'Brevibacterium ihuae' sp. nov., isolated from a stool sample of a healthy 25-year-old woman

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Abstract

We report the main characteristics of 'Brevibacterium ihuae' strain cv3^T isolated from a stool sample of a healthy 25-year-old woman. © 2017 The Authors. Published by Elsevier Ltd.

Keywords: 'Brevibacterium ihuae', culturomics, emerging bacteria, gut microbiota, human microbiota

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In 2016, as part of the study of the human microbiome by culturomics [1], 'Brevibacterium ihuae' strain cv3 was isolated. We failed to identify this bacterium using matrix-assisted desorption ionization—time of flight mass spectrometry (MALDI-TOF MS) screening on a Microflex spectrometer (Bruker Daltonics, Bremen, Germany) [2]. The stool samples were isolated in Marseille, France, after obtaining the oral approval of the patient. The clinical stool sample was then stored at +4°C before carrying out any experimentation. The ethics committee of the Institut Federatif de Recherche IFR48 validated this study under the number 09-022.

Strain cv3 was first isolated in January 2015 after lyophilization of the stool, which was resuspended and incubated for 24 hours in aerobic atmosphere at 37°C on 5% blood-enriched Columbia agar (bioMérieux, Marcy l'Etoile, France). Cells were Gram-positive rods, and colonies were translucent and 3 mm in diameter on blood-enriched Columbia agar. Strain cv3

exhibited catalase activity but was negative for oxidase. Growth occurred between 25 and 45°C on blood-enriched Columbia agar, with optimal growth being obtained at 28°C after 48 hours of incubation. Growth of the strain was tested under anaerobic and microaerophilic conditions, and under aerobic conditions with or without 5% CO2. Optimal growth was achieved aerobically. Weak cell growth was observed under microaerophilic and anaerobic conditions. The motility test was positive, and the cells were nonsporulating. 16S rRNA gene sequencing was performed using fDI-rP2 primers as previously described, using a 3130-XL sequencer [3]. Strain cv3 showed a 97.25% nucleotide sequence similarity with Brevibacterium senegalense strain JC43 (accession no. JF824806), the phylogenetically closest species with a validly published name [4] (Fig. 1). This similarity value is below the 16S rRNA gene sequence threshold of 98.65% set by Stackebrandt and Ebers [5] to delineate a new species without carrying out DNA-DNA hybridization. Thus, it can be classified as a member of the genus Brevibacterium within the family Brevibacteriaceae and under the Actinobacteriae phylum.

We suggest the discovery of the new species 'Brevibacterium ihuae' (i.hu'ae, N.L. neut. adj. ihuae, from the IHU (Institut Hospitalo-Universitaire), where strain cv3 was isolated). Strain $cv3^{T}$ is the type strain of the new species 'Brevibacterium ihuae.'

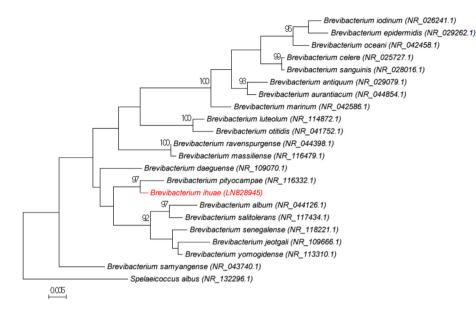


FIG. 1. Phylogenetic tree representing position of 'Brevibacterium ihuae' strain cv3 among other phylogenetically closely related neighbours. Using CLUSTALW tool, sequences of strains involved in this tree were aligned. MEGA software and maximumlikelihood method were used to obtain phylogenetic inferences. Bootstrap values obtained after 500 repeats are shown on nodes. Note that only bootstrap score of at least 90% were retained and that scale bar indicates 0.5% nucleotide sequence divergence.

MALDI-TOF MS spectrum

The MALDI-TOF MS spectrum of strain cv3 is available online (http://www.mediterranee-infection.com/article.php?laref=256 &titre=urms-database).

Nucleotide sequence accession number

The I6S rRNA gene sequence was deposited in GenBank under accession number LN828945.

Deposit in a culture collection

Strain cv3 was deposited in the Collection de Souches de l'Unité des Rickettsies (CSUR, WDCM 875) under number P1577.

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Conflict of interest

None declared.

References

- 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] Huson DH, Auch AF, Qi J, Schuster SC. MEGAN analysis of metagenomic data. Genome Res 2007;17:377–86.
- [5] Stackebrandt E, Ebers J. Taxonomic parameters revisited: tarnished gold standards. Microbiol Today 2006;33:152–5.