'Lactobacillus raoultii' sp. nov., a new bacterium isolated from the vaginal flora of a woman with bacterial vaginosis

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Abstract

We report the isolation of a new bacterium species, *'Lactobacillus raoultii'* strain Marseille P4006 (CSUR P4006), isolated from a vaginal sample of a 45-year-old woman with bacterial vaginosis.

Keywords: Bacterial vaginosis, culturomics, emerging bacteria, human microbiota, *Lactobacillus raoultii*, vaginal microbiota
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In 2017, as part of the of the human microbiome description by culturomics, a vaginal swab sample was taken from a 45-year-old woman with bacterial vaginosis [1]. An approval (number 09-022) was obtained from the ethics committee of the Institut Fédératif de Recherche IFR48 (Marseille, France) along with signed informed consent from the patient. The vaginal swab was preincubated in a customized medium under anaerobic conditions at 37°C. The customized medium comprised tryptone (10 g/L), yeast extract (5 g/L), peptone (5 g/L), meat extract (3 g/L), L-cysteine HCl (0.1 g/L), dextrose (2.5 g/L), NaCl (5 g/L), $MgSO_4$ (0.1 g/L), $FeSO_4$ (0.02 g/L), K_2HPO_4 (0.83 g/L), Tris(hydroxymethyl)aminomethane (3.69 g/L) and 10% sheep's blood, pH 5. Strain Marseille P4006 was first isolated after preincubation for 72 hours. A sample of the medium with vaginal swab was inoculated on 5% blood-enriched Columbia agar (bioMérieux, Marcy l'Etoile, France), and the agar plate was incubated for 2 days at 37°C under anaerobic conditions. Colonies were an opaque white with a diameter of approximately 0.5 mm. Bacterial cells were rod shaped and Gram positive, and were facultative anaerobic with a mean breadth of 0.5 µm and a mean length of 1.5 µm. Catalase and oxidase reactions were negative. Matrix-assisted desorption ionization–time of flight mass spectrometry (MALDI-TOF MS) performed on a Microflex spectrometer (Bruker Daltonics, Bremen, Germany) failed to identify the bacterium [2]. Thus, 16S rRNA sequencing was performed with fD1–rP2 primers as previously described (Eurogentec, Seraing, Belgium) using a 3130-XL sequencer (Applied Biosciences, Saint Aubin, France) [3].

16S rRNA gene sequence–based identification of strain Marseille P4006 exhibited a 98.1% sequence similarity with *Lactobacillus farraginis* strain JCM8627 (GenBank accession no. AB690214.1), the phylogenetically closest species with standing in nomenclature (Fig. 1).

Created in 1901 by Beijerinck, the genus *Lactobacillus* contains 222 other species isolated in plants and human that are facultative anaerobic or anaerobic bacteria. *Lactobacillus farraginis,* the phylogenetically closest validated species of strain P4006, was first isolated in 2007 from distilled *shochu* residue (*shochu* is a Japanese spirit made from rice, sweet potato, barley or other starchy materials) but was never isolated from humans [4].

Because the 16S rRNA gene sequence was in the range of 98.7% to define a new species [5], strain Marseille P4006 was

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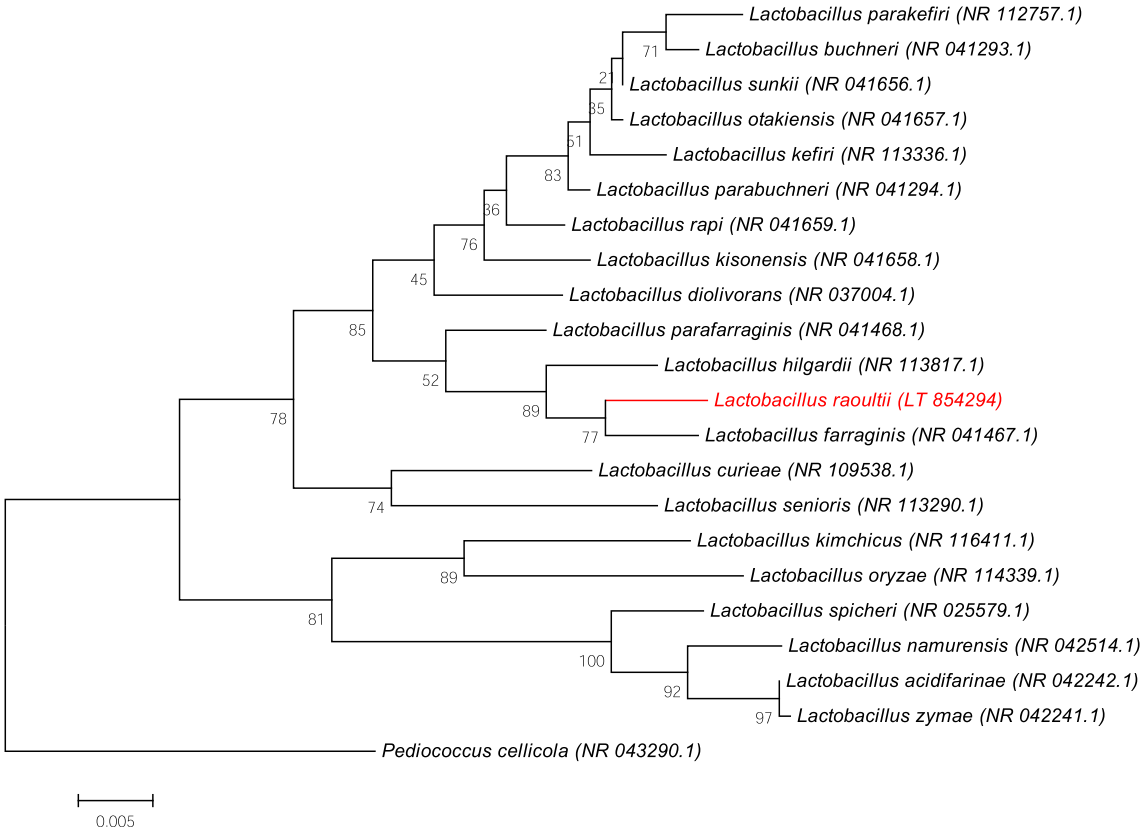

FIG. 1. Phylogenetic tree showing positioning of *'Lactobacillus raoultii'* strain Marseille P4006 relative to its phylogenetically closest bacterial species with standing in nomenclature. 16S rRNA sequences were aligned by MUSCLE 3.8.31 with default parameters, and phylogenetic interferences were obtained using neighbour-joining method with 500 bootstrap replicates by MEGA6 software. Scale bar indicates 0.5% nucleotide sequence divergence.

considered as a new species within the *Lactobacillus* genus in the *Lactobacillaceae* family. Thus, we propose that strain Marseille P4006 may be the putative representative of a novel species named *'Lactobacillus raoultii'* (ra.oul'ti.i, N.L. masc. gen. n., *raoultii,* 'of Raoult,' named after French scientist Didier Raoult in honor of his outstanding contributions to the field of medical microbiology). Strain Marseille P4006^T is the type strain of the new species *'Lactobacillus raoultii'* sp. nov.

MALDI-TOF MS spectrum

The MALDI-TOF MS spectrum of *'Lactobacillus raoultii'* 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 LT854294.

Deposit in a culture collection

Strain Marseille P4006 was deposited in the 'Collection de Souches de l'Unité des Rickettsies' (CSUR, WDCM 875) under number P4006.

Conflict of Interest

None declared.

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