NEW SPECIES

Christensenella massiliensis, a new bacterial species isolated from the human gut

S. Ndongo[1], S. Khelaifia[1], P.-E. Fournier[1] and D. Raoult[1,2]

1) *Unité de Recherche sur les Maladies Infectieuses et Tropicales Emergentes, CNRS (UMR 7278), IRD (198), INSERM (U1095), AMU (UM63), Marseille, France and* 2) *Special Infectious Agents Unit, King Fahd Medical Research Center, King Abdulaziz University, Jeddah, Saudi Arabia*

Abstract

We describe the main characteristics of *Christensenella massiliensis,* strain Marseille-P2438^T (CSUR P2438), isolated from a stool specimen of a 66-year-old patient.

Keywords: *Christensenella massiliensis*, culturomics, gut microbiota, taxonogenomics, taxonomy
Original Submission: 26 April 2016; **Revised Submission:** 29 April 2016; **Accepted:** 29 April 2016
Article published online: 4 May 2016

Corresponding author: D. Raoult, URMITE, CNRS (UMR 7278), IRD (198), INSERM (U1095), AMU (UM63), Faculté de Médecine, Aix-Marseille Université, 27 Boulevard Jean Moulin, 13385 Marseille Cedex 5, France
E-mail: didier.raoult@gmail.com

Here we present an early description of a new bacterium isolated as part of culturomics study of the human gut microbiota [1] from the stool specimen of a 66-year-old patient hospitalized in November 2015 at the Timone Hospital in Marseilles, France. Approval of the study by the local ethics committee of IFR48 (Marseille, France) was obtained beforehand under agreement 09-022. After obtaining the consent signed by the patient, the stool sample was directly preincubated for 7 days in an anaerobic blood bottle culture supplemented with 5 mL of rumen fluid. Then isolated colonies of the strain were obtained by subculturing on 5% sheep's blood agar (bioMérieux, Marcy l'Etoile, France) in anaerobic conditions generated by AnaeroGen (bioMérieux) after 4-day incubation. Our systematic matrix-assisted laser desorption/ionization time-of-flight mass spectrometry (MALDI-TOF MS) screening on a MicroFlex spectrometer (Bruker Daltonics, Leipzig, Germany) [2] was unable to identify this strain.

The strain Marseille-P2438 features Gram-negative bacilli which was strict anaerobic, nonmotile and non–spore forming (0.3–0.5 × 1.2–1.5 μm). It presents no catalase and oxidase activity. The colonies are yellow-white, circular and about 0.1 to 0.2 mm in diameter after 4-day incubation on 5% sheep's blood agar. Optimal growth occurs at 37°C, pH 7 and 0.5% NaCl. Using a 3130-XL sequencer (Applied Biosciences, Saint Aubin, France), the 16S rRNA gene was sequenced using the universal primers fD1 and rp2 (Eurogentec, Angers, France), as previously described [3]. Strain Marseille-P2438 (GenBank accession no. LT161898) exhibited a 97.5% sequence identity with *Christensenella minuta* strain YIT 12065^T (GenBank accession no. NR112900), the phylogenetically closest species with standing in nomenclature (Fig. 1), which putatively classifies it as a member within the genus *Christensenella* in the Firmicutes phylum [4].

C. minuta strain YIT 12065T^T is strictly anaerobic, Gram-negative, nonmotile and non–spore forming, and are short, straight rods isolated from human faeces. Catalase and oxidase activities for *C. minuta* were negative. To date, it is the first species of *Christensenella* genus published with a validated name [4].

Strain Marseille-P2438 shows a 16S rRNA gene sequence divergence of >1.3% with its phylogenetically closest species with a validly published name with standing in nomenclature [5]. On the basis of these results, strain Marseille-P2438^T is proposed as a novel species within the genus *Christensenella,* namely *Christensenella massiliensis* sp. nov.

http://dx.doi.org/10.1016/j.nmni.2016.04.014

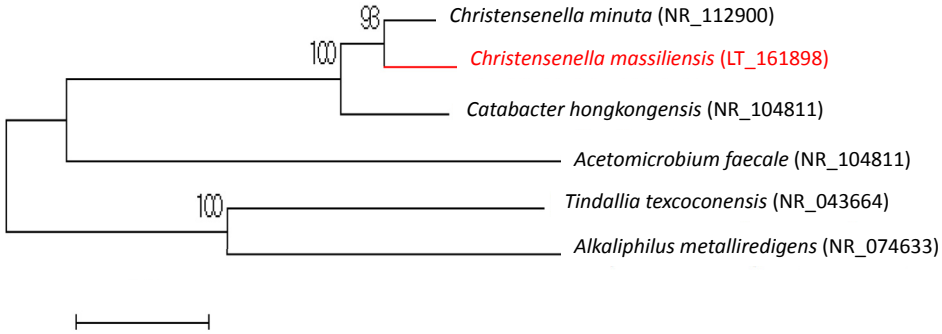

FIG. 1. Phylogenetic tree showing position of *Christensenella massiliensis* strain Marseille-P2438^T relative to other phylogenetically close members of family Christensenellaceae. GenBank accession numbers are indicated in parentheses. Sequences were aligned using CLUSTALW, and phylogenetic inferences were obtained using maximum-likelihood method within MEGA software. Numbers at nodes are percentages of bootstrap values obtained by repeating analysis 500 times to generate majority consensus tree. Only values greater than 95% were displayed. Scale bar = 2% nucleotide sequence divergence.

MALDI-TOF MS spectrum accession number

The MALDI-TOF MS spectrum of *Christensenella massiliensis* is available online (http://www.mediterraneeinfection.com/article.php?laref=256&titre=urms-database).

Nucleotide sequence accession number

The 16S rRNA gene sequence was deposited in GenBank under accession number LT161898.

Deposit in a culture collection

Strain Marseille-P2438^T was deposited in the Collection de Souches de l'Unité des Rickettsies (CSUR) under number P2438.

Acknowledgement

This study was funded by the Fondation Méditerranée Infection.

Conflict of Interest

None declared.

References

[1] Lagier JC, 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 JC, 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] Morotomi M, Nagai F, Watanabe Y. Description of *Christensenella minuta* gen. nov., sp. nov., isolated from human faeces, which forms a distinct branch in the order Clostridiales, and proposal of Christensenellaceae fam. nov. Int J Syst Evol Microbiol 2012;62:144–9.
[5] Kim M, Oh HS, Park SC, 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.