



LETTER TO THE EDITOR

First report of a clinical isolate of *bla*_{OXA-48}- carbapenemase producing *Raoultella ornithinolytica* in South America

Primer reporte de un aislado clínico de *Raoultella ornithinolytica* productora de carbapenemasa *bla*_{OXA-48} en América del Sur

Dear Editor:

Raoultella spp., a bacterium named after Didier Raoult, is a Gram-negative belonging to the *Enterobacteriaceae* family. There have been described 4 species: *Raoultella planticola*, *Raoultella ornithinolytica*, *Raoultella terrigena*, and *Raoultella electrica*, all species are inhabitants of soil and plants. Recently, *R. planticola* and *R. ornithinolytica* have been associated with human infections and resistance to different antibiotics including carbapenems⁵. In this report, we describe an infection caused by *bla*_{OXA-48} producing *R. ornithinolytica*. A 64-year-old male patient was admitted into a 700-bed hospital in Quito, in order to restore intestinal transit after ileocecal resection. Two days after admission the patient presented sepsis characterized by hypotension, fever, and leukocytosis. The patient was directed to intensive care unit where he was treated with Ampicillin/sulbactam for 10 days, subsequent blood cultures were negative however *Escherichia coli* was recovered from tracheal aspirate. The patient was submitted to the surgical department to continue treatment and 10 days later he presented a surgical site infection, *R. planticola* (resistant to imipenem and piperacillin/tazobactam but susceptible to third generation cephalosporins and ciprofloxacin) was isolated and detected using a Vitek[®] compact 2 system (Biomérieux, France), CARD AST 272 (Table 1). The patient

was treated with ciprofloxacin 500 mg i.v. q12h and recovered satisfactorily.

Isolate analysis using DNA sequences of *rpoB* and 16S rRNA genes and MALDI-TOF Vitek MS system, (bioMérieux) indicated that the isolate was *R. ornithinolytica* (99.9% identification score). The RAPIDEC CARBA NP[®] (bioMérieux) assay was positive and polymerase chain reaction (PCR) amplifying the *bla*_{OXA-48} associated with Tn1999 was performed³. The amplicon was cleaned, using Wizard[®] SV gel, and sequenced in Macrogen (South Korea); nucleotide sequences showed the presence of *bla*_{OXA-48} gene (accession no.: MH507508) and Tn1999 (accession no.: MK359485). The amplicon showed 99% nucleotide identity to a Tn1999 harboring *bla*_{OXA-48} previously described in *Klebsiella pneumoniae* (accession no.: JN626286). We were unable to determine the plasmid incompatibility group (conjugation using *E. coli* J53 as the recipient was unsuccessful), nor could we establish whether the patient was colonized by *R. ornithinolytica* before the surgery. To the best of our knowledge, this is the first description of a clinical isolate of *R. ornithinolytica* harboring *bla*_{OXA-48} in Ecuador and possibly in South America. Nevertheless, *bla*_{OXA-48} genes in *Enterobacteriaceae* have been described in Latin American and Caribbean countries². In Ecuador, a *bla*_{OXA-48}-like gene was found previously in *K. pneumoniae* (accession number KY609322.1). The *bla*_{OXA-48}-like gene has been found associated with 5 isoforms of Tn1999 in *Enterobacteriaceae*⁴; interestingly, an *R. ornithinolytica* strain containing *bla*_{OXA-48} associated with Tn1999.2 was detected in Lebanon¹. The genetic closeness of *Raoultella* spp. and *Klebsiella* spp., may lead to misidentification using biochemical tests e.g., Vitek 2 system, the introduction of *rpoB* and 16S rRNA genes analysis and the new technology based in MALDI-TOF MS allowed us a correct identification to species level of *Raoultella* spp. Thus, our results underline the accuracy of MALDI-TOF MS in *R. ornithinolytica* identification. This report was approved by the institutional human ethics committee Cod: 02-01-2018-003.

Table 1 Antibiotic sensitivity tests, carbapenemase assays and carbapenemase gene detection in an Ecuadorian *Raoultella ornithinolytica* isolate

Minimum inhibitory concentration (µg/ml) using vitek 2 system										CIM	Rapidec CarbaNP	PCR and sequencing	
Amp/Sul	Pip/Taz	Cfz	Ctx	Imi	Mer	Cip	Ami	Col				Positive	Positive
>32	>128	<1	<1	8	2	<0.25	≤2	≤0.5				<i>bla</i> _{OXA-48}	Tn1999

Amp/Sul: Ampicillin-sulbactam, Pip/Taz: Piperacillin-tazobactam, Cfz: Cefazidime, Ctx: Cefotaxime, Imi: Imipenem, Mer: Meropenem, Cip: Ciprofloxacin, Ami: Amikacin, Col: Colistin. CIM: Carbapenem inactivation method. MGE: Mobile genetic element. PCR: polymerase chain reaction.

Ethical approval

This study was approved by Ethics Committee in Humans of Carlos Andrade Marín hospital Cod: 02-01-2018-003.

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Contributions

JR and GT: conception, design of the study and drafting the article. EV, FV, EP, NC, ME: acquisition of data, analysis and interpretation. BN and GT revising critically for important intellectual content of article. GT: final approval of the version to be submitted.

Conflict of interest

The authors declare that they have no conflict of interest.

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Presentación del sitio web de la Red Nacional de Identificación Microbiológica por Espectrometría de Masas. Manual para la interpretación de resultados de MALDI-TOF MS



Presentation of the National Network for Microbiological Identification by Mass Spectrometry website. Guide for the interpretation of MALDI-TOF MS results

Sr. Editor:

En las últimas décadas, la tecnología MALDI-TOF, basada en espectrometría de masas, ha demostrado su potencial para la rápida identificación de microorganismos patógenos mediante el análisis del perfil de proteínas ribosomales⁴. Actualmente existen dos plataformas comerciales para el

diagnóstico clínico en humanos: MicroFlex[®] LT (Bruker Daltonics) y Vitek[®] MS (bioMérieux). Estas plataformas fueron aprobadas por la Food and Drug Administration en 2009; pero dicha validación incluyó solo algunos grupos taxonómicos de importancia clínica^{5,6}.

En Argentina, 20 instituciones de salud han incorporado la técnica de MALDI-TOF al laboratorio clínico. Algunos constituyen laboratorios nacionales de referencia (LNR) y han documentado la evaluación de la metodología para la identificación de bacterias y hongos, con un enfoque taxonómico polifásico^{1–3}. Es por ello que desde 2013, se ha recopilado información detallada sobre más de 2.000 microorganismos; también se han desarrollado bases de datos complementarias con espectros proteicos de microorganismos que no están incluidos en las bases comerciales o están escasamente representados, y espectros de variantes biológicas regionales.

La creación e incorporación de estas bases de datos nacionales han optimizado el desempeño de las plataformas comerciales. Consecuentemente, surgió la