Nosocomial Outbreak of OXA-48-Producing *Klebsiella pneumoniae* in a Chinese Hospital: Clonal Transmission of ST147 and ST383

Ling Guo¹, Jingna An¹,², Yanning Ma¹, Liyan Ye¹, Yanping Luo¹, Chuanmin Tao², Jiyong Yang¹*  

¹ Department of Microbiology, Chinese PLA General Hospital, Beijing, China, ² Department of Laboratory Medicine, West China Hospital, Sichuan University, Chengdu, China  

* yangjy301@hotmail.com

Abstract

Background

In China, the spread and outbreak of OXA-48-producing *Enterobacteriaceae* remains largely unknown.

Methods

OXA-48-producing isolates were analyzed for genetic relatedness by pulsed-field gel electrophoresis (PFGE), antimicrobial susceptibility by E-test, and sequence type (ST) by multilocus sequence typing. S1-PFGE and southern blotting were used for plasmid profiling, and PCR and subsequent sequencing were performed to determine the genetic environment of *bla*<sub>OXA-48</sub> gene.

Results

In total, 37 non-duplicated OXA-48-producing *K. pneumoniae* (OXAKp) isolates were recovered. From December 2013 to August 2014, an outbreak was observed at a respiratory ICU. The 37 isolates of *K. pneumoniae* were categorized into four PFGE types (A, B, C, and D). The predominant strains associated with the outbreak were strains with PFGE type A and B, which belonged to ST383 and ST147, respectively. Plasmid sequencing revealed that the *bla*<sub>OXA-48</sub>-carrying plasmid is 69,069 bp in length and belongs to the IncL/M incompatibility group. Sequence analysis revealed that the IS1999 element was located upstream of the *bla*<sub>OXA-48</sub> gene and was truncated by IS1R.

Conclusions

In this study, the dissemination and outbreak of OXAKp isolates were clonal, and ST147 and ST383 *K. pneumoniae* were the predominant clones that were associated with the outbreak. Meanwhile, the horizontal transfer of plasmids potentially mediate the spread of *bla*<sub>OXA-48</sub> gene between different *K. pneumoniae* strains.
Introduction

Global spread of carbapenemase-producing *Klebsiella pneumoniae* is a growing clinical problem and public health threat [1]. In 2004, a novel class D carbapenemase, OXA-48 oxacillinase, was identified in a clinical *K. pneumoniae* isolate [2]. Since then, OXA-48-producing *K. pneumoniae* (OXAKp) has been primarily reported in Turkey, and in countries of the Middle East, North Africa, and Europe [1,3]. In some countries, OXAKp accounted for the majority of carbapenemase-producing *Enterobacteriaceae* [4,5].

To date, OXA-48 and its several variants have been identified in *Enterobacteriaceae* [6,7]. These variants differ from OXA-48 by one to five amino acid substitutions [6]. OXA-48-type carbapenemases weakly hydrolyze carbapenems, but does not exhibit activity against extended-spectrum cephalosporins and aztreonam. However, some OXA-48-like variants confer resistance to broad spectrum cephalosporins and cephalosporins resistance that is associated with impaired permeability or the production of extended-spectrum β-lactamases (ESBLs) [3]. A recent study demonstrated that the OXA-48-like variants possessed different carbapenems hydrolytic properties, while OXA-163 variant did not exhibit significant carbapenemase activity [8].

In Europe, diverse sequence types (STs) of dominant OXAKp have been identified in outbreaks or solitary case reports (STs 11, 14, 15, 16, 17, 45, 101, 104, 147, 326, 392, 395 and 405) [9,10,11,12,13,14]. In Asia, OXAKp ST15 have been identified in India [13], and OXAKp ST11 and ST116 were present in Taiwan [15].

The *bla*<sub>OXA-48</sub> gene is found on a IncL/M-type self-transferable plasmid of approximately 62 kb that was disseminated in various enterobacterial species [3]. Other types of *bla*<sub>OXA-48</sub>-carrying plasmids (e.g. IncA/C, IncFIA, and IncF) have also been identified in *Enterobacteriaceae* [11,12,15]. The *bla*<sub>OXA-48</sub> gene is flanked by two IS1999 elements to form a functional composite transposon Tn1999, which does not carry any other antibiotic resistance gene [2,16]. In addition, a novel Tn1999 transposon variant (Tn1999.2) has been identified in which the IS1999 element was located upstream of the *bla*<sub>OXA-48</sub> gene and truncated by ISIR [17]. Two other Tn1999 transposon derivatives (Tn1999.3 and Tn1999.4) were also discovered located downstream of the *bla*<sub>OXA-48</sub> gene and truncated by ISIR or by a more complicated genetic structure (named Tn2015) [18,19].

Currently, OXA-48-producing *Enterobacteriaceae* has been reported only in regions of Taiwan [15] and has not been found in other regions of China. In this study, we report a nosocomial outbreak of OXAKp at our hospital involving 34 patients. The phenotypic and genotypic characteristics of OXAKp isolates were analyzed.

Materials and Methods

Bacterial isolates

All clinical enterobacterial isolates were collected from a 4000-bed tertiary-care hospital and were identified by VITEK<sup>®</sup> MS (bioMérieux SA, Marcy-l’Etoile, France). No ethical approval was obtained for using the clinical samples since they were collected during routine bacteriologic analyses in public hospitals. All data were anonymously analyzed.

Antimicrobial susceptibility testing

The MICs of cefotaxime (CTX), piperacillin-tazobactam (TZP), imipenem (IMP), meropenem (MEM), ertapenem (ETP), amikacin (AK) and levofloxacin (LEV) were measured by E-test (AB bioMérieux, Solna, Sweden). *E. coli* ATCC 25922 was used as the quality control strains.
for antimicrobial susceptibility testing. Results were interpreted according to the interpretive standards of the Clinical Laboratory Standards Institute [20].

Detection of specific porin and resistance genes

The isolates that exhibited non-susceptibility to carbapenems were screened for \( \text{bla}_{OXA-48} \) by PCR amplification and subsequent amplicon sequencing as previously described [2]. PCR detection of \( \text{bla}_{CTX-M} \) genes was performed as previously described [21]. The DNA sequences of outer membrane protein (OMP) genes were analyzed as previously described [22]. All PCR products were purified and sequenced.

Pulsed-field gel electrophoresis (PFGE) and MLST analysis

PFGE with \( XbaI \) was performed for OXAKp isolates [23]. \textit{Salmonella} ser. Braenderup strain (H9812) was used as a reference standard for PFGE. MLST was carried out according to protocols provided on MLST websites (http://www.pasteur.fr/recherche/genopole/PF8/mlst/Kpneumoniae).

Plasmid analysis and Southern blot

The transferability of plasmids was demonstrated by conjugation experiments [23]. Sodium azide resistant \textit{E. coli} J53 was used as the recipient for conjugation testing. The resistance plasmids were typed by using several simplex and multiplex PCR [24,25]. A \( \text{bla}_{OXA-48} \) probe was generated by labeling a \( \text{bla}_{OXA-48} \) PCR product by the PCR DIG Probe Synthesis Kit (Roche Applied Sciences, Mannheim, Germany). Plasmid analysis was performed in one representative of each PFGE profile, and the S1-PFGE and Southern blot were performed [23]. A \( \text{bla}_{OXA-48} \)-carrying plasmid from type A strain was sequenced using the Illumina MiSeq system [26].

Genetic environment analysis of \( \text{bla}_{OXA-48} \) gene

To confirm the upstream genetic structures of the \( \text{bla}_{OXA-48} \) gene, a primer pair Tn1999-F (5’-AGTTCTGGGCGATTTGTTG) and Tn1999-R (5’-ACACGCATAACGTCCGCTTG) were used to amplify a 192 bp or 970 bp fragments from the IS1999 (GenBank no. JN626286) or the IS1R-truncated IS1999 (GenBank no. JN714122), respectively. All PCR products were purified and sequenced.

Results

Emergence and outbreak of OXAKp

In total, 2310 \textit{K. pneumoniae} isolates were recovered from various clinical specimens at our hospital between March 2013 and July 2015, and 247 (10.69%) of these were found to be non-susceptible to carbapenems. During this period, 37 non-duplicated OXAKp isolates were recovered from 34 patients in two ICUs and three other clinical wards (Fig 1). There were three patients from whom the OXAKp isolates were recovered from different sites of the same patient. Among these isolates, 23 (62.2%) were recovered from sputum, while 5 and 9 strains were recovered from blood and urine sample, respectively. In April 2013, OXAKp first appeared at the surgical ICU. This followed by an outbreak from December 2013 to August 2014 at a respiratory ICU (RICU) (Fig 1). Among 34 patients with OXAKp isolates, 22 (64.7%) died. Nine patients (26.5%) showed improvement, while two patients and another patient were successfully cured and discharged, respectively.
The 37 isolates of *K. pneumoniae* were categorized into four PFGE types (A, B, C, and D). The majority of the isolates belonged to types A and B, while types C and D isolates were detected in two and one samples respectively (Table 1). Before December 2013, only type A strain was sporadically found at SICU and RICU. Eight months after the emergence of type A OXAKp strain, only sporadic cases were identified in two ICUs. This followed by an outbreak in a respiratory ICU in the next nine months. Meanwhile, new OXAKp clones (types B and C) emerged and were found spread to other wards (Fig 1). MLST was performed in one representative of each PFGE profile and three sequence types (STs) were identified. Types A and D strains belonged to ST383, while types B and C were categorized with ST147 and ST13, respectively.

**Antimicrobial susceptibilities**

The antimicrobial susceptibility patterns for the isolates are listed in Table 1. All *K. pneumoniae* isolates presented resistance to CTX and TZP and exhibited heterogeneous carbapenem resistance patterns.

**OMP and CTX-M beta-lactamase genes**

No mutations were found in the *ompK36* coding region. For the *ompk35* gene, an intact open reading frame of 738 bp was found among types B and C isolates, whereas a much larger DNA fragments (1505 bp) were amplified from types A and D isolates. Sequencing analysis found that an additional insertion sequence, IS1R (768 bp), was inserted after nucleotide position 105 of the *ompK35* gene. Furthermore, a 9 bp duplication of the target site (CTGGACTTC) was identified (Fig 2). Types B and C isolates produced both CTX-M-14 and CTX-M-15, while only CTX-M-14 was detected among types A and D isolates (Table 1).

**Plasmid analysis and genetic environment of blaOXA-48 gene**

S1-PFGE and southern blot analysis showed that the *bla*OXA-48* gene in all types of strains was located on an approximately 60 kb plasmid. Plasmid sequencing revealed that the *bla*OXA-48*-carrying plasmid is 69,069 bp in length and belongs to the IncL/M incompatibility group. A BLAST search against all completely sequenced *bla*OXA-48*-harboring plasmids in GenBank (http://www.ncbi.nlm.nih.gov/GenBank/) showed that the plasmid analyzed in this study displayed overall nucleotide identity (99.28%) to pOXA48-PM (GenBank: KP025948.1), a *bla*OXA-48*-carrying IncL/M plasmid from a *Proteus mirabilis* strain (Pm-OXA-48) [27]. Sequence
Table 1. Phenotypic and genotypic characteristics of OXA-48-producing K. pneumoniae isolates.

| No. | Isolate No. | Source | PFGE type | ST  | ompk35  | ompk36  | CTX-M | Minimal inhibitory concentration (mg/L) |
|-----|-------------|--------|-----------|-----|---------|---------|-------|---------------------------------|
|     |             |        |           |     |         |         |       | CTX   | TZP   | IPM   | MEM   | ETP   | AK    | LEV   |
| 1   | IR5065      | Blood  | A         | ST383| Truncated*| Intact  | 14    | >256 | >256 | 2  | 2  | 32  | 64  | >32 |
| 2   | IR5067      | Urine  | A         | ST383| Truncated*| Intact  | 14    | >256 | >256 | 4  | 1  | 4   | >256 | >32 |
| 3   | IR5070      | Sputum | A         | ST383| Truncated*| Intact  | 14    | >256 | >256 | 2  | 1  | 4   | 128  | >32 |
| 4   | IR5075      | Blood  | A         | ST383| Truncated*| Intact  | 14    | >256 | >256 | >32 | >32 | >32 | 64   | >32 |
| 5   | IR5082      | Blood  | A         | ST383| Truncated*| Intact  | 14    | >256 | >256 | 2  | 1  | 32  | 128  | >32 |
| 6   | IR5083      | Sputum | A         | ST383| Truncated*| Intact  | 14    | >256 | >256 | >32 | >32 | >32 | >256 | >32 |
| 7   | IR5088      | Sputum | A         | ST383| Truncated*| Intact  | 14    | >256 | >256 | >32 | >32 | >32 | >256 | >32 |
| 8   | IR5090      | Urine  | A         | ST383| Truncated*| Intact  | 14    | >256 | >256 | 8  | >32 | >32 | >256 | >32 |
| 9   | IR5097      | Urine  | A         | ST383| Truncated*| Intact  | 14    | >256 | >256 | >32 | >32 | >32 | >256 | >32 |
| 10  | IR5098      | Sputum | A         | ST383| Truncated*| Intact  | 14    | >256 | >256 | >32 | >32 | >32 | 96   | >32 |
| 11  | IR5099      | Sputum | A         | ST383| Truncated*| Intact  | 14    | >256 | >256 | >32 | >32 | >32 | >256 | >32 |
| 12  | IR5100      | Sputum | A         | ST383| Truncated*| Intact  | 14    | >256 | >256 | 2  | 1  | 32  | 48   | >32 |
| 13  | IR5602      | Sputum | A         | ST383| Truncated*| Intact  | 14    | >256 | >256 | 8  | >32 | >32 | 48   | >32 |
| 14  | IR5604      | Sputum | A         | ST383| Truncated*| Intact  | 14    | >256 | >256 | >32 | >32 | >32 | >256 | >32 |
| 15  | IR5607      | Urine  | A         | ST383| Truncated*| Intact  | 14    | >256 | >256 | 4  | 2  | 4   | 64   | >32 |
| 16  | IR5609      | Urine  | A         | ST383| Truncated*| Intact  | 14    | >256 | >256 | >32 | >32 | >32 | 128  | >32 |
| 17  | IR5610      | Sputum | A         | ST383| Truncated*| Intact  | 14    | >256 | >256 | 2  | 1  | 4   | 64   | >32 |
| 18  | IR5614      | Urine  | A         | ST383| Truncated*| Intact  | 14    | >256 | >256 | 2  | 1  | 4   | 96   | >32 |
| 19  | IR5615      | Sputum | A         | ST383| Truncated*| Intact  | 14    | >256 | >256 | 4  | 4  | 32  | 128  | >32 |
| 20  | IR5618      | Sputum | A         | ST383| Truncated*| Intact  | 14    | >256 | >256 | >32 | >32 | >32 | >256 | >32 |
| 21  | IR5629      | Urine  | A         | ST383| Truncated*| Intact  | 14    | >256 | >256 | 2  | 4  | 4   | 64   | >32 |
| 22  | IR5630      | Sputum | A         | ST383| Truncated*| Intact  | 14    | >256 | >256 | 2  | 4  | 4   | 32   | >32 |
| 23  | IR5632      | Blood  | A         | ST383| Truncated*| Intact  | 14    | >256 | >256 | >32 | >32 | >32 | >256 | >32 |
| 24  | IR5633      | Sputum | A         | ST383| Truncated*| Intact  | 14    | >256 | >256 | 2  | 4  | 4   | 64   | >32 |
| 25  | IR5639      | Sputum | A         | ST383| Truncated*| Intact  | 14    | >256 | >256 | >32 | >32 | >32 | 64   | >32 |
| 26  | IR5647      | Sputum | A         | ST383| Truncated*| Intact  | 14    | >256 | >256 | 2  | 4  | 4   | 64   | >32 |
| 27  | IR5085      | Sputum | B         | ST147| Intact    | Intact  | 14,15 | >256 | >256 | 4  | 2  | 16  | >256 | >32 |
| 28  | IR5086      | Sputum | B         | ST147| Intact    | Intact  | 14,15 | >256 | >256 | 4  | 1  | 16  | 64   | >32 |
| 29  | IR5087      | Sputum | B         | ST147| Intact    | Intact  | 14,15 | >256 | >256 | 4  | 1  | 8   | 64   | >32 |
| 30  | IR5089      | Sputum | B         | ST147| Intact    | Intact  | 14,15 | >256 | >256 | 2  | 1  | 2   | 64   | >32 |
| 31  | IR5092      | Sputum | B         | ST147| Intact    | Intact  | 14,15 | >256 | >256 | 4  | 1  | 4   | 64   | >32 |
| 32  | IR5601      | Blood  | B         | ST147| Intact    | Intact  | 14,15 | >256 | >256 | 2  | 1  | 8   | 64   | >32 |
| 33  | IR5603      | Sputum | B         | ST147| Intact    | Intact  | 14,15 | >256 | >256 | 4  | 1  | 6   | >256 | >32 |
| 34  | IR5612      | Sputum | B         | ST147| Intact    | Intact  | 14,15 | >256 | >256 | 2  | 1  | 4   | 64   | >32 |
| 35  | IR5093      | Urine  | C         | ST13 | Intact    | Intact  | 14,15 | >256 | >256 | 2  | 1  | 16  | >256 | >32 |
| 36  | IR5094      | Sputum | C         | ST13 | Intact    | Intact  | 14,15 | >256 | >256 | 4  | 4  | 16  | 64   | >32 |
| 37  | IR5649      | Sputum | D         | ST383| Truncated*| Intact  | 14    | >256 | >256 | 8  | 4  | 16  | 64   | >32 |

CTX: cefotaxime, TZP: piperacillin-tazobactam, IMP: imipenem, MEM: meropenem, ETP: ertapenem, AK: amikacin.
*: truncated by IS1R.

doi:10.1371/journal.pone.0160754.l001

analysis revealed that the IS1999 element was located upstream of the bla<sub>OXA-48</sub> gene and was truncated by IS1R.

**Discussion**

OXA-48 oxacillinase was identified in a K. pneumoniae isolate from Turkey in 2001 [2].
Since then, OXAKp isolates have been detected worldwide [3]. In Asia, OXAKp have only been identified in India [13] and Taiwan area of China [15]. In this study, 37 OXAKp isolates were collected between March 2013 and July 2015. A total of 247 clinical K. pneumoniae isolates exhibited non-susceptibility to carbapenems. In general, 14.98% (37/247) of clinical K. pneumoniae isolates that were non-susceptible to carbapenem produced OXA-48. PFGE analysis demonstrated that the majority (n = 26) of OXAKp isolates belonged to the same type (A clone) followed by type B clone (n = 8). OXAKp mainly disseminate in the respiratory ICU (Fig 1) and appeared to be clonal. Hospital outbreaks linked to patient transfer have also been observed in some European countries [3,4,5]. In France, OXAKp presents two-thirds of the carbapenemase-producing enterobacteriaceae [4]. Therefore, it is very important to recognize the impact of clonal dissemination on the prevalence of OXAKp and to strengthen the surveillance and reporting system of OXAKp in hospital infection control measures. In this study, 22 of 34 (64.7%) patients died, possibly from illnesses related to OXAKp-associated infections. However, most patients were hospitalized in the ICU and presented with severe conditions. As a result, it is difficult to determine whether the death of the patients were associated with the infections or adverse physical conditions. It has been confirmed that antimicrobial therapy is strongly associated with patient survival [28]. However, a high mortality rate was observed, although most patients had received carbapenems therapy in this study. Therefore, the mortality may be associated with the clinical conditions of the patients upon infection, rather than with infection itself.

OXAKp exhibits high-level carbapenem resistance when OXA-48 carbapenemases are associated with the production of ESBLs and impaired permeability [3]. In this study, type A and type B isolates produced both OXA-48 and CTX-M; however, none of these isolates exhibited high-levels of carbapenem resistance (Table 1). In addition, although all of the ompK35 genes have been truncated by IS1R, about half of the type A isolates also presented low-level carbapenem resistance (Table 1). It is unclear to what extent the impacts of ESBLs and impaired outer membrane protein are on the carbapenem resistance of OXAKp. In most clinical microbiology laboratories in China, isolates with carbapenem-sensitive phenotype were undetected, regardless of whether they produce carbapenemases or not. The weak hydrolytic ability of OXA-48 to carbapenems may lead to an underestimation of the prevalence of OXA-48-producing isolates in China, and calls for changes in clinical microbiology analysis procedures that would enhance phenotypic and molecular detection of OXA-48 types of carbapenems.

In outbreaks of OXAKp worldwide, diverse sequence types of OXAKp have been reported. This included several dominant ones (e.g., STs 11, 14, 15, 16, 17, 45, 101, 104, 147, 326, 392, 395 and 405) [9,10,11,12,13]. In this study, the predominant clone of OXAKp belonged to ST383 and ST147, which were found in 27 and 8 isolates, respectively. Outbreaks of OXAKp
ST147 are common in Europe [9,10,11,29], whereas prevalence of OXAKp ST383 have been sporadically reported in the United Kingdom [29]. ST383 contributes significantly to the dissemination of the VIM-producing K. pneumoniae [30,31,32], suggesting that the carbapenem-resistant phenotype may confer some fitness advantage to the spread of the pathogen. To our knowledge, this is the first study to report the outbreak of OXAKp ST383 worldwide.

In this study, all of the strains carry plasmids of the same size and incompatibility type (approximately 70 kb, IncL/M), suggesting that horizontal transfer of bla_{OXA-48}-carrying plasmid may have occurred during the outbreak. The high nucleotide sequence homology between the bla_{OXA-48}-carrying plasmid analyzed in this study and the previously reported plasmid sequences (i.e., bla_{OXA-48}-carrying IncL/M plasmid from a Proteus mirabilis strain [Pm-OXA-48] [27]) also reveals possible sources of the plasmid. Other studies have confirmed that a single IncL/M plasmid of approximately 62 kb is the main source of the bla_{OXA-48} gene disseminated in a variety of enterobacterial species [16], and that the inter-genus transfer of bla_{OXA-48}-carrying plasmids might occur more frequently in vivo than previously estimated by in vitro experiments [33]. All of these data indicated the important role of the self-conjugative IncL/M plasmids with approximately 60–70 kb in size on the prevalence bla_{OXA-48} gene. The genetic environment of the bla_{OXA-48} gene has been characterized as a functional composite transposon, which was identified as Tn1999 [2,16] and several isoforms (Tn1999.2, Tn1999.3 and Tn1999.4) [17,18,19]. For strains analyzed in this study, the genetic environment of the bla_{OXA-48} are consistent with those of Tn1999.2, Tn1999.3, or Tn1999.4. In these strain, the IS1999 element is located upstream of the bla_{OXA-48} gene and is truncated by IS1R [19]. However, PCR and subsequent sequencing analyses failed to differentiate variants such as Tn1999.2, Tn1999.2 inverted, Tn1999.3, Tn1999.4, or other new variants. Additional overlapping PCRs and sequencing analyses are needed to reveal the downstream sequence of bla_{OXA-48} gene.

In conclusion, a clonal dissemination and outbreak of OXAKp has been observed. ST147 and ST383 K. pneumoniae were the predominant clones that were associated with the outbreak. Meanwhile, the horizontal transfer of bla_{OXA-48}-carrying plasmids potentially mediate the spread of bla_{OXA-48} gene among different K. pneumoniae strains.

**Author Contributions**

**Conceptualization:** JY YL CT.

**Data curation:** JY.

**Formal analysis:** JY.

**Funding acquisition:** JY.

**Investigation:** LG JA YM LY.

**Methodology:** JY YL CT.

**Project administration:** JY YL CT.

**Resources:** JY YL CT.

**Software:** JY.

**Supervision:** JY YL CT.

**Validation:** JY.

**Visualization:** JY.

**Writing - original draft:** JY.
Writing - review & editing: JY.

References
1. Pitout JD, Nordmann P, Poirel L (2015) Carbapenemase-Producing Klebsiella pneumoniae, a Key Pathogen Set for Global Nosocomial Dominance. Antimicrob Agents Chemother 59: 5873–5884. doi: 10.1128/AAC.01915-15 PMID: 26169401
2. Poirel L, Hertlit C, Tolun V, Nordmann P (2004) Emergence of oxacillinase-mediated resistance to imipenem in Klebsiella pneumoniae. Antimicrob Agents Chemother 48: 15–22. PMID: 14693513
3. Poirel L, Potron A, Nordmann P (2012) OXA-48-like carbapenemases: the phantom menace. J Antimicrob Chemother 67: 1597–1606. doi:10.1093/jac/dks121 PMID: 22499996
4. Vaux S, Carbone A, Thiolet JM, Jarlier V, Coignard B (2011) Emergence of carbapenemase-producing Enterobacteriaceae in France, 2004 to 2011. Euro Surveill 16.
5. Dautzenberg MJ, Ossewaarde JM, de Kraker ME, van der Zee A, van Burgh S, de Greeff SC, et al. (2014) Successful control of a hospital-wide outbreak of OXA-48 producing Enterobacteriaceae in the Netherlands, 2009 to 2011. Euro Surveill 19.
6. Dortet L, Oueslati S, Jeannot K, Tande D, Naas T, Nordmann P (2015) Genetic and biochemical characterization of OXA-405, an OXA-48-type extended-spectrum beta-lactamase without significant carbapenemase activity. Antimicrob Agents Chemother 59: 3823–3828. doi:10.1128/AAC.05058-14 PMID: 25870062
7. Evans BA, Amyes SG (2014) OXA beta-lactamases. Clin Microbiol Rev 27: 241–263. doi: 10.1128/CMR.00117-13 PMID: 24696435
8. Oueslati S, Nordmann P, Poirel L (2015) Heterogeneous hydrolytic features for OXA-48-like beta-lactamases. J Antimicrob Chemother 70: 1059–1063. doi: 10.1093/jac/dku524 PMID: 25583748
9. Voulgari E, Zarkotou O, Ranellou K, Karageorgopoulou DE, Vrioni G, Marnali V, et al. (2013) Outbreak of OXA-48 carbapenemase-producing Klebsiella pneumoniae in Greece involving an ST11 clone. J Antimicrob Chemother 68: 84–88. doi: 10.1093/jac/dks5916
10. Liapis E, Pantel A, Robert J, Nicolas-Chanoine MH, Cavaile L, van der Mee-Meunet N, et al. (2014) Molecular epidemiology of OXA-48-producing Klebsiella pneumoniae in France. Clin Microbiol Infect 20: O1121–O1123. doi: 10.1111/1469-0691.12727 PMID: 24942039
11. Poirel A, Poirel E, Rondinaud E, Nordmann P (2013) Intercontinental spread of OXA-48 beta-lactamase-producing Enterobacteriaceae over a 11-year period, 2001 to 2011. Euro Surveill 18.
12. Poirel A, Kalpoe J, Poirel L, Nordmann P (2011) European dissemination of a single OXA-48-producing Klebsiella pneumoniae clone. Clin Microbiol Infect 17: E24–E26. doi: 10.1111/j.1469-0691.2011.03669.x PMID: 21973185
13. Lascols C, Peirano G, Hackel M, Laupland KB, Pitout JD (2013) Surveillance and molecular epidemiology of Klebsiella pneumoniae isolates that produce carbapenemases: first report of OXA-48-like enzymes in North America. Antimicrob Agents Chemother 57: 130–136. doi: 10.1128/AAC.01686-12 PMID: 23070171
14. Oteo J, Hernandez JM, Espasa M, Fleites A, Saez D, Bautista V, et al. (2013) Emergence of OXA-48-producing Klebsiella pneumoniae and the novel carbapenemases OXA-244 and OXA-245 in Spain. J Antimicrob Chemother 68: 317–321. doi: 10.1093/jac/dks383 PMID: 23034714
15. Ma L, Wang JT, Wu TL, Siu LC, Chuang YC, Lin JC, et al. (2015) Emergence of OXA-48-Producing Klebsiella pneumoniae in Taiwan. PLoS One 10: e0139152. doi: 10.1371/journal.pone.0139152 PMID: 26414183
16. Poirel L, Bonnin RA, Nordmann P (2012) Genetic features of the widespread plasmid coding for the carbapenemase OXA-48. Antimicrob Agents Chemother 56: 559–562. doi: 10.1128/AAC.05289-11 PMID: 22083465
17. Carrer A, Poirel L, Eraksoy H, Cagatay AA, Badur S, Nordmann P (2008) Spread of OXA-48-positive carbapenem-resistant Klebsiella pneumoniae isolates in Istanbul, Turkey. Antimicrob Agents Chemother 52: 2935–2937. doi: 10.1128/AAC.01672-07 PMID: 18519712
18. Giani T, Conte V, Di Pilato V, Aschbacher R, Weber C, Larcher C, et al. (2012) Escherichia coli from Italy producing OXA-48 carbapenemase encoded by a novel Tn1999 transposon derivative. Antimicrob Agents Chemother 56: 2211–2213. doi:10.1128/AAC.00035-12 PMID: 22290939
19. Poirel A, Nordmann P, Rondinaud E, Jaureguy F, Poirel L (2013) A mosaic transposon encoding OXA-48 and CTX-M-15: towards pan-resistance. J Antimicrob Chemother 68: 476–477. doi: 10.1093/jac/dks397 PMID: 23027715
20. CLSI (2014) Performance Standards for Antimicrobial Susceptibility Testing; Twenty-Fourth Informational Supplement (M100-S24). Clinical and Laboratory Standards Institute, 950 West Valley Road, Suite 2500, Wayne, PA 19087 USA Vol.34 No.1.

21. Dallen C, Da Costa A, Decre D, Favier C, Arlet G (2010) Development of a set of multiplex PCR assays for the detection of genes encoding important beta-lactamases in Enterobacteriaceae. J Antimicrob Chemother 65: 490–495. doi:10.1093/jac/dkp498 PMID: 20071363

22. Lee CH, Chu C, Liu JW, Chen YS, Chiu CJ, Su LH (2007) Collateral damage of flomoxef therapy: in vivo development of porin deficiency and acquisition of blaDHA-1 leading to ertapenem resistance in a clinical isolate of Klebsiella pneumoniae producing CTX-M-3 and SHV-5 beta-lactamases. J Antimicrob Chemother 60: 410–413. PMID:17576696

23. Yang J, Ye L, Guo L, Zhao Q, Chen R, Tian S, et al. (2013) A nosocomial outbreak of KPC-2-producing Klebsiella pneumoniae in a Chinese hospital: dissemination of ST11 and emergence of ST37, ST392 and ST395. Clin Microbiol Infect.

24. Johnson TJ, Wannemuehler YM, Johnson SJ, Logue CM, White DG, Doetkott C, et al. (2007) Plasmid replicon typing of commensal and pathogenic Escherichia coli isolates. Appl Environ Microbiol 73: 1976–1983. PMID: 17277222

25. Johnson TJ, Bielak EM, Fortini D, Hansen LH, Hasman H, Debroy C, et al. (2012) Expansion of the IncX plasmid family for improved identification and typing of novel plasmids in drug-resistant Enterobacteriaceae. Plasmid 68: 43–50. doi: 10.1016/j.plasmid.2012.03.001 PMID: 22470007

26. Chen L, Hu H, Chavda KD, Zhao S, Liu R, Liang H, et al. (2014) Complete sequence of a KPC-producing IncN multidrug-resistant plasmid from an epidemic Escherichia coli sequence type 131 strain in China. Antimicrob Agents Chemother 58: 2422–2425. doi: 10.1128/AAC.02567-13 PMID: 24395232

27. Chen L, Al Laham N, Chavda KD, Mediavilla JR, Jacobs MR, Bonomo RA, et al. (2015) First report of an OXA-48-producing multidrug-resistant Proteus mirabilis strain from Gaza, Palestine. Antimicrob Agents Chemother 59: 4305–4307. doi: 10.1128/AAC.00565-15 PMID: 25896692

28. Daikos GL, Tsaousi S, Tzouvelekis LS, Anyfantis I, Psychogiou M, Argyropoulou A, et al. (2014) Carbapenemase-producing Klebsiella pneumoniae bloodstream infections: lowering mortality by antibiotic combination schemes and the role of carbapenems. Antimicrob Agents Chemother 58: 2322–2328. doi: 10.1128/AAC.02166-13 PMID: 24514083

29. Dimou V, Dhanji H, Pike R, Livermore DM, Woodford N (2012) Characterization of Enterobacteriaceae producing OXA-48-like carbapenemases in the UK. J Antimicrob Chemother 67: 1660–1665. doi: 10.1093/jac/dks124 PMID: 22532467

30. Papagiannitsis CC, Giakkoupi P, Vatopoulos AC, Tryfinopoulou K, Mirigou V, Tzouvelekis LS (2010) Emergence of Klebsiella pneumoniae of a novel sequence type (ST383) producing VIM-4, KPC-2 and CMY-4 beta-lactamases. Int J Antimicrob Agents 36: 573–574. doi: 10.1016/j.ijantimicag.2010.07.018 PMID: 20983669

31. Samuelson O, Toleman MA, Hasselvetd V, Fuursted K, Leegaard TM, Walsh TR, et al. (2011) Molecular characterization of VIM-producing Klebsiella pneumoniae from Scandinavia reveals genetic relatedness with international clonal complexes encoding transferable multidrug resistance. Clin Microbiol Infect 17: 1811–1816. doi: 10.1111/j.1469-0691.2011.03532.x PMID: 21595797

32. Papagiannitsis CC, Izdebski R, Baraniak A, Flett J, Herda M, Hrabáková J, et al. (2015) Survey of metallo-beta-lactamase-producing Enterobacteriaceae colonizing patients in European ICUs and rehabilitation units, 2008–11. J Antimicrob Chemother 70: 1981–1988. doi: 10.1093/jac/dkv055 PMID: 25759034

33. Gottig S, Gruber TM, Sticher B, Wichelhaus TA, Kempf VA (2015) In vivo horizontal gene transfer of the carbapenemase OXA-48 during a nosocomial outbreak. Clin Infect Dis 60: 1808–1815. doi: 10.1093/cid/civ191 PMID: 25759432