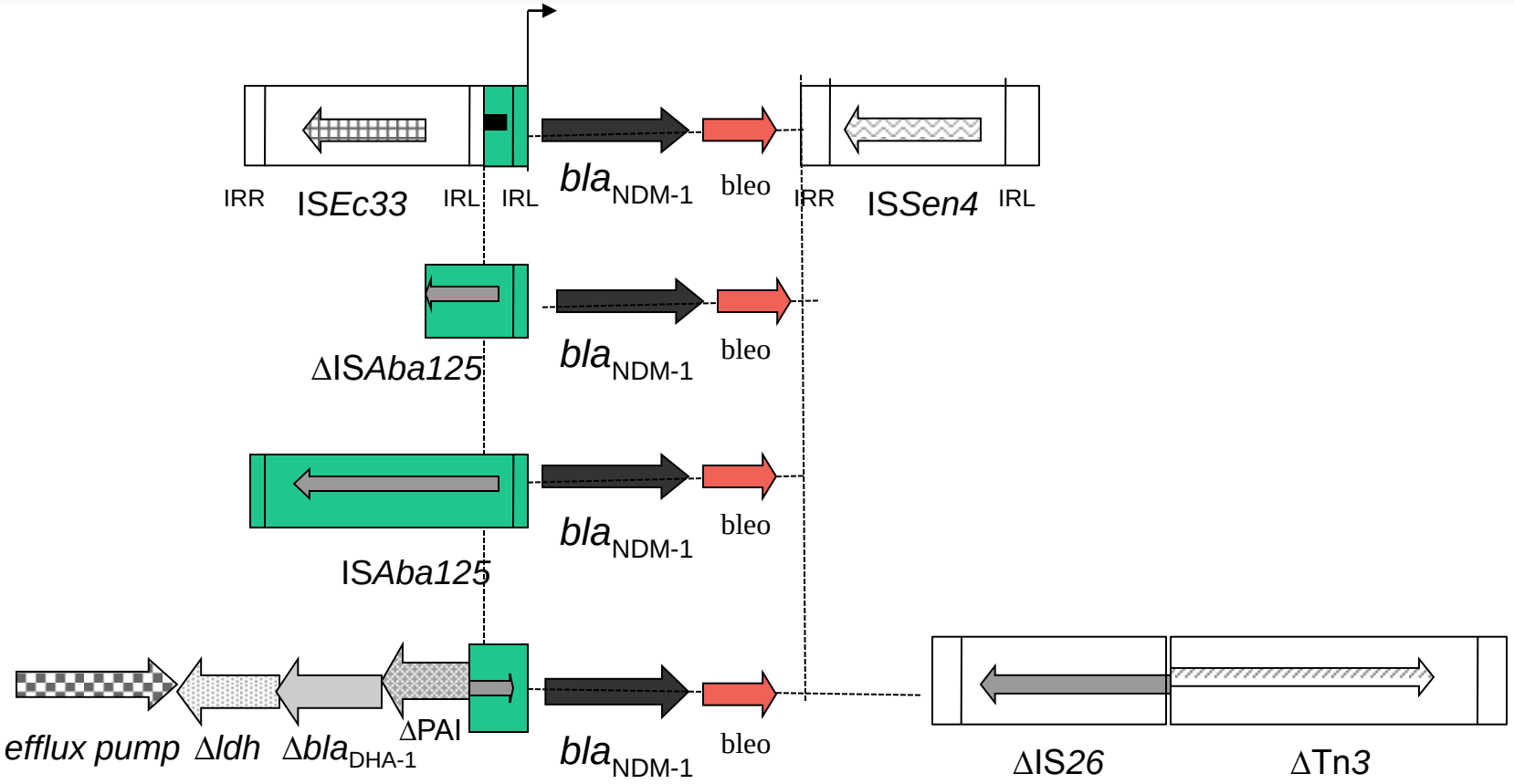
C. freundii NDM-1, France

Resistance genes including
-9 ß-lactamase genes including -3 carbapenemase gene and 1 ESBL gene
-16S RNA methylase gene
-Rifampicin ribosylation gene
-Chloramphenicol acetylase
-Point mutation in gyrases



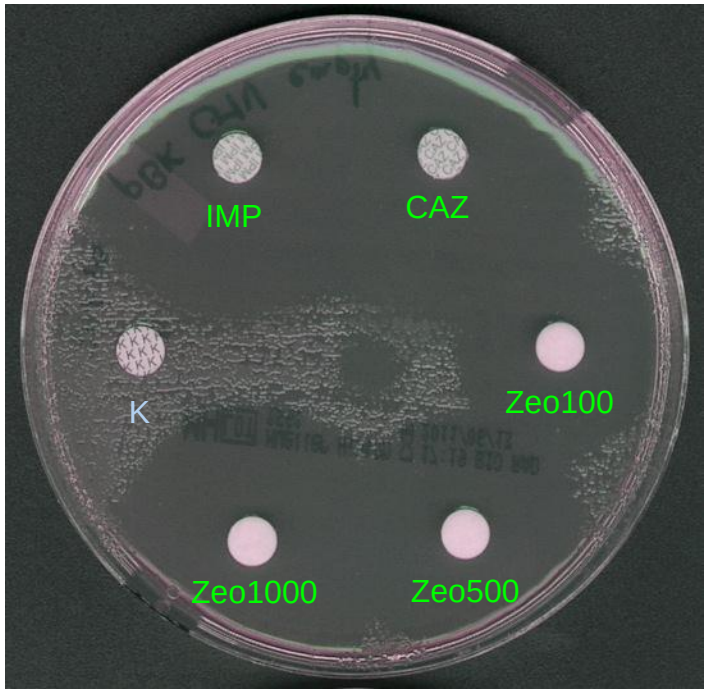
Poirel L, Ros L, Carricajo A, Berthelot P, Pozetto B, , Bernabeu S,,Nordmann P
Extremely drug-resistant C. freundii in a patient returning from India and producing NDM-1 and other Carbapenemases. Antimicrob. Agents Chemother, 2010,

bla_{NDM-1} gene environment

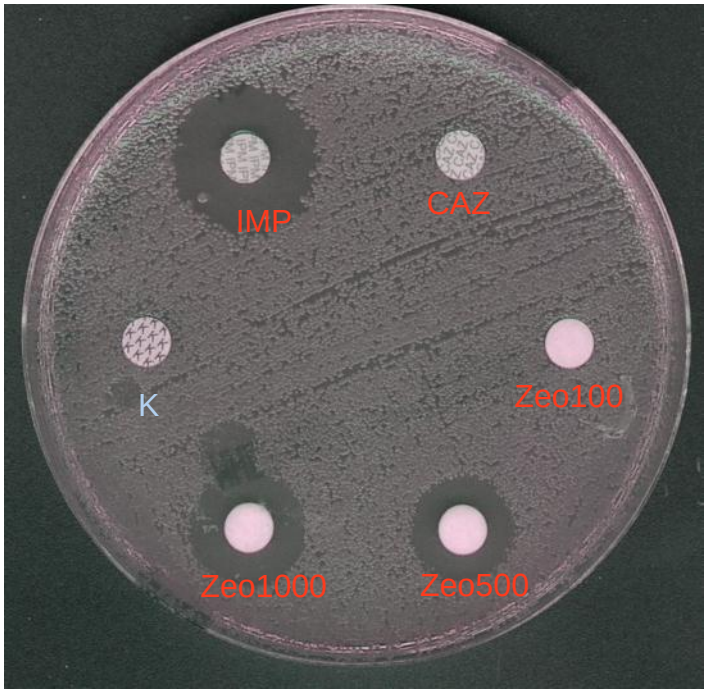


The *bla*NDM-1 and *ble*MBL gene in *E. coli*

E. coli



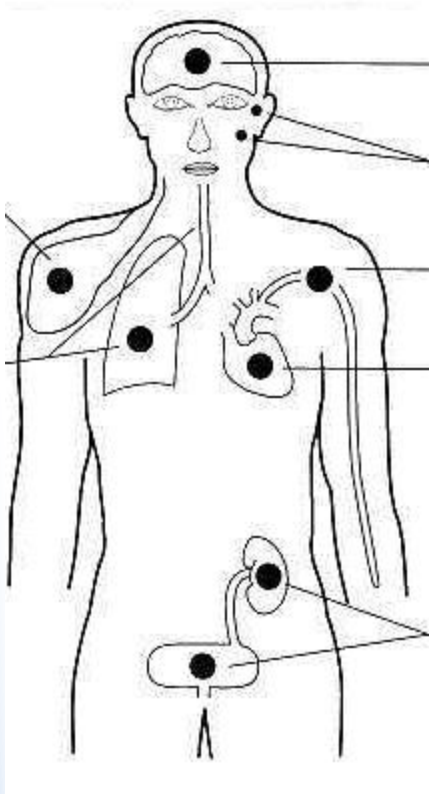
E. coli + bla_{NDM-1} + *ble*



Stabilization effect of bleomycin resistance protein

Infections with NDM producers

***E. coli, Klebsiella**, Enterobacter, Serratia, Citrobacter, Pseudomonas, Acinetobacter*



Asymptomatic colonisation

Wound infection / Diabetic foot

Lower urinary tract infection

Upper urinary tract infection

Nosocomial pneumonia / VAP

Intra-abdominal / pelvic infection

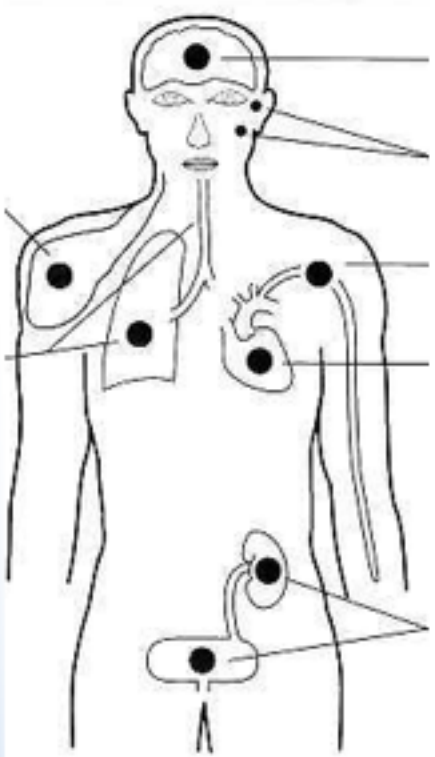
Bacteraemia / septicaemia

Neurosurgical meningitis

SEVERITY

Infections with NDM producers

E. coli, Klebsiella, *Enterobacter, Serratia, Citrobacter, Pseudomonas, Acinetobacter*



Asymptomatic colonisation

Wound infection / Diabetic foot

Lower urinary tract infection

Upper urinary tract infection

Nosocomial pneumonia / VAP

Intra-abdominal / pelvic infection

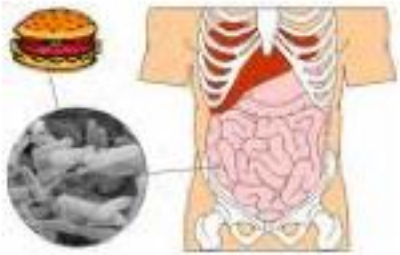
Bacteraemia / septicaemia

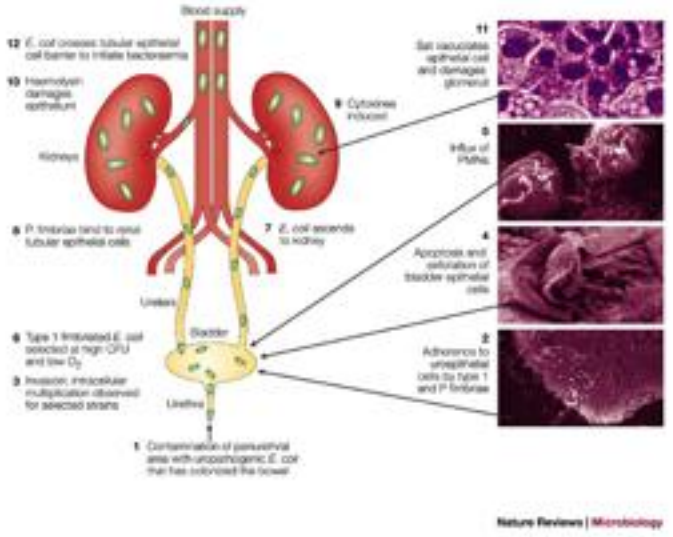
Neurosurgical meningitis

- No difference between NDM and non-NDM producers
- No known virulence factors for NDM producers
- NDM producers will not respond to conventional antibiotics !!!!

Escherichia coli

- 1st human bacterial pathogen
- 1st community-acquired pathogen
- 1st cause of urinary tract infections and diarrhea





A successful story

Hygiena

Diarrhea

Population; overcrowded

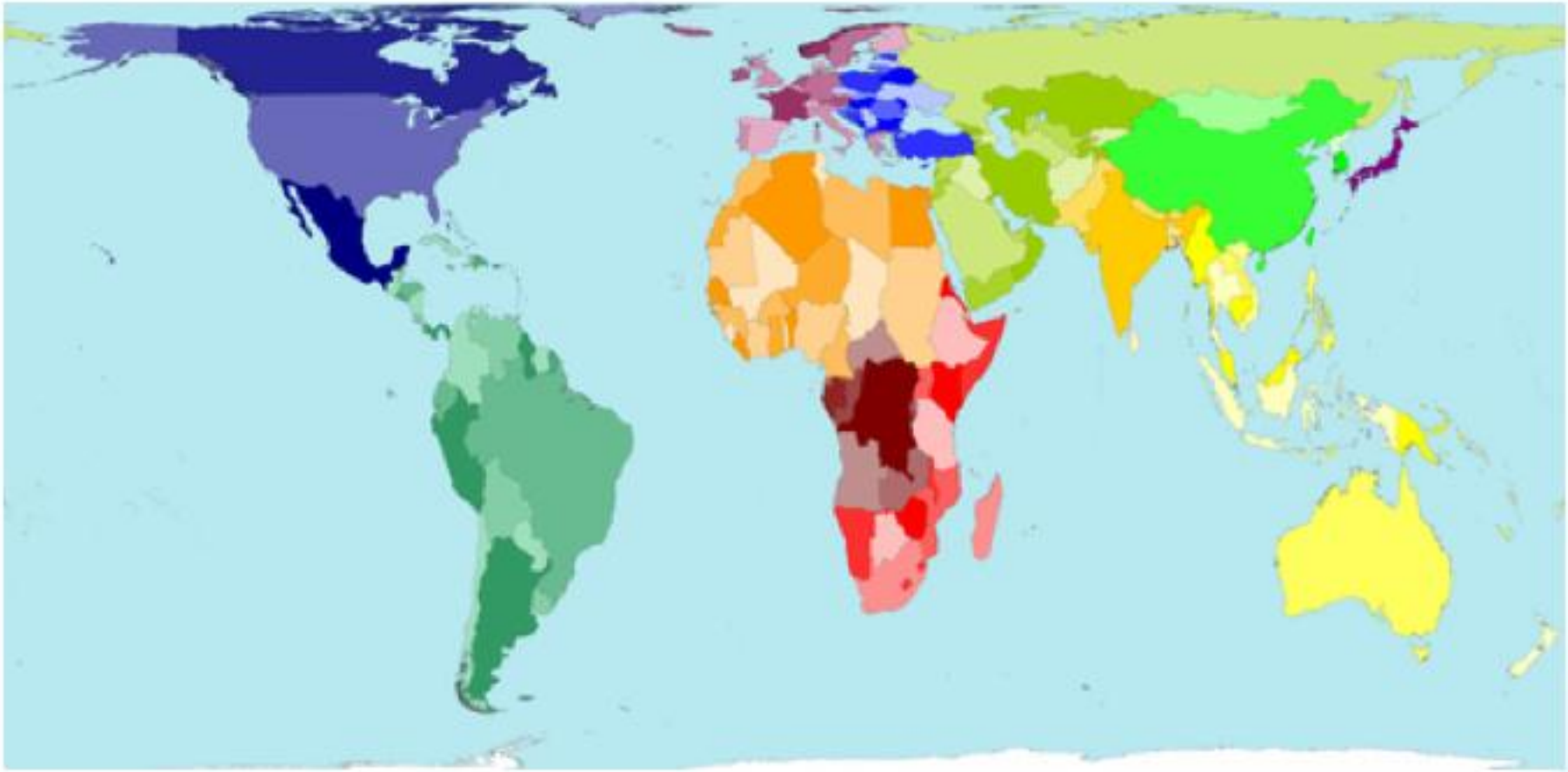
Antibiotics; misuse and overuse, over-the-counter sale

Subtropical continent

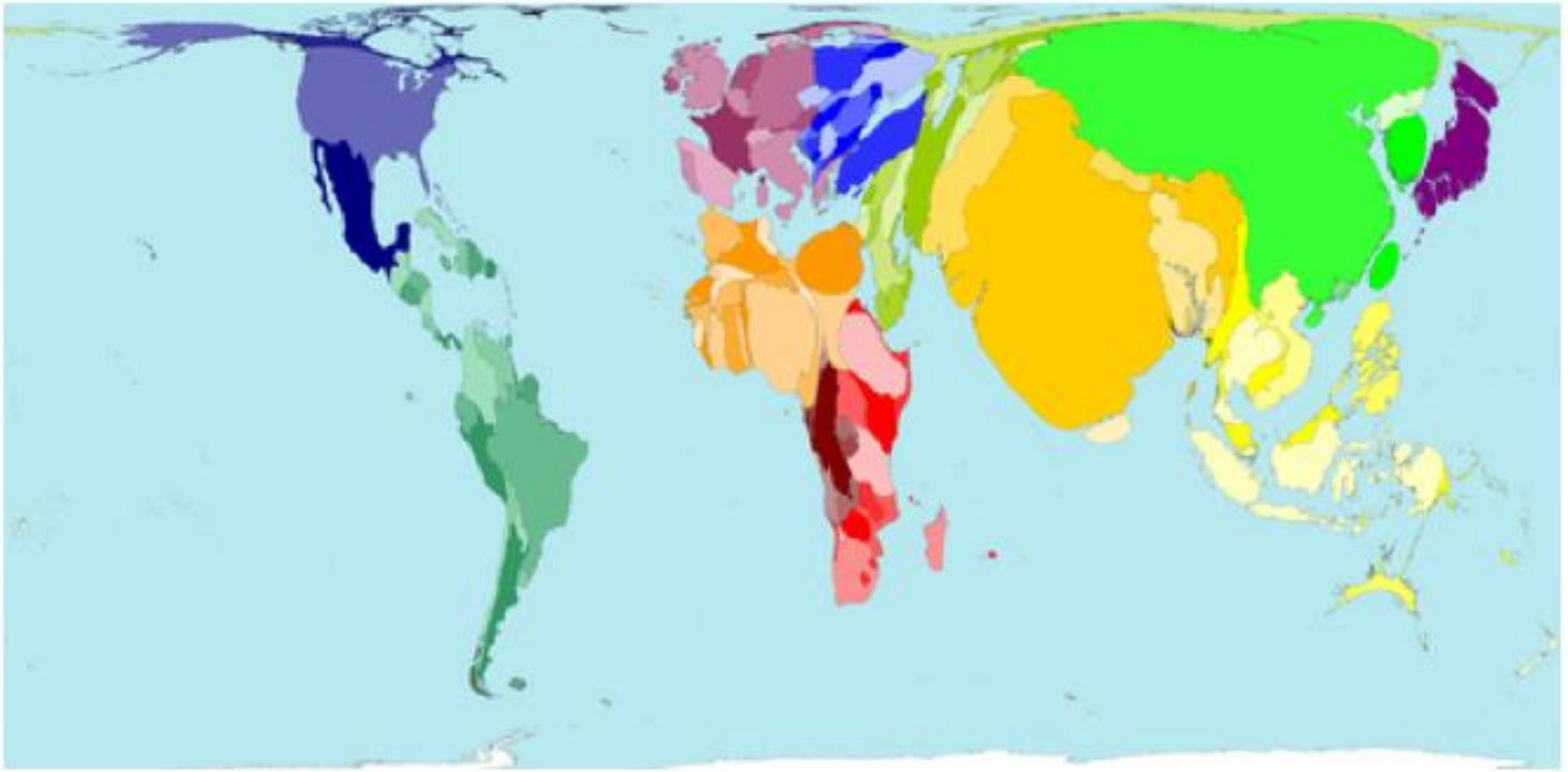
Spread of NDM-1 producers in *E. coli*, *K. pneumoniae*...

...and then higher mortality rate and length of hospitalization, overuse of broad-spectrum of antibiotics....

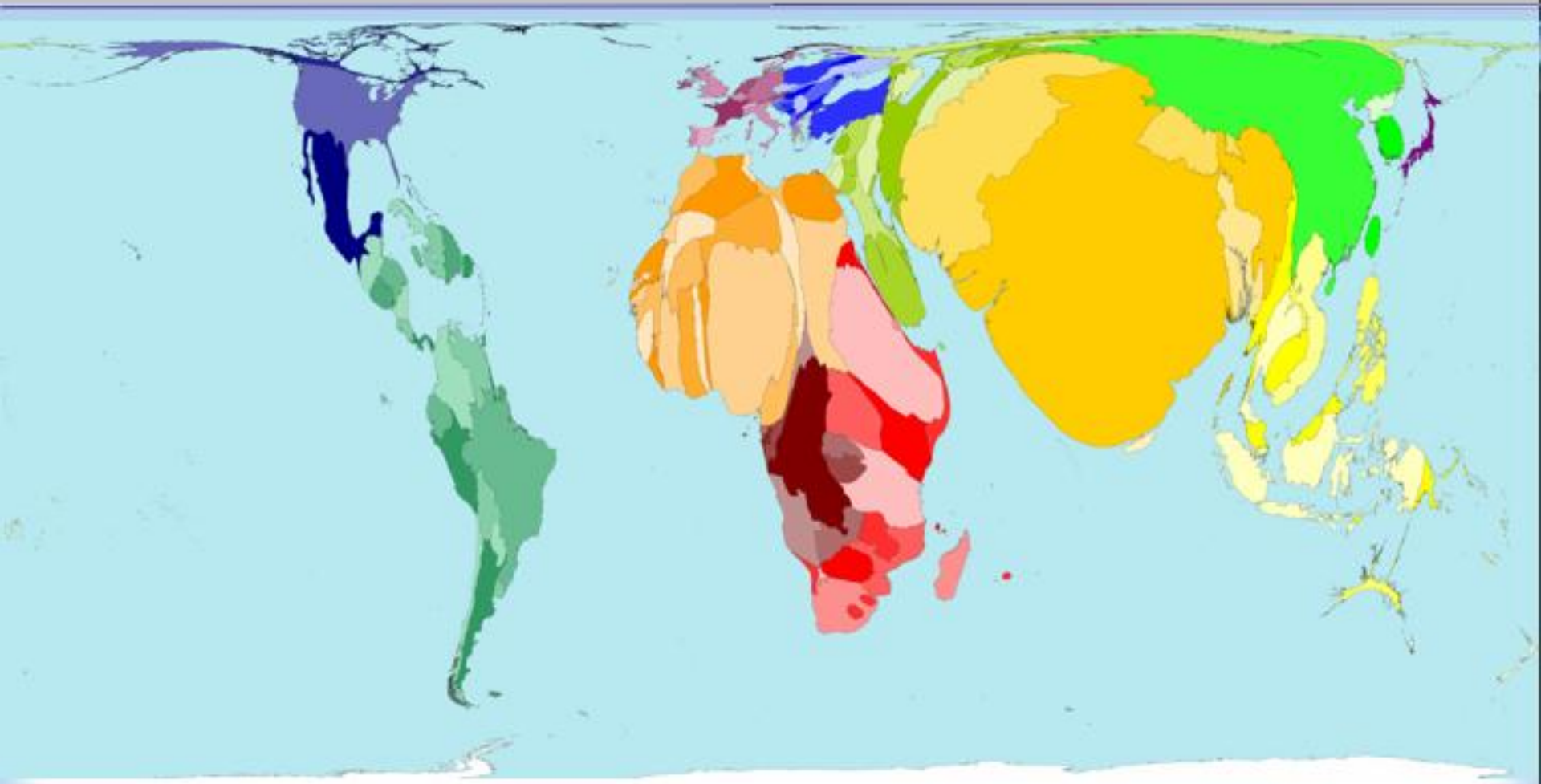
World map according to land size



World map according to population size



World map according to diarrhea



Children deaths below 10 years old

Journal of Antimicrobial Chemotherapy Advance Access published July 25, 2011

J Antimicrob Chemother
doi:10.1093/jac/dkr299

Journal of Antimicrobial Chemotherapy

Prevalence of faecal carriage of Enterobacteriaceae with NDM-1 carbapenemase at military hospitals in Pakistan, and evaluation of two chromogenic media

John D. Perry[1]*, Sakeenah Hussain Naqvi[2], Irfan Ali Mirza[2], Shehla Ambreen Alizai[2], Aamir Hussain[2], Sandrine Ghirardi[3], Sylvain Orenga[3], Kathryn Wilkinson[1], Neil Woodford[4], Jiancheng Zhang[4], David M. Livermore[4], Shahid Ahmad Abbasi[2] and Muhammad W. Raza[1]

[1]*Microbiology Department, Freeman Hospital, Newcastle upon Tyne NE7 7DN, UK;* [2]*Microbiology Department, Armed Forces Institute of Pathology (AFIP), Rawalpindi, Pakistan;* [3]*bioMérieux, 38390 La Balme-les-Grottes, France;* [4]*Antibiotic Resistance Monitoring & Reference Laboratory (ARMRL), HPA Microbiology Services—Colindale, 61 Colindale Avenue, London NW9 5EQ, UK*

*Corresponding author. Tel: +44-191-2231226; Fax: +44-191-2231224; E-mail: johnperry@nuth.nhs.uk

Received 21 April 2011; returned 8 June 2011; revised 14 June 2011; accepted 23 June 2011

Objectives: To determine the prevalence and antimicrobial susceptibility of carbapenemase-producing Enterobacteriaceae among hospitalized patients and outpatients attending two military hospitals in Rawalpindi, Pakistan, and to compare the performance of two chromogenic culture media for the isolation of these organisms.

Methods: Stool samples from 200 distinct patients were cultured on MacConkey agar and subsequently on two chromogenic media—Colorex KPC and a prototype chromogenic medium, ID Carba—designed for the isolation of carbapenemase-producing Enterobacteriaceae. All Gram-negative isolates growing on either chromogenic medium were investigated for carbapenemases by phenotypic and molecular methods. Producers were subjected to susceptibility testing with 40 antimicrobials by VITEK 2 or agar dilution.

Results: In total, 64 NDM-1-positive isolates of Enterobacteriaceae, belonging to seven distinct species, were recovered from 37 (18.5%) of the stool samples. No other carbapenemase types were confirmed. Nineteen positive samples were identified among 70 from inpatients (prevalence 27.1%) and there were 18 positive samples among 130 from outpatients (prevalence 13.8%). Fifty-six isolates (87.5%) harbouring the NDM-1 enzyme were recovered on ID Carba compared with 41 isolates (64.1%) on Colorex KPC ($P=0.012$). Multidrug resistance was prevalent, but no pan-resistant isolates were found, with most isolates susceptible *in vitro* to colistin (97%), mecillinam (95%), fosfomycin (94%), tigecycline (89%) and nitrofurantoin (78%).

Conclusions: This study shows a high prevalence of multidrug-resistant Enterobacteriaceae with the NDM-1 enzyme in Rawalpindi. The new chromogenic medium, ID Carba, was more sensitive than Colorex KPC and has potential as a screening medium for isolation of Enterobacteriaceae harbouring the NDM-1 enzyme.

Keywords: β-lactamases, antimicrobial resistance mechanisms, *Escherichia coli*

E. coli NDM-1: community-acquired !

3 Institut Scientifique de la Santé Publique. Guidelines for control of infections in case of cross-border transfer of patients hospitalized in countries with high endemicity of carbapenemase-producing Enterobacteriaceae. Surveillance de germes multi-résistants dans les hôpitaux belges. Sept 23, 2010. http://www.nsih.be/download/MDR/NewDelhi_fiche_alert_VS_FR.pdf (in French) (accessed Nov 8, 2010).

The plasmid-mediated bla_{NDM-1} gene that encodes a powerful carbapenemase was first identified in *Escherichia coli* and in *Klebsiella pneumoniae* in Sweden from a patient who was transferred from India.[1] It was then identified from many patients in the UK, India, and Pakistan in different enterobacterial species.[2] Here we report a woman aged 60 years who was admitted to hospital in April, 2009, for treatment of a breast cancer.

The patient came from Darjeeling, India, where she had lived for several years and had never been hospitalised. Upon her admission in France, bacterial cultures from the surface of her breast tumour were grown. The cultures were of the *E coli* isolate GUE that was resistant to most β-lactams (remaining susceptible to aztreonam) and that had reduced susceptibility to carbapenems (minimum inhibitory concentrations of imipenem 3 µg/mL, ertapenem 3 µg/mL, and meropenem 2 µg/mL).[3] This isolate was also resistant to gentamicin, kanamycin, tobramycin, sulfonamides, tetracycline, and fluoroquinolones, but remained susceptible to amikacin, chloramphenicol, rifampicin, and colistin. PCR and sequencing revealed that *E coli* GUE harboured the bla_{NDM-1} gene. Mating-out assays[4] allowed the bla_{NDM-1} gene to be identified on a 110 kb plasmid, with markers for kanamycin, gentamicin, tobramycin, trimethoprim, and sulfonamide resistance. Multilocus sequence typing[5] identified *E coli* GUE as an ST131-type strain, which corresponds to a genetic background that is also responsible for the worldwide diffusion of another common resistance determinant, CTX-M-15.

This case is the first identification of an NDM-1-producing *E coli* isolate in France, and corresponds again to an imported case from India. This example confirms the recent data suggesting that the Indian subcontinent might represent an important reservoir, and therefore a source, of NDM-producing isolates. The patient had not been hospitalised in India; therefore, the multidrug-resistant isolate had likely been community acquired. Worringly, this resistance gene has been identified here in an *E coli* strain belonging to a genotype that has proved its ability to disseminate widely in the community.

We declare that we have no conflicts of interest. This study was mostly funded by the INSERM (U914), France, and by grants from the Ministère de l'Education Nationale et de la Recherche (UPRES-EA3539), Université Paris XI, France, and from the European Community (TEMPOtest-QC, HEALTH-2009-241742).

*Laurent Poirel, Cécile Hombrouck-Alet, Claire Freneaux, Sandrine Bernabeu, *Patrice Nordmann*
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Hôpital de Bicêtre, Department of Bacteriology-Virology, Le Kremlin Bicêtre, Paris 94275, France

1 Yong D, Toleman MA, Giske CG, et al. Characterization of a new metallo-β-lactamase gene, bla_{NDM-1}, and a novel erythromycin esterase gene carried on a unique genetic structure in *Klebsiella pneumoniae* sequence type 14 from India. *Antimicrob Agents Chemother* 2009; 53: 5046–54.

2 Kumarasamy KK, Toleman MA, Walsh TR, et al. Emergence of a new antibiotic resistance mechanism in India, Pakistan, and the UK: a molecular, biological, and epidemiological study. *Lancet Infect Dis* 2010; 10: 597–602.

3 Clinical and Laboratory Standards Institute. 2010. Performance standards for antimicrobial susceptibility testing; twentieth informational supplement. Jan, 2010. http://www.clsi.org/source/orders/free/m100-s20.pdf (accessed Nov 6, 2010).

4 Rodriguez-Martinez JM, Nordmann P, Fortineau N, Poirel L. VIM-19, a metallo-β-lactamase with increased carbapenemase activity from *Escherichia coli* and *Klebsiella pneumoniae*. *Antimicrob Agents Chemother* 2010; 54: 471–76.

5 Tartof SY, Solberg OD, Manges AR, Riley LW. Analysis of a uropathogenic *Escherichia coli* clonal group by multilocus sequence typing. *J Clin Microbiol* 2005; 43: 5860–64.

Clinical Infectious Diseases Advance Access published September 29, 2011

0 MONTH

Correspondence

Emergence of an Autochthonous and Community-Acquired NDM-1–Producing *Klebsiella pneumoniae* in Europe

To the Editor—The recently identified carbapenemase New Dehli metallo-β-lactamase (NDM-1) inactivates all β-lactams except aztreonam [1]. The corresponding gene that is usually plasmid-borne has spread mostly in *Escherichia coli* and *Klebsiella pneumoniae* [1, 2]. NDM-1 producers are multidrug resistant or even resistant to all antibiotics [1, 2]. Whereas contamination with NDM-1 producers is mostly hospital associated, rare cases of community acquisition are known and have been traced to the Indian subcontinent [2].

Here, we report a woman aged 83 years who had cystitis due to a multidrug-resistant *K. pneumoniae* in June 2011. She had a history of multiple and recurrent episodes of urinary tract infections caused by diverse Enterobacteriaceae that were always treated with narrow-spectrum antibiotics. Because the patient's symptoms tended to to disappear spontaneously and rapidly, the latest cystitis episode had not been treated.

K. pneumoniae EDU was resistant to all β-lactams, including carbapenems, as detected with a Vitek-2 automated suscep-

Polymerase chain reaction, sequencing, and plasmid analysis, performed as described elsewhere [5], revealed that *K. pneumoniae* EDU harbored the bla_{NDM-1} carbapenemase gene and the $bla_{CTX-M-15}$ extended-spectrum ß-lactamase gene, which were located on 2 different plasmids (both being approximately 150 kb in size). The isolate coexpressed the CMY-2 cephalosporinase gene, which was located on the bla_{NDM-1} plasmid. In addition, it possessed the *qnrB* gene encoding resistance to quinolones and the bla_{OXA-1} gene encoding a restricted-spectrum oxacillinase, both genes being located on the $bla_{CTX-M-15}$ plasmid. Both plasmids were self-transferable by conjugation, and the bla_{NDM-1} plasmid was found to be of the IncA/C broad-host range type [6]. Multilocus sequence typing [7] results showed that *K. pneumoniae* EDU belonged to the sequence type 1, whereas previously reported NDM-1–positive *K. pneumoniae* isolates were of other sequence types (eg, ST14 and ST147) [6].

Neither this patient nor her husband had traveled to any country in the previous 3 years, including countries with a high prevalence of NDM-1 producers (India, Pakistan, Bangladesh, United Kingdom, Balkan states, and Middle Eastern nations) of NDM-1 producers outside its main reservoir (Indian subcontinent). The source of contamination remains unknown but may be difficult to find, because persistence of NDM-1 producers in human flora has been evidenced to be >1 year [9].

This present report may indicate the ongoing spread of NDM producers in the community worldwide. A nightmare perspective could be its spread similar to that reported for extended-spectrum β-lactamases of the CTX-M-type, which are now uncontrolled.

Notes

Financial support. This work was supported by INSERM (U914), France, and by grants from the Ministère de l'Education Nationale et de la Recherche (UPRES-EA3539), Université Paris XI, France, and from the European Community (TEMPOtest-QC, HEALTH-2009-241742).

Potential conflicts of interest. All authors: No reported conflicts.

All authors have submitted the ICMJE Form for Disclosure of Potential Conflicts of Interest. Conflicts that the editors consider relevant to the content of the manuscript have been disclosed.

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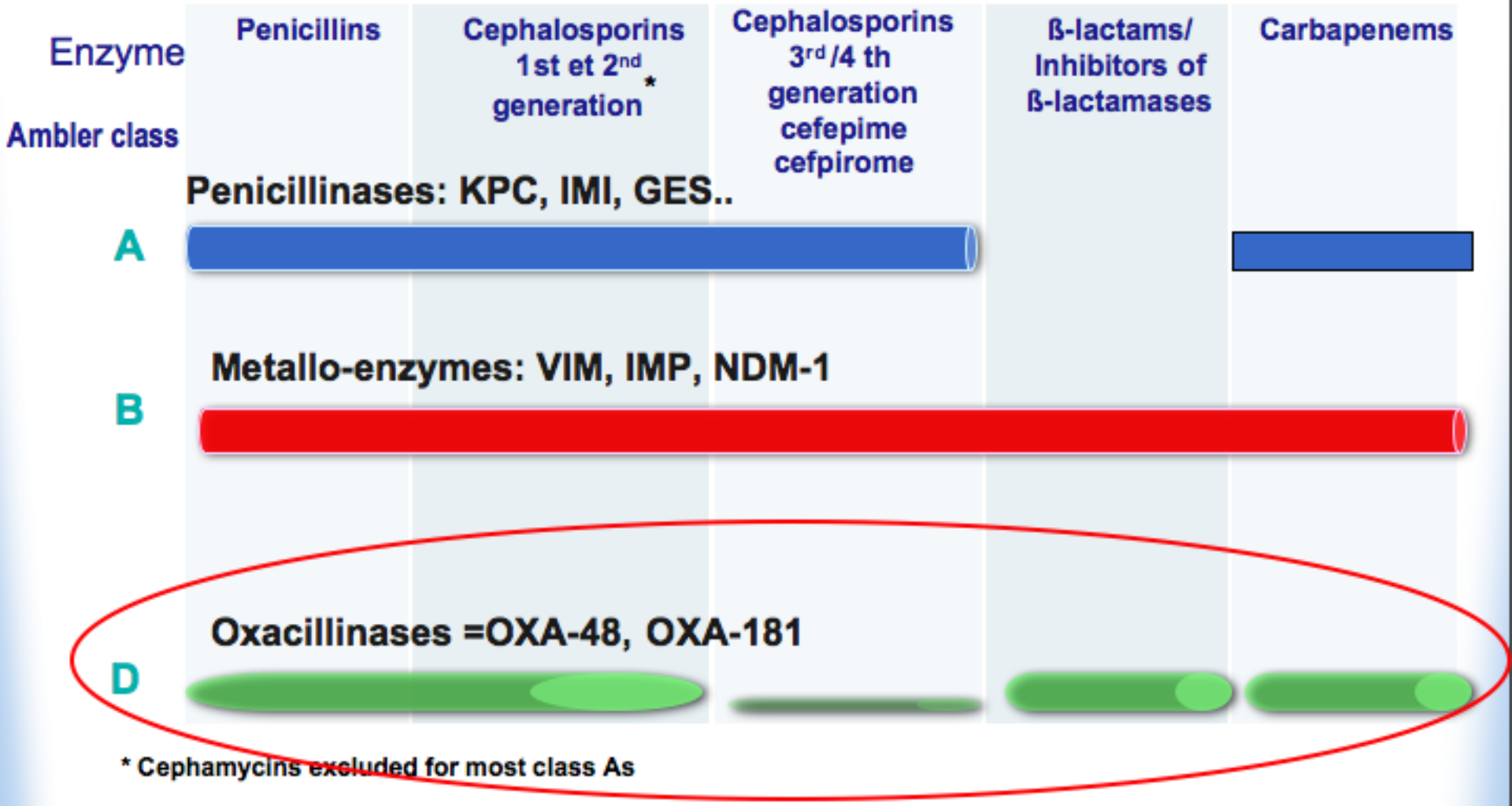
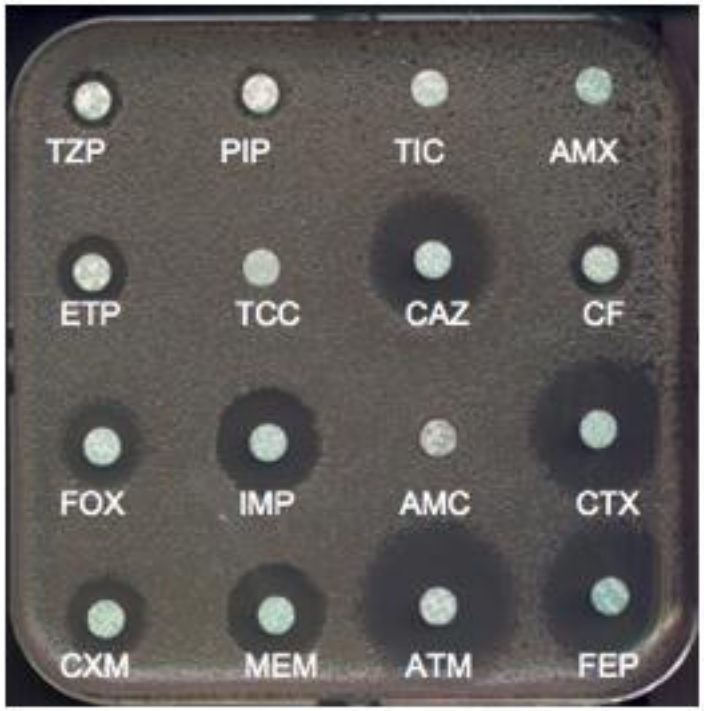
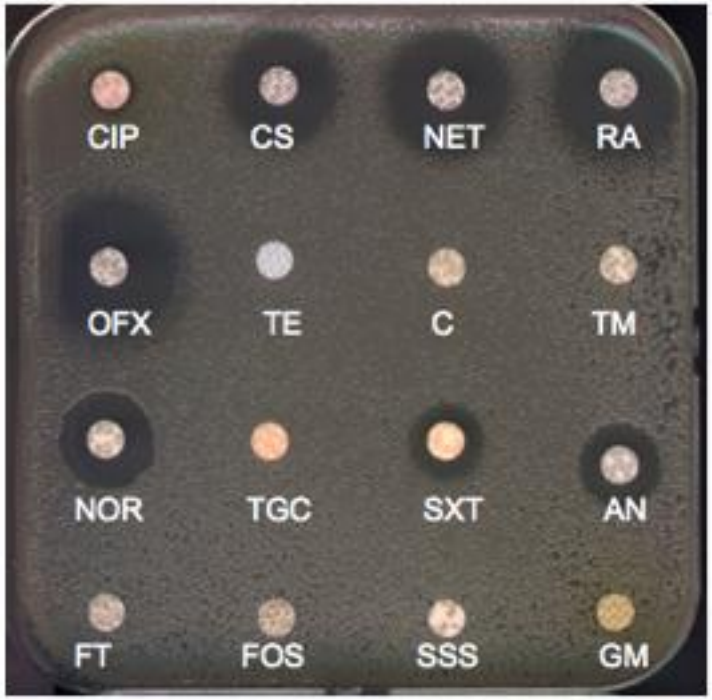
The carbapenemases in *Enterobacteriaceae*



* Cephamycins excluded for most class As

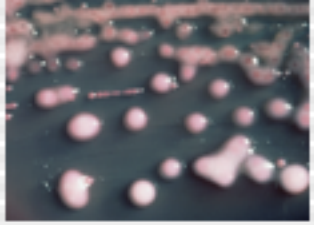
OXA-48





Emergence of oxacillinase-mediated resistance to Imipenem in Klebsiella pneumoniae
Poirel L, Héritier , Nordmann P. Tolün, AAC 2004

OXA-48 + CTX-M-15



India
(OXA-181/OXA-48)

The Netherlands
Germany
Italy
Belgium
Turkey
UK
Lebanon
Switzerland
Israel
France
Egypt
Spain
Morocco
Algeria
Tunisia
Senegal

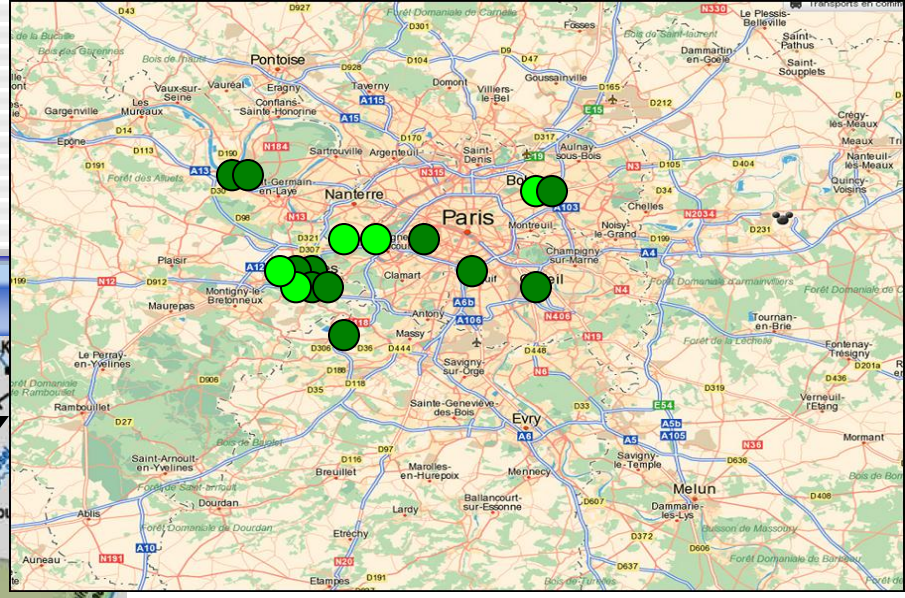
Combating the Superbug Klebsiella Oxa-48 Outbreak in a Dutch Hospital 15 August 2011

Over 80 patients have been infected with at Maasstad hospital, Rotterdam in the Netherlands due to the outbreak of a multiple antibiotic-resistant bacterium named Klebsiella pneumoniae Oxa-48. The hospital has estimated that over 2000 people were at risk of being infected.

- Single OXA-48-producing isolates
- Outbreaks of OXA-48-producing isolates
- Nationwide distribution of OXA-48-producing isolates

EID, Nordmann et al. 2011

OXA-48



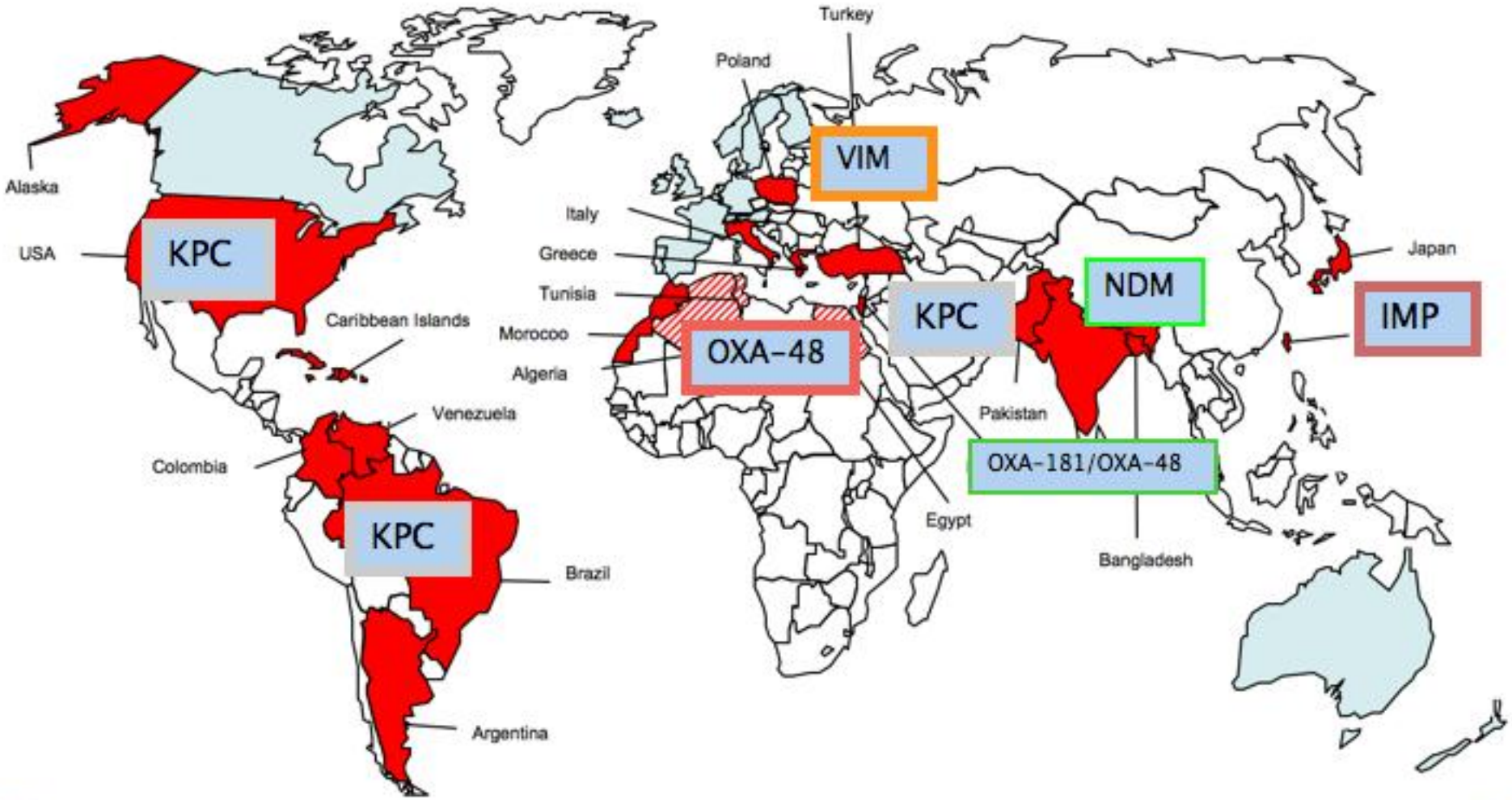
● *K.pneumoniae*

● *E.coli*

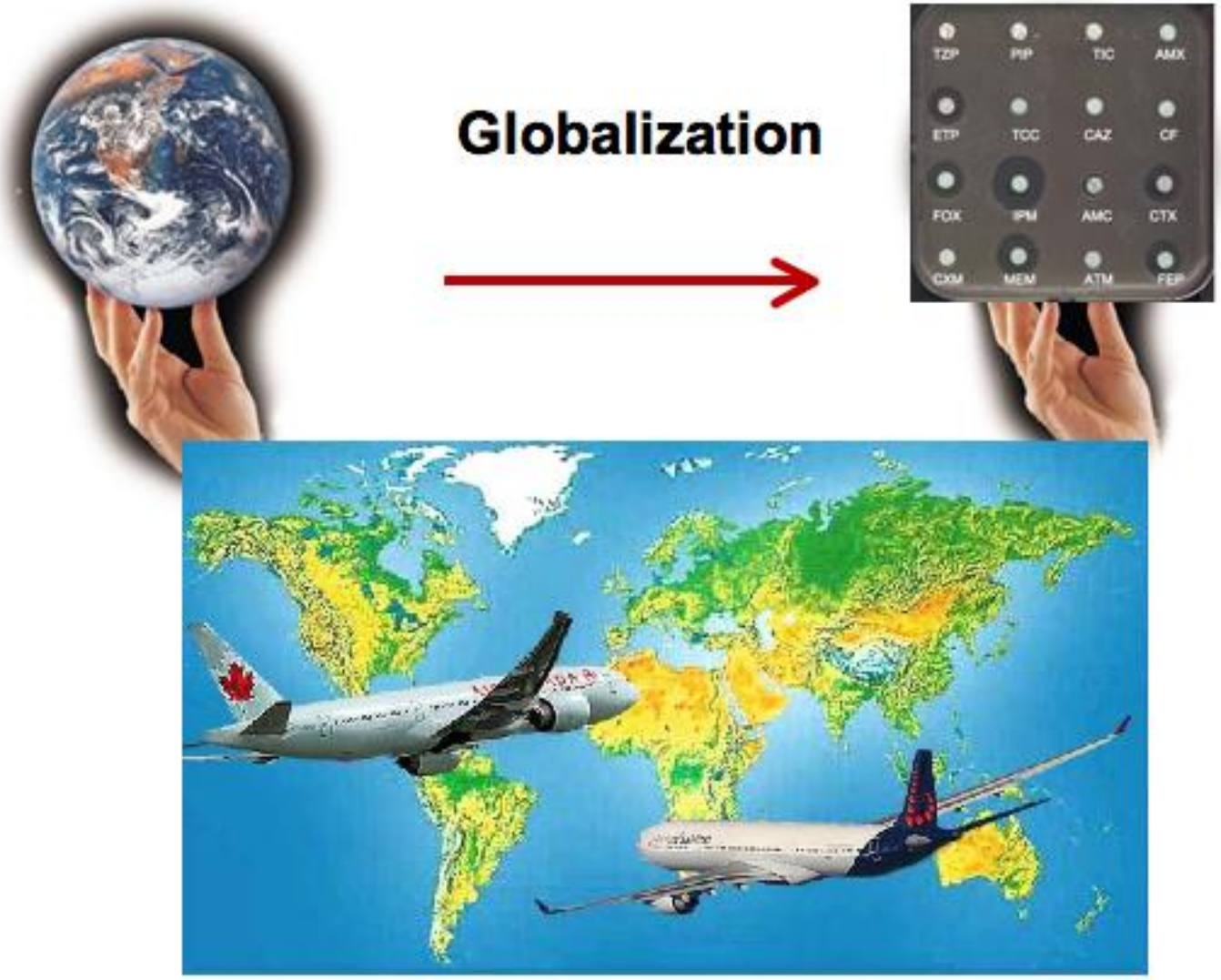
***E.coli* OXA-48 : 7**

***K.pneumoniae* OXA-48 : 20**

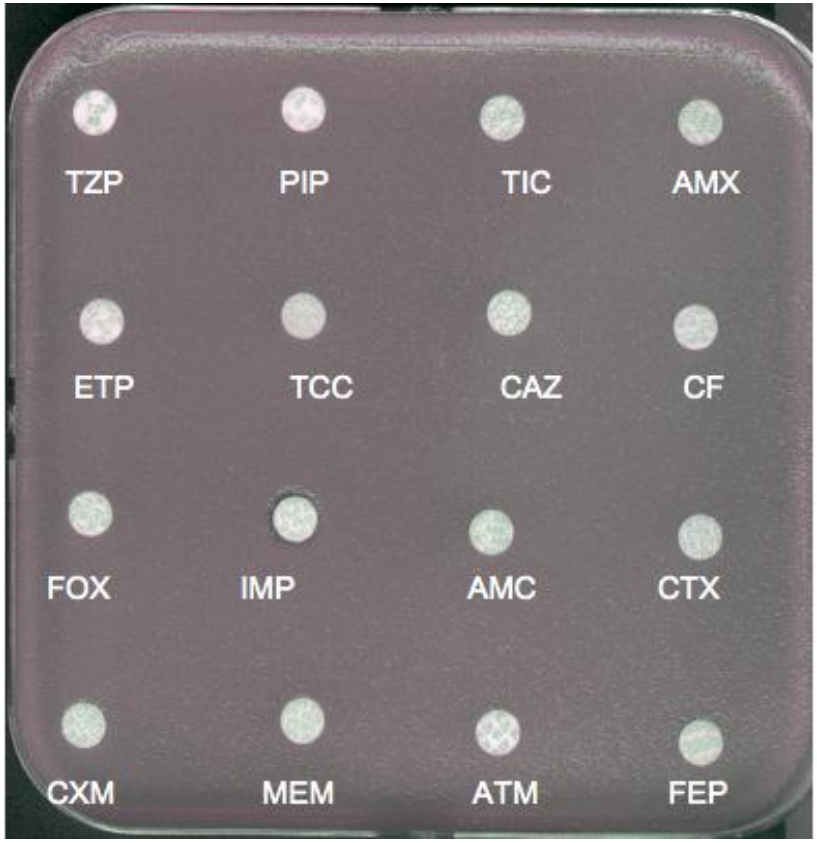
Carbapenemases/Enterobactericeae reservoirs



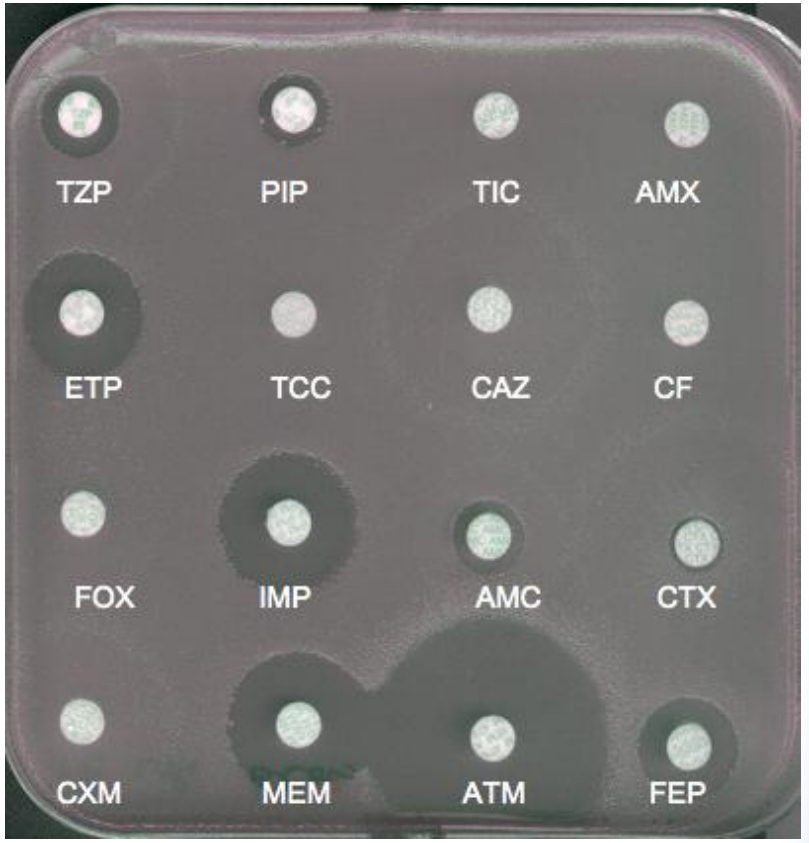
MDR Gram negatives: transfer 962 millions -2010



Susceptibilitiy testing



K. pneumoniae NDM-1

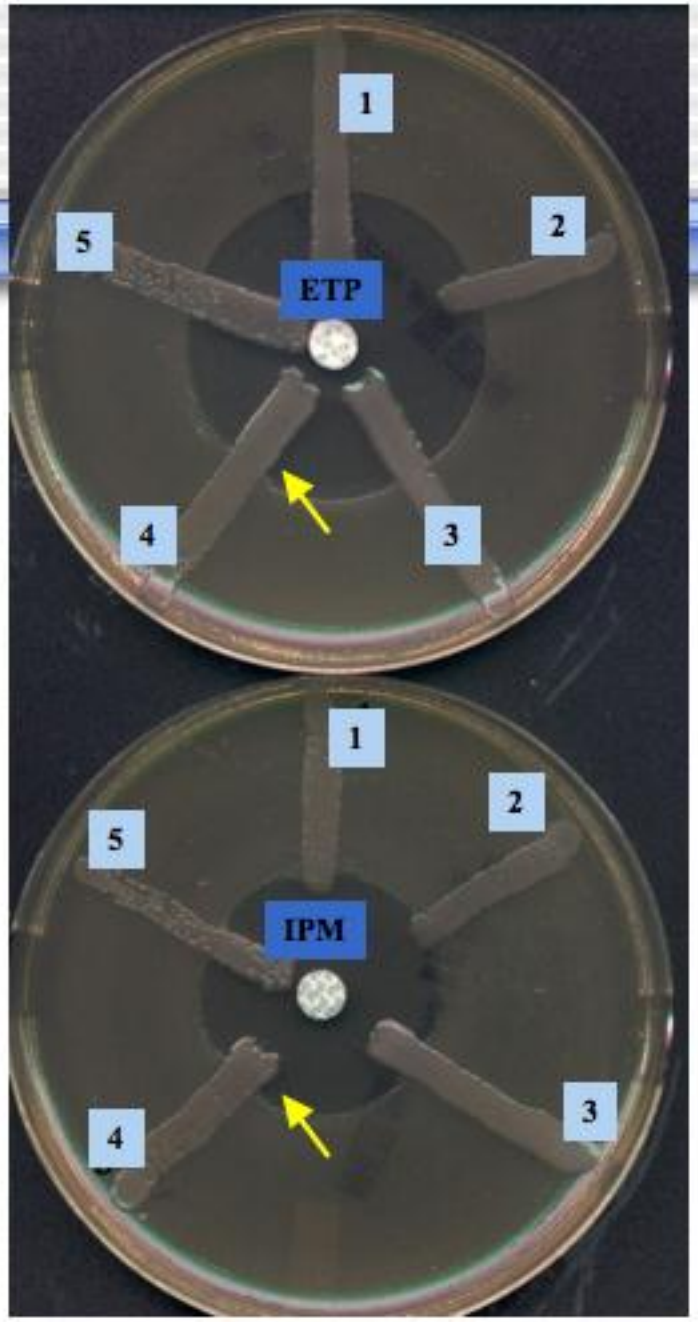


E. coli NDM-1

Heterogeneous expression of metallo-carbapenemases in *Enterobacteriaceae*

		MIC (mg/L) imipenem	MIC (mg/L) meropenem
S. marcescens	IMP-type	32 - >128	128 - >128
	VIM-type	64	64
K. pneumoniae	IMP-type	0.25 - >128	0.25 - 64
	VIM-type	2	0.5
	NDM-type	0.75->32	2->32
Enterobacter *Citrobacter* *E. coli* *S. flexneri*	IMP-type	0.25 - 8	0.25 - 8
	VIM-type	1 - 8	0.5 - 4
	NDM-type	1.5->32	2->32
CLSI		≤ 1 / ≥4	≤ 1/ ≥4
EUCAST		≤ 2/ >8	≤ 2/ >8

Hodge test



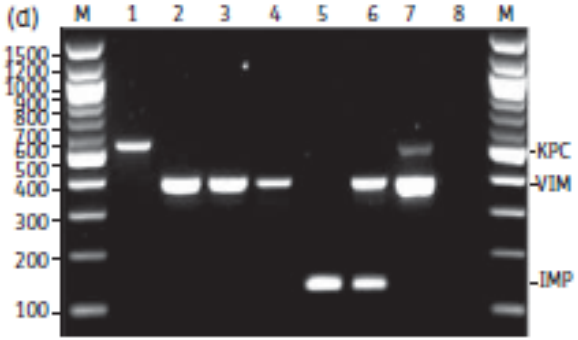
1 : *K pneumoniae* NDM-1
2 : *K pneumoniae* NDM-1
3 : *K pneumoniae* NDM-1
4 : *K pneumoniae* KPC-2
5 : *E. coli* CTX-M-15

False positive; *A. baumanniii*
Enterobacter sp…

Hodge test

	MIC (µg/ml) ETP	Hodge test	
OXA-48			
E. coli (n=4)	0.5 - 6	+	
K. pneumoniae (n=4)	2 ® 16	+	
KPC			
E. coli (n=3)	0.5 - 16	+	
K. pneumoniae (n=2)	4 - 12	+	
NDM-1			
E. coli (n=6)	3 - 32	+ (n=1)	- (n=5)
K. pneumoniae (n=6)	6 ® 32	+ (n=4)	- (n=2)

Molecular tests - PCR- Sequencing; Gold Standard



Multiplex PCR for detection of acquired carbapenemase genes

Laurent Poirel[a,*], Timothy R. Walsh[b], Vincent Cuvillier[a], Patrice Nordmann[a]

[a]*Service de Bactériologie-Virologie, INSERM U914 "Emerging Resistance to Antibiotics", Hôpital de Bicêtre, Assistance Publique/Hôpitaux de Paris, Faculté de Médecine Paris-Sud, Université Paris XI, 94275 K.-Bicêtre, France*

[b]*Department of Medical Microbiology, School of Medicine, Cardiff University, UK*

Received 20 September 2010; accepted 1 December 2010

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Diagnostic Microbiology and Infectious Disease xx (2011) xxx–xxx

DIAGNOSTIC MICROBIOLOGY AND INFECTIOUS DISEASE

www.elsevier.com/locate/diagmicrobio

Updated multiplex polymerase chain reaction for detection of 16S rRNA methylases: high prevalence among NDM-1 producers☆

Béatrice Berçot[a,b], Laurent Poirel[a,*], Patrice Nordmann[a]

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Received 5 April 2011; accepted 25 August 2011

Detection of carriers: stools

JOURNAL OF CLINICAL MICROBIOLOGY, May 2010, p. 1913–1914 Vol. 48, No. 5
0095-1137/10/$12.00 doi:10.1128/JCM.02277-09

Use of ChromID Extended-Spectrum β-Lactamase Medium for Detecting Carbapenemase-Producing *Enterobacteriaceae*[▽]

Amélie Carrër, Nicolas Fortineau, and Patrice Nordmann*

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Received 20 November 2009/Returned for modification 12 January 2010/Accepted 5 March 2010

ChromID extended-spectrum β-lactamase (ESBL) culture medium is routinely used for screening ESBL producers. This medium was tested for detecting carbapenemase-producing *Enterobacteriaceae* isolates from a collection of reference strains and compared to the CHROMagar KPC culture medium previously evaluated for detecting KPC-producing isolates. Producers of IMP-, VIM-, and KPC-type carbapenemases with high levels of resistance to cephalosporins and to carbapenems were detected at 1×10^1 CFU/ml. The OXA-48 producers were not detected on ChromID ESBL medium unless coexpressing ESBLs, whereas carbapenemase-producing isolates with MICs of <4 μg/ml were not detected on CHROMagar KPC medium.

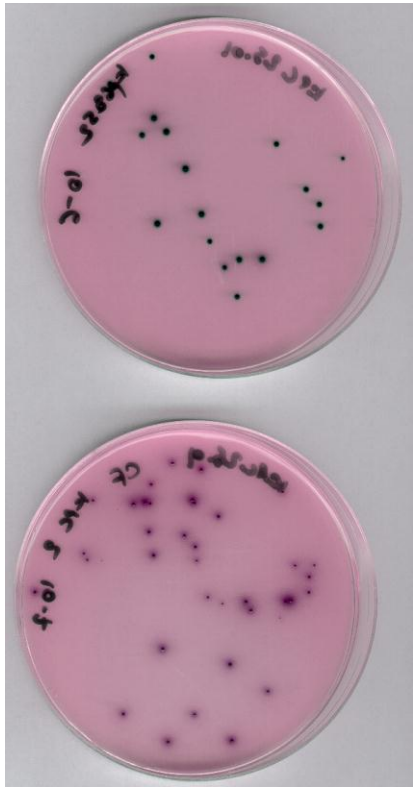
JOURNAL OF CLINICAL MICROBIOLOGY, Feb. 2011, p. 718–721 Vol. 49, No. 2
0095-1137/11/$12.00 doi:10.1128/JCM.01773-10

How To Detect NDM-1 Producers[▽]

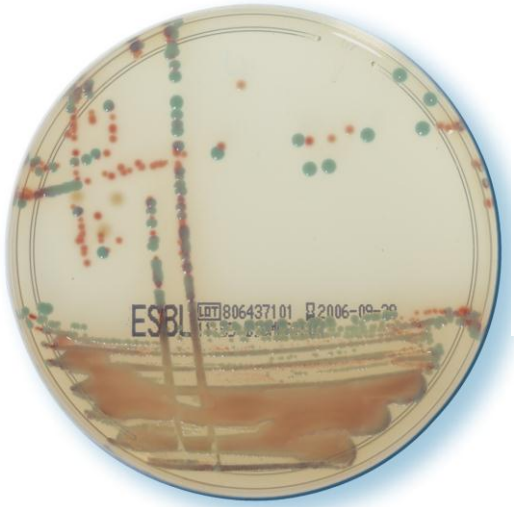
Patrice Nordmann,[1]* Laurent Poirel,[1] Amélie Carrër,[1] Mark A. Toleman,[2] and Timothy R. Walsh[2]

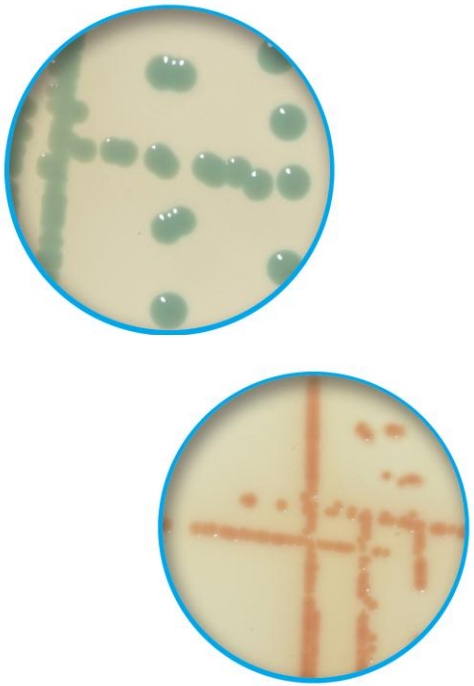
Service de Bactériologie-Virologie, INSERM U914 Emerging Resistance to Antibiotics, Hôpital de Bicêtre, Assistance Publique/Hôpitaux de Paris, Faculté de Médecine Paris Sud, Le Kremlin-Bicêtre, France,[1] and Department of Immunology, Immunity and Biochemistry, School of Medicine, Cardiff University, Cardiff, United Kingdom[2]

Detection of carbapenemase producers in stools



CHROMagar KPC
Selector: CARBAPENEM
++





ChromID ESBL bioMerieux

Selector : CEPHALOSPORIN

+++

Real-Time PCR for detection of NDM-1-carbapenemase gene from stools

ANTIMICROBIAL AGENTS AND CHEMOTHERAPY, Sept. 2011, p. 4038–4043 Vol. 55, No. 9
0066-4804/11/$12.00 doi:10.1128/AAC.01734-10

Real-Time PCR for Detection of NDM-1 Carbapenemase Genes from Spiked Stool Samples[▽]

Thierry Naas,†* Ayla Ergani,† Amélie Carrër, and Patrice Nordmann

Service de Bactériologie-Virologie, INSERM U914: Emerging Resistance to Antibiotics, Hôpital de Bicêtre, 94275 Le Kremlin-Bicêtre, and Assistance Publique-Hôpitaux de Paris, Faculté de Médecine Paris-Sud, Paris, France

Highly specific and sensitive qPCR (limit of detection +/-10^1 bacteria / 100 mg of stool

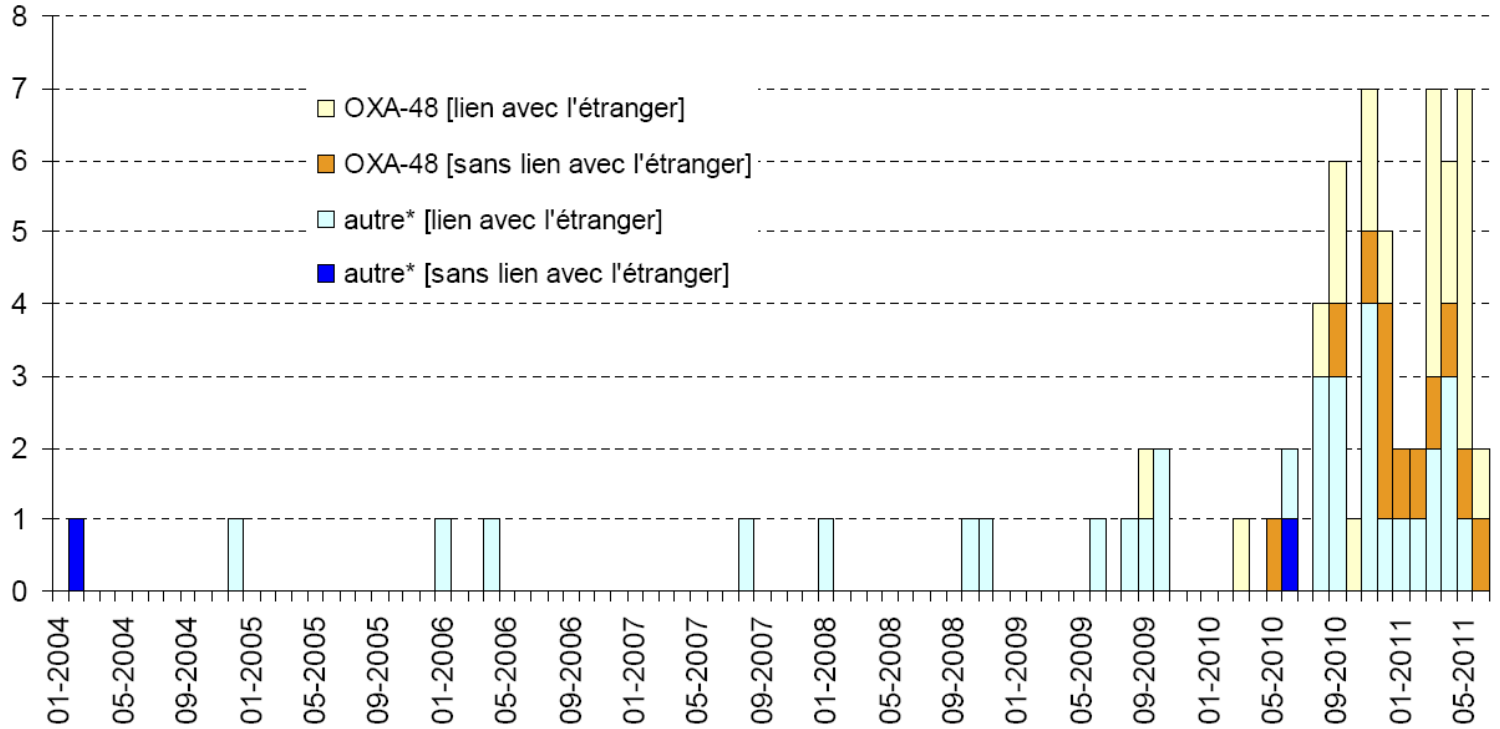
=> use of enrichment culture (increased detection?)

Useful tool for outbreak management (rapid cohorting of patients, infection control measure evaluation, ….)

At least as sensitive as culture on stool samples,
but 4h turn around time vs 48h-72h for culture

Needs to be evaluated on rectal swabs &/ in outbreak situation. Extraction can be automated

Nombre d'épisodes impliquant des EPC par mois de signalement



* autre carbapénèmase (KPC, VIM, IMP, NDM-1)

67 épisodes au total

2009 : 6 2010 : 26 2011 : 27

Espèces et mécanismes

Espèces impliquées dans les épisodes signalés (N=67)

Bactérie	Nombre d'épisodes
K. pneumoniae	43
E. coli	18
E. cloacae	8
E. aerogenes	3
C. freundii	2
P. mirabilis	1
S. marcescens	1
Total	**67***

** 2 ou 3 entérobactéries présentant le même mécanisme de résistance impliquées dans 8 épisodes*

Enregistrer une copie

Mécanismes impliqués dans les épisodes signalés (N=67)

Mécanisme de résistance	Nombre d'épisodes
OXA-48	33
KPC	19
VIM	9
NDM-1	8
IMP	1
Total	**67***

** 2 mécanismes de résistance associés dans 3 épisodes*

1 épisode = 1 ou plusieurs cas

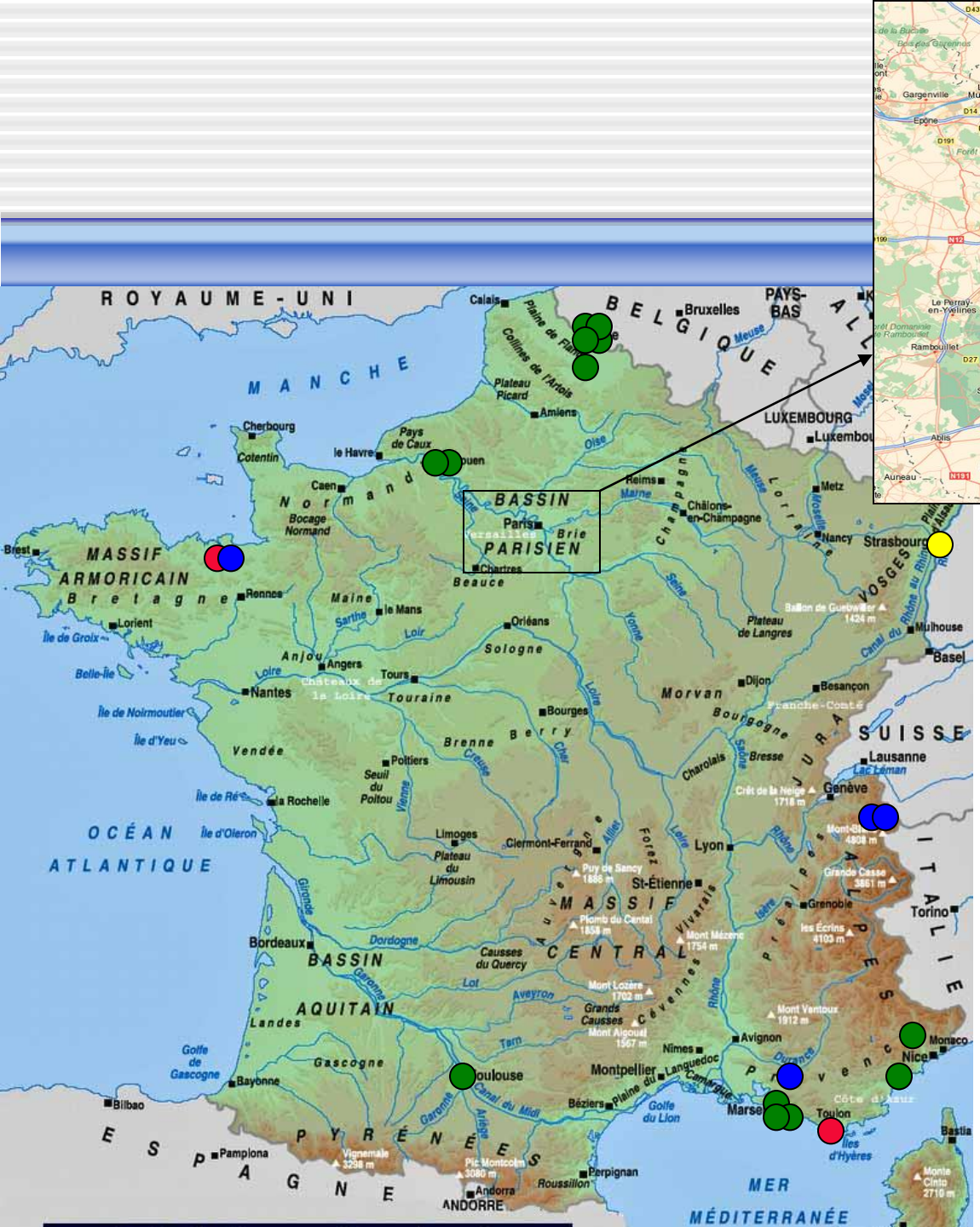
67 épisodes = 193 cas (infectés ou colonisés)

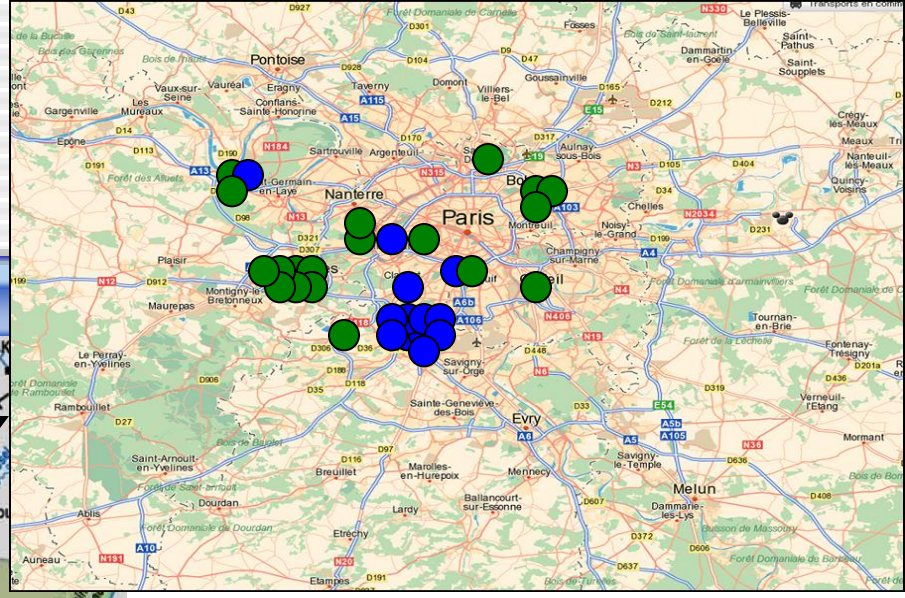
- **Lien avec un pays étranger pour 53 épisodes**

Contexte	Nombre d'épisodes	(%)
Rapatriement sanitaire (transfert)	34	(64%)
Hospitalisation pendant le séjour	11	(21%)
Délai avant hospitalisation en France		
Quelques jours	2	
Quelques semaines	1	
Dans les 1 à 6 mois	6	
Dans l'année	2	
Résident en France, voyage à l'étranger	5	(9%)
Résident à l'étranger sans hospitalisation rapportée	3	(6%)
Total	53	(100%)

Pays	Total
Grèce	16
Maroc	11
Inde	7
Italie	4
Algérie	3
Tunisie	3
Egypte	3
Sénégal	2
Turquie	2
Irak	1
Etats-Unis	1
Total	53

14 épisodes sans lien avec un pays étranger, OXA-48 pour 12

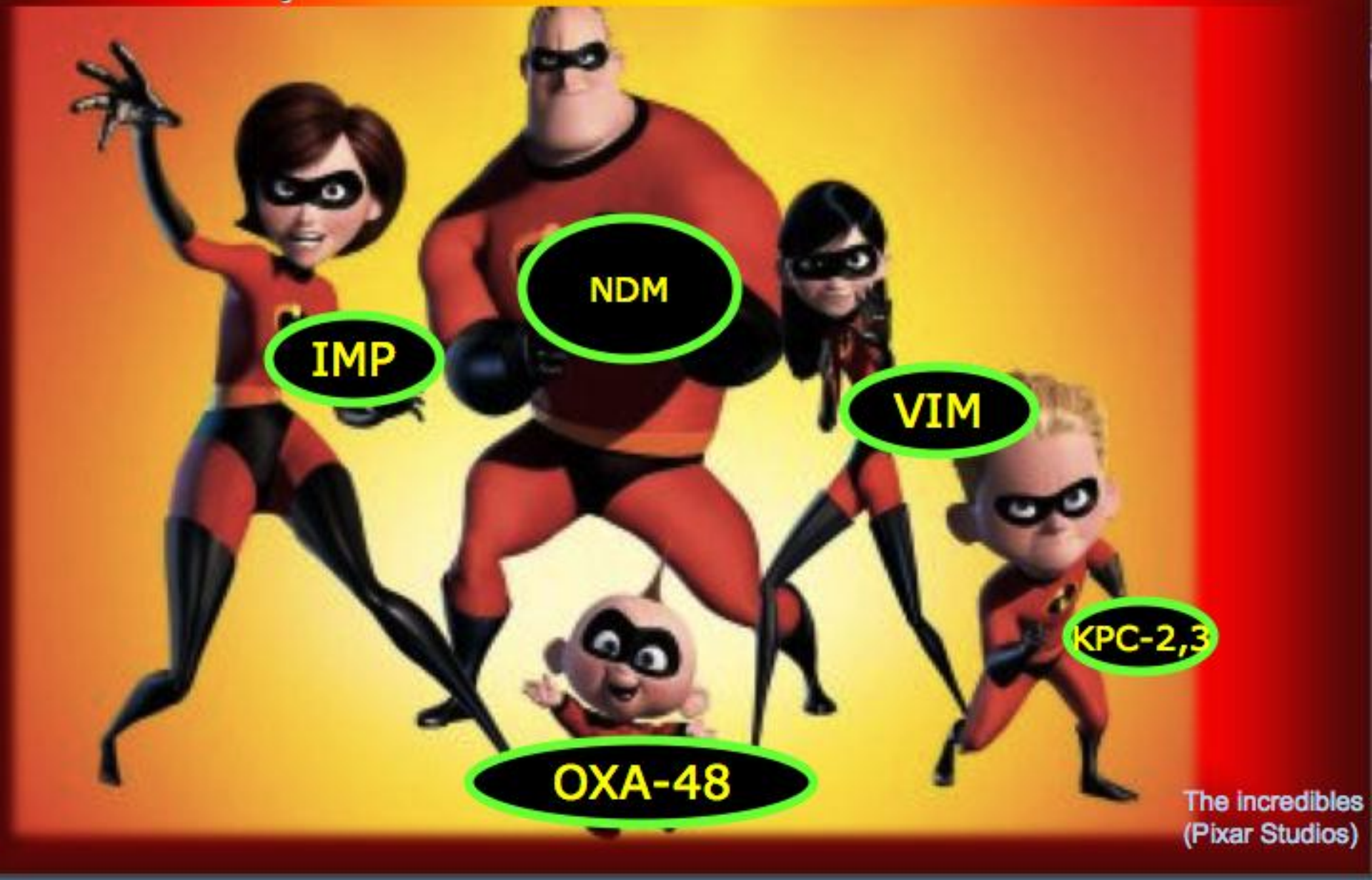




- VIM
- OXA 48
- NDM
- KPC

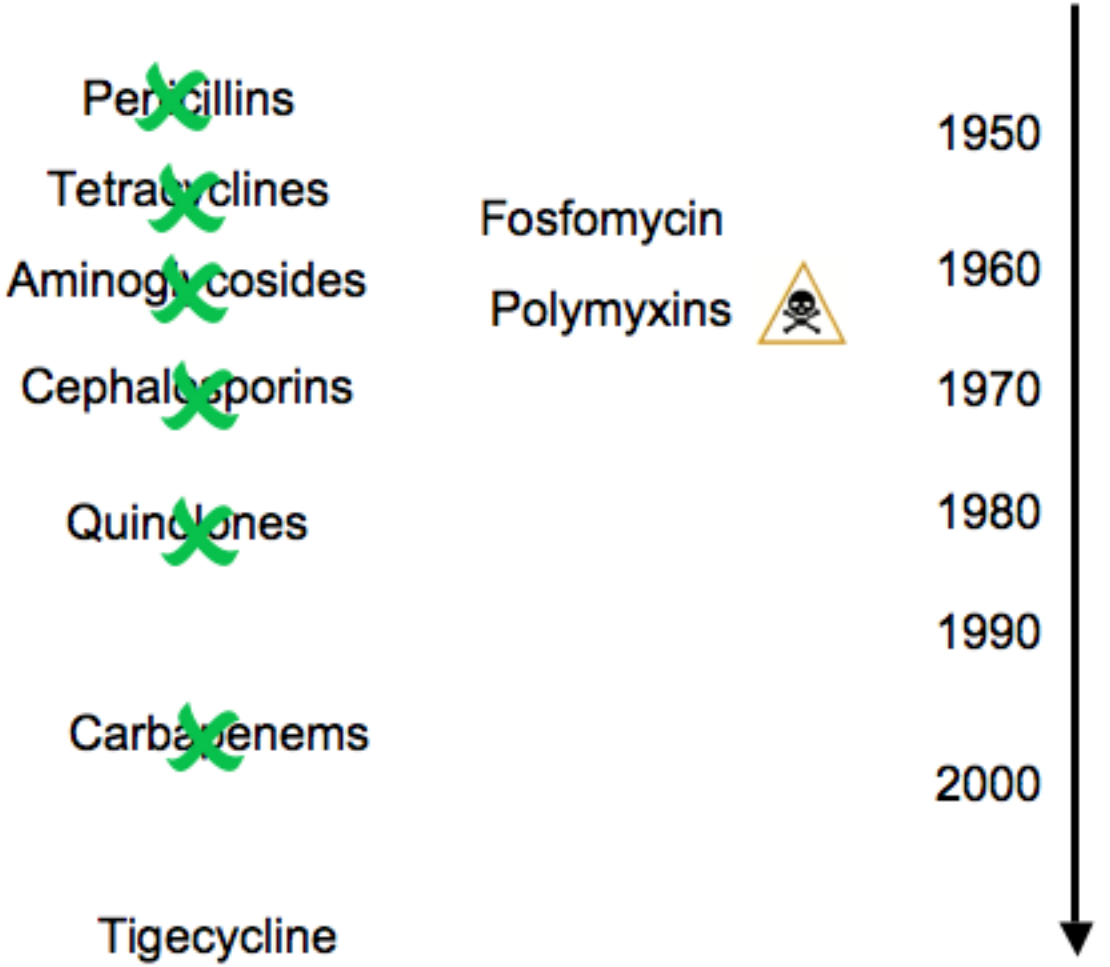
Ile de France : 32/52 (62%)

Carbapenemases in Enterobacteriaceae



The Incredibles
(Pixar Studios)

Rise of Antimicrobial Resistance in Gram-negatives



Take home message

- Increase prevalence of carbapenemase producers worldwide

- Carbapenemase producers; multiplicity of clones and of genetic vectors

- Multidrug resistance and pandrug resistance
- Reversion of multidrug resistance in Gram negatives; uncommon

- Difficult detection; a need for rapid identification techniques
- Nosocomial outbreaks are reported worldwide with all types of carbapenemase producers

- Spread of carbapenemase producers (NDM, OXA-48) in the community is an important source of concern
- An urgent need for new drugs for this next decade

Carbapenemases; a nightmare !