OXA-48-like carbapenemases: the phantom menace

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OXA-48-type carbapenem-hydrolysing class D β-lactamases are increasingly reported in enterobacterial species. To date, six OXA-48-like variants have been identified, with OXA-48 being the most widespread. They differ by a few amino acid substitutions or deletions (one to five amino acids). The enzymes hydrolyse penicillins at a high level and carbapenems at a low level, sparing broad-spectrum cephalosporins, and are not susceptible to β-lactamase inhibitors. When combining permeability defects, OXA-48-like producers may exhibit a high level of resistance to carbapenems. OXA-163 is an exception, hydrolysing broad-spectrum cephalosporins but carbapenems at a very low level, and being susceptible to β-lactamase inhibitors. The blaOXA-48-type genes are always plasmid-borne and have been identified in association with insertion sequences involved in their acquisition and expression. The current spread of the blaOXA-48 gene is mostly linked to the dissemination of a single IncL/M-type self-transferable plasmid of 62 kb that does not carry any additional resistance gene. OXA-48-type carbapenemases have been identified mainly from North African countries, the Middle East, Turkey and India, those areas constituting the most important reservoirs; however, occurrence of OXA-48 producers in European countries is now well documented, with some reported hospital outbreaks. Since many OXA-48-like producers do not exhibit resistance to broad-spectrum cephalosporins, or only decreased susceptibility to carbapenems, their recognition and detection can be challenging. Adequate screening and detection methods are therefore required to prevent and control their dissemination.

Keywords: oxacillinases, β-lactamases, carbapenems, class D

Introduction

In the last few years the emergence of carbapenem resistance in Gram-negatives has been observed worldwide, both in non-fermenters (Acinetobacter baumannii and Pseudomonas aeruginosa) and in fermenters (Enterobacteriaceae). This phenomenon is mostly related to the spread of different types of β-lactamases. In Enterobacteriaceae, the main carbapenem-hydrolysing β-lactamases are the Ambler class A β-lactamases (e.g. KPC) and the Ambler class B β-lactamases/metallo-β-lactamases (e.g. IMP, VIM and NDM).1 In addition, the class D β-lactamase OXA-48 (and its variants) possessing weak but significant carbapenemase activity is increasingly reported in Enterobacteriaceae. Recent studies and general epidemiological observations showed that OXA-48-like producers are being increasingly identified in many countries.1

This review aims to summarize the main characteristics of the OXA-48-type carbapenemases, including genetic, enzymatic, microbiological and epidemiological features.

General properties of OXA-48

Class D β-lactamases are not inhibited by clavulanic acid, tazobactam and sulbactam (apart from very few exceptions), whereas their activity may be in vitro inhibited by NaCl.2 Some of these class D β-lactamases hydrolyse carbapenems and are therefore defined as carbapenem-hydrolysing class D β-lactamases (CHDLs).2 This is true for OXA-48, which was first identified from a carbapenem-resistant Klebsiella pneumoniae isolate that had been recovered in Istanbul, Turkey, in 2001.3 That very first OXA-48 producer isolate was multidrug resistant and exhibited a high level of resistance to all β-lactams, including broad-spectrum cephalosporins, cephemycins, monobactams and carbapenems. The identified blaOXA-48 gene was plasmid-located and encoded a β-lactamase weakly related to other class D β-lactamases, sharing only 46%, 36%, 32% and 21% amino acid identity with OXA-10, OXA-23, OXA-40 and OXA-1, respectively.1

Further investigations showed that this OXA-48-producing K. pneumoniae co-expressed several β-lactamases, including the class A extended-spectrum β-lactamase (ESBL) SHV-2a and the narrow-spectrum β-lactamases TEM-1 and OXA-47, and exhibited defects in several outer membrane proteins, leading to its high-level antibiotic resistance pattern.3 Although OXA-48 hydrolyses penicillins at a high level, it hydrolysates carbapenems only at a low level. In addition, it shows very weak activity against expanded-spectrum cephalosporins.1,4 In fact, it hydrolysates cefotaxime very poorly, but does not significantly
The analysis of the crystal structure of OXA-48 revealed that this β-lactamase possessed a structure similar to that of the narrow-spectrum class D β-lactamase OXA-10.6 Interestingly, the carbapenemase activity of OXA-48 is likely resulting from subtle changes in the active site region compared with other class D β-lactamases. The efficient hydrolysis of carbapenems might rely on the rotation of the substrate α-hydroxyethyl group promoted by the nature and conformation of residues located in or close to the β5–β6 loop, which allows the movement of the deacylating water molecule toward the acylated serine residue.6

**Epidemiology of OXA-48 producers**

After the first identification of an OXA-48-producing *K. pneumoniae* from Istanbul, an outbreak of OXA-48-producing *K. pneumoniae* isolates was reported in Istanbul from May 2006 to January 2007.5 Two distinct clones (differing from the index OXA-48 producer) were identified in the same hospital, both producing different ESBL determinants (SHV-12 and CTX-M-15, respectively).5 In addition, the *bla*OXA-48 gene has been identified in *Escherichia coli* and *Citrobacter freundii*, again first in Turkey.6,7 For many years, almost all the reports of OXA-48 producers remained from patients hospitalized in Turkey or from patients with a link to Turkey.8,9

More recently, the identification of the *bla*OXA-48 gene has been reported in many countries, most often from *K. pneumoniae* isolates. It appears that OXA-48 producers have spread in Turkey, the Middle East and North African countries (Table 1 and Figure 1). All those countries can now be considered to be important reservoirs of OXA-48 producers. In Turkey, a series of sporadic cases, but also outbreaks, have been reported during the last 8 years since the first description of OXA-48 in 2004.5,10 In the Middle East, sporadic cases have been recently reported, some of them actually corresponding to identification of the OXA-181 variant (see below). However, there have been OXA-48-producing isolates reported in Lebanon,11,12 Sultanate of Oman,13 Saudi Arabia14 and Kuwait15 (Table 1). In Africa, most of the data are from the northern countries (Morocco, Tunisia, Egypt and Libya), and occurrence of OXA-48 producers has also been reported in Senegal16 and South Africa (Table 1). In Morocco, a nosocomial dissemination of OXA-48-producing *K. pneumoniae*, *Klebsiella oxytoca* and *Enterobacter cloacae* has been reported.17 Notably, the occurrence of some OXA-48-producing *E. cloacae* isolates in France was demonstrated to originate from Morocco.18,19 In Tunisia, OXA-48-producing *K. pneumoniae* isolates have been reported in different hospitals located in different cities.20–22 There are no published data yet from Algeria, but our own observations with patients transferred from that country strongly suggests that it is also a country where OXA-48 producers might be endemic (Figure 1).

Additionally, OXA-48 producers have been identified sporadically in several European countries, including France, Germany, The Netherlands, Italy, Belgium, the UK, Ireland, Slovenia, Switzerland and Spain (Table 1). In countries such as France,
Notably, the recently identified occurrence of true in France and Belgium, and probably in Germany, where from Georgia or in Jordan. Medical tourism involving patients who had been transferred in hospital settings (Table 1 and Figure 1). However, the spread of emergence of OXA-48-producing enterobacterial isolates, at least in the UK, Germany and Belgium, recent studies revealed an emergence of OXA-48-producing enterobacterial isolates, at least in hospital settings (Table 1 and Figure 1). However, the spread of the \(\text{bla}_{\text{OXA-48}}\) gene might be much more important than thought. In fact, the detection of OXA-48-like producers is difficult since the level of acquired resistance to carbapenems may remain quite low (see below).

Interestingly, an emergence of outbreaks related to OXA-48-producing \(K.\ pneumoniae\) is currently observed, involving strains always exhibiting multidrug resistance patterns, being in particular highly resistant to carbapenems. That phenomenon is no longer observed in Turkey only, since hospital outbreaks have been reported in France,\(^{22}\) Belgium,\(^{26}\) Israel,\(^{25}\) Russia\(^{26}\) and The Netherlands.\(^{27}\) The same OXA-48-producing \(K.\ pneumoniae\) of sequence type (ST) 395 was actually identified in Morocco, France and Amsterdam, indicating a clonal dissemination.\(^{28}\)

One of the main sources of concern corresponds to the occurrence OXA-48 producers in the community, often as a consequence of importations from endemic countries, but not systematically. In North Africa, since those countries are probably facing endemic situations, it is likely that OXA-48 producers have spread in the community. This has been exemplified with cases reported in Morocco.\(^{29}\) In Europe, this may be particularly true in France and Belgium, and probably in Germany, where OXA-48-producing isolates have been established already (Figure 1).\(^{30}\) Notably, the recently identified occurrence of OXA-48 producers in Israel was demonstrated to be linked with medical tourism involving patients who had been transferred from Georgia or in Jordan.\(^{21}\)

**Variants of OXA-48**

Since its discovery in 2004, and until very recently, prospective and retrospective studies were always identifying the exact OXA-48 variant. Then the OXA-162 variant (differing by a single amino acid substitution) was identified from \(K.\ pneumoniae\) isolates in Turkey (GenBank ACZ73269) (Figure 2). OXA-162 shares identical hydrolitic activity against \(\beta\)-lactams in general, and carbapenems in particular (L. Poirel, personal data). Recently it has been identified in Germany in various species (\textit{e.g.} \(E.\ coli\), \(C.\ freundii\) and \(R.\ ornithinolitic\)).\(^{32}\)

Then the OXA-163 variant, which exhibits very specific enzymatic characteristics, was identified from Argentinean isolates (see below).

The OXA-181 variant, differing from OXA-48 by four amino acid substitutions, has been identified quite concomitantly by different groups (Figure 2). Interestingly, it has been found to be associated with other carbapenemase genes, such as the \(\text{bla}_{\text{NDM-1}}\) and \(\text{bla}_{\text{VIM-5}}\) genes, in particular in isolates for which a link with the Indian subcontinent could be traced.\(^{33-35}\) The \(\text{bla}_{\text{OXA-181}}\) gene has been identified in multiple clonally unrelated \(K.\ pneumoniae\) isolates in India,\(^{36}\) in \(K.\ pneumoniae\) isolates in The Netherlands,\(^{36}\) New Zealand\(^{37}\) and the Sultanate of Oman;\(^{35}\) in \(C.\ freundii\) and \(P.\ rettgeri\) (L. Poirel, personal data); and in \(E.\ coli\) in India.\(^{36}\) OXA-181 shares the same hydrolytic properties as OXA-48.\(^{35}\) Since the main reservoir of OXA-181 producers seems to correspond to the Indian subcontinent, we believe that their spread worldwide will mirror that of the NDM-1 producers.\(^{38}\)

OXA-204 was recently identified from a series of \(K.\ pneumoniae\) isolates recovered from patients having a link with Algeria or Tunisia. OXA-204 exhibits two amino acid substitutions compared with OXA-48, and preliminary data indicate a substrate profile very similar to that of OXA-48 (L. Poirel, unpublished data) (Figure 2). OXA-232 has been recently identified from \(K.\ pneumoniae\) isolates in France, from patients who had been transferred from Mauritius or India (L. Poirel, A. Paton and P. Nordmann, unpublished data). It exhibits five amino acid substitutions compared with OXA-48, but is just a point mutant derivative of OXA-181 (Figure 2). Again, preliminary data indicate a very similar hydrolysis spectrum for that variant.

**OXA-163, an expanded-spectrum class D \(\beta\)-lactamase**

Even though OXA-48 exhibits a substrate profile including penicillins and carbapenems, but not expanded-spectrum cephalosporins, OXA-163 hydrolyses expanded-spectrum cephalosporins, but very weakly carbapenems.\(^{39}\) Its peculiar substrate profile enables that enzyme to be classified into the group named extended-spectrum OXAs (ES-OXAs),\(^ {2}\) some ES-OXAs being just point-mutant derivatives of narrow-spectrum class D \(\beta\)-lactamases that have been mainly identified in \(P.\ aeruginosa\). OXA-163 is also peculiar in the way that its activity is partially inhibited by clavulanic acid and tazobactam, in contrast to the other class D \(\beta\)-lactamases.\(^ {2}\) It actually exhibits a substrate profile that is similar to that of OXA-18 identified in \(P.\ aeruginosa\).\(^ {40}\) Sequence analysis of the amino acid sequence...
Origin of the OXA-48-like \(\beta\)-lactamases

Even if the progenitors of the widespread \(\text{bla}_{\text{OXA-48}}\), \(\text{bla}_{\text{PER}}\), \(\text{bla}_{\text{IMP}}\), \(\text{bla}_{\text{VIM}}\) and \(\text{bla}_{\text{DHD}}\) carbapenemase genes remain unknown, the origin of some CHDLs has been identified.\(^1\) The waterborne species *Shewanella oneidensis* does possess an intrinsic \(\text{bla}_{\text{OXA-54}}\) gene encoding a \(\beta\)-lactamase that shares 92% amino acid identity with OXA-48.\(^{2,4}\) Recently, *Shewanella xiamenensis* was identified as the progenitor of the \(\text{bla}_{\text{OXA-181}}\) gene, with the exact same sequence identified on the chromosomes of some of the waterborne species.\(^{2,4,5}\) More generally, the *Shewanella* spp. constitute reservoirs of CHDL-encoding genes.\(^6\) If mobilized on mobile genetic structures, they might lead to acquisition of carbapenem resistance in Enterobacteriaceae.

Therefore, considering that the waterborne, environmental and non-human-pathogenic *Shewanella* spp. are the progenitors of the \(\text{bla}_{\text{OXA-48}}\)-like genes, it might be speculated that those genes have been mobilized in the environment. The main hypothesis is that the involved genetic tools, being insertion sequences (see later), have mobilized the chromosomal genes onto plasmids that have subsequently disseminated in clinically relevant species such as in *K. pneumoniae* or *E. coli*. That link between the donor (*Shewanella*) and the recipient might have been direct, or indirect through intermediate reservoirs. This latter hypothesis is exemplified by the recent identification of a plasmid-borne \(\text{bla}_{\text{OXA-48}}\) gene in a *Serratia marcescens* strain recovered from an aquatic environment in Morocco,\(^6,5\) suggesting that *S. marcescens*, being an environmental and opportunistic enterobacterial pathogen, could have played the role of intermediate reservoir, being in close contact with the donor in the environment, but also with clinically relevant enterobacterial species in the human gut.

### Genetic platforms of OXA-48-like \(\beta\)-lactamase-encoding genes

The \(\text{bla}_{\text{OXA-48}}\) gene has been originally identified in association with insertion sequence IS1999 in *K. pneumoniae*, providing promoter sequences responsible for its expression.\(^7\) It has been demonstrated that \(\text{bla}_{\text{OXA-48}}\) was part of a composite transposon named Tn1999 and made of two copies of IS1999 bracketing this gene.\(^6\) In *in vitro* experiments confirmed that Tn1999 was functional, even though transposition occurred at very low frequency in *E. coli* (<10\(^{-7}\)). Then transposon Tn1999.2 was identified from *K. pneumoniae* isolates from Istanbul, differing by the insertion of IS1R.\(^6,7\) In fact, IS1R had targeted the region upstream of \(\text{bla}_{\text{OXA-48}}\), thus enhancing its expression by providing strong promoter sequences. Interestingly, the isolates harbouring the Tn1999.2 structure exhibited higher MICs of carbapenems.
compared with those possessing Tn1999. Very recently a third isofrom of Tn1999 has been identified in an E. coli isolate from Italy, with a second copy of IS1R located downstream of blaOXA-48.48

Interestingly, the blaOXA-181 gene has been identified in a totally different genetic environment, without any IS199 feature in the surrounding sequences. Insertion sequence IScep1 was identified upstream of the blaOXA-181 gene. That insertion sequence is known to be widely responsible for the acquisition of the broad-spectrum β-lactamase blaCTX-M and blaCMY genes. The transposition process mediated by IScep1 is peculiar, since only a single copy can mobilize sequences located at its right-end extremity by recognizing imperfect right inverted repeat sequences (one-ended transposition). In K. pneumoniae KP3 from Oman, the blaOXA-181 gene was identified inside a 3139 bp long IScep1-made transposon named Tn2013, flanked by a 5 bp duplicated sequence being the signature of the transposition. The same association between IScep1 and blaOXA-181 was found in the other OXA-181 producers we have identified (L. Poirel, A. Potron and P. Nordmann, personal data).

Plasmids involved in the spread of OXA-48-like-encoding genes

So far, acquisitions of blaOXA-48-like genes have been identified only in Enterobacteriaceae, but have never been found in other Gram-negative such as A. baumannii or P. aeruginosa, even though other CHDL-encoding genes are identified in these species. This absence of transfer to non-enterobacterial species might be explained by the narrow host range of plasmids bearing the blaOXA-48-like genes. Indeed, the acquired blaOXA-48 gene has always been identified on plasmids. This is also true for the blaOXA-163 and blaOXA-181 genes. Several studies initially reported that the blaOXA-48 gene was located on ∼70 kb plasmids that were self-transferable and did not carry additional resistance determinants. All those observations led us to speculate on the epidemiology of a single plasmid carrying blaOXA-48. That hypothesis has been recently confirmed through the complete sequencing of the blaOXA-48-carrying plasmid pOXA-48a recovered from a K. pneumoniae isolate in 2001 in Istanbul, Turkey. The whole sequence of pOXA-48a was obtained and subsequent analysis revealed that it was a 62.3 kb IncI/M-type plasmid backbone on which the Tn1999 composite transposon had inserted. IncI/M-type plasmids are common in Enterobacteriaceae, and have been identified at the origin of the acquisition of a variety of antibiotic resistance genes. They are broad host range plasmids, being identified in Erwinia spp., Ralstonia spp. and Pseudomonas spp. A single replication module was identified on pOXA-48a, as for other IncLM-type plasmids, and the transfer operon was also very similar to those of that plasmid group. The conjugation rate of plasmid pOXA-48a among enterobacterial species was found to be quite high, at $3.3 \times 10^{-5}$. Molecular investigations showed that plasmid pOXA-48a was identified in all OXA-48-producing isolates recovered from many countries. This observation is noteworthy since it indicates that the current spread of OXA-48 producers is related to the spread of a single plasmid among different enterobacterial isolates.

Few studies have identified OXA-181-producing isolates so far. However, it has been shown that the blaOXA-181 gene might be found on different plasmid scaffolds. In K. pneumoniae KP3 from Oman, the blaOXA-181 gene was found on a small (7605 bp) ColE2-type plasmid, which was not self-transferable (but mobilizable), did not carry additional resistance determinants and was of broad host range. In a C. freundii isolate from France co-harbouring a blaNDM-1-positive plasmid, the blaOXA-181 gene was located onto a 83577 bp IncF-type plasmid that was self-conjugative, though at a low frequency, and mobilizable.

Detection of OXA-48-like enzymes

One of the major concerns for controlling the spread of OXA-48-like producers is the absence of phenotypical tests that could contribute to their easy recognition. In particular, a search for carbapenemase production in any enterobacterial isolates with a slight decrease of susceptibility to carbapenems is important, because there is currently a paucity of clinical experience for treating infections due to carbapenemase producers, and also very limited knowledge about the possibility to select in vivo mutants with increased levels of resistance to carbapenems through additional mechanisms.

There are actually two main concerns: (i) recognizing the production of OXA-48 in those isolates recovered from infections; and (ii) isolating the OXA-48 producers by using a reliable screening process. First, based on the literature and our own experience, we propose that production of a carbapenemase should be suspected only for enterobacterial isolates with MIC values of ertapenem ≥0.5 mg/L, or imipenem or meropenem ≥1 mg/L. What is noteworthy with OXA-48 producers is that very different β-lactam resistance patterns can be observed, with some isolates being still susceptible to broad-spectrum cephalosporins and carbapenems, some being still susceptible to broad-spectrum cephalosporins but resistant to carbapenems, and some being resistant to broad-spectrum cephalosporins and carbapenems (Figure 3). That means that suspicion of OXA-48 production is very often challenging.

Recently, several countries adopted new guidelines recommending the screening of patients transferred from foreign hospitals or patients returning from travels in foreign countries known to be endemic for multiresistant bacteria. Indeed, the prevention of spread of carbapenemase producers relies on the early and accurate detection of carriers. Those recommendations included screening procedures for colonization, by using, for instance, commercially available ESBL-targeting media or carbapenem-supplemented media. Several studies report using media containing imipenem at 1–2 mg/L, which may be too high for efficient detection of carbapenemase producers with low-level resistance, which is often the case for OXA-48/NDM-1 producers. In addition, the ESBL-targeting media approach fails to detect OXA-48-like producers (except OXA-163 producers), which do not produce any ESBL determinants, since they remain susceptible to broad-spectrum cephalosporins.

An enrichment procedure including an overnight culture in broth supplemented with ertapenem before plating on Drigalski agar medium with ertapenem and imipenem Etest strips has
inhibits a wide range of β-lactam hydrolytic activity could be inhibited by the NXL104 CHDL producers in particular. Recent data have shown that studies did not yet evaluate the reliability for OXA-48 production of carbapenemases, even though those approaches was recently highlighted by Glupczynski et al. who suggested that a high level of resistance of OXA-48 producers to the β-lactam temocillin might be presumptive of OXA-48 production. However, the efficacy of carbapenemases for treating infections due to carbapenemase producers with low-level resistance or susceptibility to several carbapenems remains debatable since imipenem-containing therapy failed to treat several OXA-48 infections. Finally, the molecular-based techniques (essentially PCR) remain the gold standard techniques for the identification of isolates carrying blaoxa-48-like genes. Primers OXA-48A (5′-TTGGT GGCAATCGATTGCGG-3′) and OXA-48B (5′-GAGCAGCTTCTTGTGTAGTG GC-3′) allow the amplification of all known blaoxa-48-like genes identified so far. In order to detect the clinically relevant carbapenemase activity, a multiplex PCR system in which screening of the blaoxa-48-like genes was included has been developed. A blaoxa-48-specific real-time TaqMan PCR method has also been developed in order to speed the process of its molecular identification. Finally, a microarray method has been developed that includes the detection of the blaoxa-48-like genes. As underlined, sequencing of the blaoxa-48-like genes is necessary to differentiate between the different variants, and this may impact the interpretation of the positivity when considering that some of those variants do not possess a significant carbapenemase activity.

Clinical consequences

The clinical consequences related to the spread of OXA-48-type producers may be quite important, because many of those producers are classified as susceptible to carbapenems according to the EUCAST or CLSI guidelines. The current recommendations proposed by those guidelines are to report susceptibility to carbapenems as found, whether or not the isolates produce a carbapenemase. It has been recently reported that imipenem could be successfully used to treat a bacteraemia due to an imipenem-susceptible but OXA-48-producing K. pneumoniae. The efficacy of carbapenemases for treating infections due to carbapenemase producers with low-level resistance or susceptibility to several carbapenems remains debatable since imipenem-containing therapy failed to treat several OXA-48 infections. For those OXA-48-like-producing isolates in which there is no ESBL association, broad-spectrum cephalosporins such as ceftazidime may be theoretically used since those molecules are weakly or not hydrolysed by those enzymes (except OXA-163). However, very few clinical data are available to support their use, except one study in which we showed success in treating a newborn infection caused by an ESBL-negative and OXA-48-producing K. pneumoniae with a combination of ceftaxime and amikacin.
The β-lactamase inhibitor NXL104 should represent an interesting therapeutic option, since it possesses potent activity against class A, B and D β-lactamases, meaning that it may inhibit both ESBL and OXA-48 in those isolates combining both mechanisms. A recent study reported that NXL104 is able to significantly reduce the MIC values of imipenem, ceftepime and ceftazidime for OXA-48 producers co-producing an ESBL, leading to susceptibility for all the tested strains. The same results were reported for the combination with cefotaxime or ceftaroline and NXL104.

Recently, an experimental model of induced peritonitis has been developed in mice, using an ESBL-negative and OXA-48-producing K. pneumoniae strain. That strain exhibited decreased susceptibility to carbapenems and susceptibility to broad-spectrum cephalosporins. Ceftazidime was shown to be an efficient therapy, whereas ertapenem and imipenem were not. That observation raises serious concerns about the efficacy of carbapenems for treating patients infected with OXA-48-type-producing Enterobacteriaceae, regardless of the observed level of resistance to carbapenems.

Colistin and tigecycline are most likely to be active in vitro against OXA-48 producers, but resistance to these molecules has been reported among OXA-48-producing isolates. Fosfomycin might be useful as a last-resort option as part of association regimens because of the high potential for emergence of resistance. Given that OXA-48 producers exhibit variable resistance profiles, selection of the appropriate therapy should be made on a case-by-case basis. Interestingly, by comparing ESBL-positive and –negative OXA-48 producers, we observed that the ESBL-negative OXA-48 producers were significantly more susceptible to aminoglycosides and fluoroquinolones than the ESBL-positive OXA-48 producers (P. Nordmann, personal data).

Conclusions

Since the beginning of the 2000s, carbapenemases of the Ambler class A KPC type or class B type, including IMP-, VIM- and NDM-like enzymes, were considered to be the most important carbapenemases in Enterobacteriaceae, since (i) their hydrolytic activity included not only carbapenems, but also broad-spectrum cephalosporins; (ii) their carbapenemase activities were significant; and (iii) their corresponding genes have been identified worldwide. In contrast, OXA-48 was not really considered so much problematic, considering that (i) its hydrolytic spectrum does not include broad-spectrum cephalosporins; (ii) its carbapenemase activity is somewhat low compared with class A and class B enzymes; and (iii) the spread of the blaOXA-48 gene was supposed to be limited to Turkey only for many years.

The difficulties linked to the detection of OXA-48 producers have played a significant role in their spread, which has been somehow silent. Indeed, the fact that expression of the blaOXA-48 gene only confers reduced susceptibility to carbapenems does not facilitate recognition of the OXA-48 producers. Also, since a significant proportion of OXA-48 producers does not co-express ESBLs, the observed susceptibility to broad-spectrum cephalosporins does not favour their recognition or even suspicion.

Is the current emergence of OXA-48 producers resulting from antibiotic selective pressure, and in particular to the overuse or misuse of carbapenems? That is an issue that is difficult to speculate about. Interestingly, the current spread of the blaOXA-48 gene is largely the consequence of the spread of a single epidemic plasmid that does not carry other resistance determinants. The absence of other resistance determinants on this plasmid suggests that other antibiotic families likely did not play a role in co-selection, as opposed to what can be hypothesized for many other carbapenemase enzymes, which are often physically associated with other resistance genes, and in particular, genes encoding resistance to aminoglycosides.

Despite the fact that blaOXA-48 is part of a functional transposon, its dissemination actually corresponds to the dissemination of its plasmid support and not of the transposon itself. Further investigations are required to better understand the reasons for such a successful spread. Another interesting observation in relation to the genetic context of the blaOXA-48 genes corresponds to the fact that all have been identified in association with insertion sequences, whereas most of the class D β-lactamase genes are very often identified as gene cassettes located in class 1 integron structures. Despite being closely related in terms of nucleotide sequence, the blaOXA-48 and blaOXA-181 genes have always been identified in distinct genetic contexts (an IS1999- or an ISEC1-made transposon), suggesting that those two genes did not evolve from each other through mutations, but rather occurred through two separate events originally corresponding to mobilizations of two different genes from two Shewanella donor strains.

From a clinical point of view, further studies need to be performed in order to evaluate whether carbapenem-based treatments could be used for treating infections caused by OXA-48 producers remaining in vitro in the susceptibility ranges, and also whether treatments based on broad-spectrum cephalosporins could be safely used when facing infections caused by ESBL-negative OXA-48 producers. Currently, OXA-48-like enzymes represent a potential source of clinical failure for many β-lactams in Enterobacteriaceae. The lack of their detection may enhance their hidden and rapid spread among clinical isolates. In addition, since OXA-48 producers are mostly E. coli and K. pneumoniae, it is possible that non-specialized laboratory or infectious disease practitioners may let the outbreak continue because of lack of recognition and thus adequate management of those particular isolates. Unfortunately we foresee further and extensive spread (likely uncontrollable), at least in European countries as a consequence of a tight relationship with many North African and Middle East countries and Turkey.

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None to declare.
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