Fluoroquinolone resistance developed during ciprofloxacin therapy and was associated with a gyrA mutation. These data show for the first time the ability of *A. radioresistens* to develop fluoroquinolone resistance during therapy. Additionally, ISAcrA1 has the potential to spread OXA-23-mediated carbapenem resistance in *A. radioresistens*.

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### Transparency declarations

None to declare.

### Supplementary data

Tables S1 and S2 are available as Supplementary data at JAC Online (http://jac.oxfordjournals.org/).

### References


**Acquisition of plasmid-borne *bla*IMP-19 gene by a VIM-1-positive *Pseudomonas aeruginosa* of the sequence type 235 epidemic lineage**

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**Keywords:** metallo-β-lactamases, integrons, multidrug resistance

Sir,

Acquired metallo-β-lactamases (MBLs) are the most common acquired carbapenemases in *Pseudomonas aeruginosa*. They confer a broad-spectrum β-lactam resistance profile, including resistance to the antipseudomonal penicillins, cephalosporins and carbapenems, that is not antagonized by the available β-lactamase inhibitors. Most MBL-producing strains exhibit a multidrug resistance profile, also including resistance to non-β-lactams, due to the accumulation of additional resistance determinants.1

Although several types of acquired MBLs have been detected in *P. aeruginosa*, the IMP- and VIM-type enzymes are currently the most widespread.1 A number of genes encoding these enzymes (e.g. *bla*VIM-1, *bla*VIM-2, *bla*VIM-4 and *bla*IMP-1) have become associated with high-risk clones, such as clonal complex (CC) 235 and CC111, which has promoted their dissemination in Europe and other continents.2

IMP-19 is an IMP allelic variant that was originally detected in *Enterobacter cloacae* (GenBank/EMBL accession no. AB201264) and *P. aeruginosa* (GenBank/EMBL accession no. AB184976) from Japan and, subsequently, in an *Aeromonas caviae* isolated in France.3 More recently, it has been reported in *Achromobacter xylosoxidans* and *Acinetobacter* spp. isolates from Japan.4,5

Here, we report on the first detection of the *bla*IMP-19 gene in Italy, in a multidrug-resistant (MDR) *P. aeruginosa* clinical isolate that also produced VIM-1 and belonged to the CC235 epidemic lineage.
*P. aeruginosa* MO-11c33 was isolated in June 2011 from the urine of an elderly (81 years old) female inpatient in Modena University Hospital (northern Italy). The patient, resident in a long-term care facility and affected by senile dementia requiring a permanent urinary catheter and nasogastric tube, had been admitted with diagnosis of sepsis and was treated with meropenem for 10 days before isolation of MO-11c33 from the urine. Clinical records reported two additional hospital admissions for septic episodes during the previous 2 months, which were successfully treated with meropenem. MO-11c33 was considered a colonizer and no specific antipseudomonal treatment was given following its isolation.

Susceptibility testing, carried out using broth microdilution and interpreted according to EUCAST criteria (http://www.eucast.org/clinical_breakpoints/), revealed an MDR phenotype including resistance to all antipseudomonal β-lactams (MIC values: piperacillin/tazobactam, 128 mg/L; ceftazidime, >128 mg/L; cefepime, >128 mg/L; imipenem, 64 mg/L; meropenem, >128 mg/L), gentamicin (MIC, 128 mg/L and ciprofloxacin (MIC, 8 mg/L) while susceptibility to amikacin (MIC, 8 mg/L) and colistin (MIC, 0.25 mg/L) was retained.

The isolate tested positive for MBL production with the MBL Etest (bioMérieux, Marcy l’Étoile, France). Multiplex PCR revealed the presence of a *bla*<sub>VIM</sub> and a *bla*<sub>IMP</sub>-19 allele, which were identified as *bla*<sub>VIM</sub>-1 and *bla*<sub>IMP</sub>-19 by sequencing of PCR amplicons of the entire coding sequences. Multilocus sequence typing (http://pubmlst.org/paeruginosa/) revealed that MO-11c33 belonged to sequence type 235 (the founder of CC235), a high-risk clone involved in the dissemination of *bla*<sub>VIM</sub>-1 and other resistance determinants in Italy and elsewhere.

Analysis of the genetic context of the two MBL genes in MO-11c33, carried out by S1 nuclease and ICeu-I mapping using 16S rRNA-, *bla*<sub>IMP</sub>-19- and *bla*<sub>VIM</sub>-1-specific probes, revealed that *bla*<sub>VIM</sub>-1 was chromosomally located, while the *bla*<sub>IMP</sub>-19 gene was carried on a ≏120 kb plasmid (data not shown). Conjuga­tion and electrotransformation experiments were not successful in transferring the plasmid to an *Escherichia coli* or *P. aeruginosa* host. Analysis of the genetic context of the two MBL genes by PCR mapping and sequencing of the flanking regions, using *bla*<sub>IMP</sub>-19- and *bla*<sub>VIM</sub>-1-specific primers and external primers (Figure 1), revealed that *bla*<sub>VIM</sub>-1 was associated with an In70.2 integron platform identical to that of the *VIM*-1-producing *P. aeruginosa* index strain and of most other *VIM*-1-producing isolates circulating in Italy but different from those of other IMP-19-producing isolates (Figure 1).

*bla*<sub>IMP</sub>-19 was associated with an In804 integron platform apparently identical to that previously detected in an *A. xylosoxidans* isolate from Japan, but different from those of other IMP-19-producing isolates (Figure 1). In In804 from MO-11c33, a copy of ISPa7, which is typical of In70.2 but present also in other integrons, was inserted upstream of the integrase gene and remnants of Tn402-like transposition modules (tnIB gene) were also detected downstream of the 3′-conserved segment (GenBank/EMBL accession no. JX421697).

To the best of our knowledge, this is the first report of *bla*<sub>IMP</sub>-19 from Italy and the second from Europe, since 2007. It is also the first report of a *P. aeruginosa* isolate coproducing...
IMP- and VIM-type MBLs. In this case, the source of blaIMP-19 remains unclear, but given the patient’s history, a direct cross-border origin could be excluded. Since the blaIMP-19-carrying plasmid could not be mobilized, its contribution in terms of carriage of additional resistance genes could not be assessed and further investigations will be necessary to understand the potential advantage conferred by acquisition of this plasmid by the VIM-producing strain. Nevertheless, the detection of blaIMP-19 in a P. aeruginosa strain of the CC235 epidemic lineage is of concern, since this could promote blaIMP-19 dissemination in the clinical setting.

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**References**


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**Two cases of macrolide resistance in Mycoplasma pneumoniae acquired during the treatment period**

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**Keywords:** 23S rRNA, azithromycin, clarithromycin

Sir,

*Mycoplasma pneumoniae* is a common cause of upper and lower respiratory tract infections, especially in children and young adults. *M. pneumoniae* infections can be treated with macrolides, which are generally considered to be the first-choice antibiotics for children. However, the prevalence of macrolide-resistant *M. pneumoniae* has gradually increased worldwide and has reached a rate of 90% in China.1 In Japan, it has been reported that the prevalence of macrolide-resistant *M. pneumoniae* isolates reached 65.5% in 2010 and 89.5% in 2011.2 *M. pneumoniae* resistance to macrolides is caused by point mutations in domain V of the 23S rRNA gene that interfere with the binding of macrolides to rRNA.3, 4 An A-to-G transition at position 2063 (A2063G) is the most frequently detected mutation among resistant strains, closely followed by an A-to-G transition at position 2064 (A2064G).5, 6 Although resistant strains of *M. pneumoniae* have been generated in vitro by selection with subinhibitory concentrations of macrolides,3, 4 in humans there is only one report of the acquisition of a macrolide-resistance mutation during the course of infection.5 We report herein two cases in which macrolide-resistant *M. pneumoniae* isolates are thought to have emerged during treatment with a macrolide.

Patient 1 is an 8-year-old child (Table 1). He visited the Yamanobe Paediatric Clinic with a 1 day history of fever of up to 39.4°C and a mild cough in November 2011. His temperature was 37.4°C. After taking a pharyngeal swab sample, he was treated with azithromycin (10 mg/kg/day) for 3 days. His temperature rapidly decreased 24 h later. Because of the persistent dry cough without fever, he visited the same clinic at 6 days after starting the antibiotic treatment and a second pharyngeal swab sample was taken. The patient’s guardian gave written informed consent for the publication of these data.

Patient 2 is a 5-year-old child (Table 1). She was brought to the Yamanobe Paediatric Clinic in January 2012, with a 4 day history of fever up to 39.8°C and coughing. On initial physical examination, her temperature was 38.7°C and rales were