'Raoultibacter timonensis' gen. nov., sp. nov., a new bacterium isolated from the human gut of a Pygmy woman

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Abstract

We report the main characteristics of a new species '*Raoultibacter timonensis*' strain Marseille-P3277 (CSUR P3277), a new member of the genus *Raoultibacter*, that was isolated from a stool sample of a healthy 11-year-old Pygmy woman. © 2017 The Authors. Published by Elsevier Ltd on behalf of European Society of Clinical Microbiology and Infectious Diseases.

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After obtaining approval under the number 09-022 from the ethics committee of the Institut Federatif de Recherche IFR48 (Marseille, France), stool samples were collected in 2015 and

analysed as part of the project aiming to describe the human gut microbiome by culturomics [1].

Blood-culture bottles, supplemented with 5% sheep blood and 5% filtered rumen, were used for our sample culture. Ahead of inoculation, samples were diluted with PBS. Bacterial growth was assessed for 30 days at 37°C. '*Raoultibacter timonensis*' was isolated on 5% blood-enriched Columbia agar (bioMérieux, Marcy l'Etoile, France). Colonies were smooth with a mean diameter of 0.2–0.4 mm. Strain Marseille-P3277 cells are cocci, Gram-negative, catalase and oxidase negative with an average

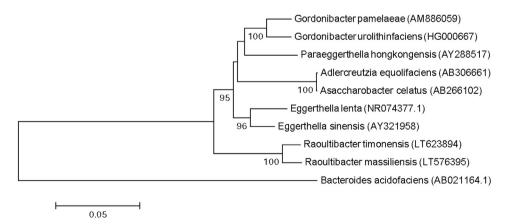


FIG. 1. This phylogenetic tree shows the position of '*Raoultibacter timonensis*' strain Marseille-P3277 among the phylogenetically closest species. Multiple sequence alignment was performed using CLUSTALW and phylogenetic inferences were made using the maximum-likelihood method by the MEGA software. Bootstrap values are shown on the nodes after 500 repeats and values <90% were eliminated. The scale bar indicates a 5% nucleotide sequence divergence. length of 1-2 µm. Colonies were not identified by our systematic matrix-assisted laser desorption/ionization time-of-flight mass spectrometry (MALDI-TOF MS) screening on a Microflex spectrometer (Bruker Daltonics, Bremen, Germany) [2]. Hence, 16S rRNA gene sequencing was performed using fD1-rP2 primers (Eurogentec, Seraing, Belgium) as previously described by a 3130-XL sequencer (Applied Biosciences, Saint Aubin, France) [3].

'Raoultibacter timonensis' exhibited a 97.96% sequence identity with Raoultibacter massiliensis strain P2849 (LT576395), the phylogenetically closest species, published with standing nomenclature (Fig. 1). Hence, strain Marseille-P3277 can be classified as a new species because it exhibited a 16S rRNA gene sequence similarity <98.65% with its phylogenetically closest species [4,5]. We propose the creation of the new species named 'Raoultibacter timonensis' (ti.mo.nen'sis. N.L. masc. adj. timonensis from the name of the Hôpital de la Timone, where strain Marseille-P3277 was isolated). Strain Marseille-P3277 is the type strain of the new species 'Raoultibacter timonensis'.

MALDI-TOF MS spectrum accession number. The MALDI-TOF MS spectrum of *R. timonensis* is available online (http://www.mediterranee-infection.com/article.php?laref=256 &titre=urms-database).

Nucleotide sequence accession number. The 16S rRNA gene sequence was deposited in GenBank under Accession number LT623894.

Deposit in a culture collection. Strain Marseille-P3277 was deposited in the Collection de Souches de l'Unité des Rickettsies (CSUR, WDCM 875) under number P3277.

Transparency declaration

None to declare.

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References

- Lagier J-C, Hugon P, Khelaifia S, Fournier PE, La Scola B, Raoult D. The rebirth of culture in microbiology through the example of culturomics to study human gut microbiota. Clin Microbiol Rev 2015;28: 237–64.
- [2] Seng P, Abat C, Rolain JM, Colson P, Lagier J-C, Gouriet F, et al. Identification of rare pathogenic bacteria in a clinical microbiology laboratory: impact of matrix-assisted laser desorption ionization-time of flight mass spectrometry. J Clin Microbiol 2013;51:2182–94.
- [3] Drancourt M, Bollet C, Carlioz A, Martelin R, Gayral JP, Raoult D. 16S ribosomal DNA sequence analysis of a large collection of environmental and clinical unidentifiable bacterial isolates. J Clin Microbiol 2000;38: 3623–30.
- [4] Kim M, Oh H-S, Park S-C, Chun J. Towards a taxonomic coherence between average nucleotide identity and 16S rRNA gene sequence similarity for species demarcation of prokaryotes. Int J Syst Evol Microbiol 2014;64:346–51.
- [5] Traore SI, Yasir M, Azhar EI, Bibi F, Bittar F, Jiman-Fatani AA, et al. "Raoultibacter massiliensis" gen. nov., sp. nov., a new bacterium isolated from the human gut of a Saudi Bedouin. New Microbes New Infect 2016;14:1–3.