

'*Citricoccus massiliensis*' sp. nov., a new bacterial species isolated from human skin by culturomics

C. Ndiaye¹, H. Bassène¹, F. Cadoret², D. Raoult², J. C. Lagier² and C. Sokhna¹

1) Aix-Marseille Université, IRD (Dakar, Marseille, Papeete), AP-HM, IHU-Méditerranée Infection, UMR Vecteurs—Infections Tropicales et Méditerranéennes (VITROME) and 2) Aix-Marseille Université, IRD, MEPHI, IHU-Méditerranée Infection, Marseille, France

Abstract

We report the main characteristics of '*Citricoccus massiliensis*' strain Marseille-P4330, a new species within the genus *Citricoccus*. This strain was isolated from the skin of a healthy human man living in Dielmo, Senegal, Western Africa.

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Corresponding author. C. Sokhna, Aix-Marseille Université, IRD (Dakar, Marseille, Papeete), AP-HM, IHU-Méditerranée Infection, UMR Vecteurs—Infections Tropicales et Méditerranéennes (VITROME), 19-21 Boulevard Jean Moulin, CP 13005, Marseille, France

E-mail: cheikh.sokhna@ird.fr

Skin was thought to harbour a limited number of microbial communities, primarily composed of aerobic cocci. Genomic studies have offered additional insight into the diversity of skin microbiota and have also allowed for quantitative analyses [1]. The earliest descriptions of skin microbiota were based on culture-dependent studies. In our laboratory, a culturomics approach, consisting of rapid identification of grown colonies using matrix-assisted desorption ionization–time of flight mass spectrometry (MALDI-TOF MS), has been developed to isolate previously uncultured human bacteria [2,3]. We used this technique to explore the human skin bacterial repertoire from skin swabs collected from healthy patients living in rural Senegal. The project was approved by the National Ethics Committee of Senegal (no. 53/MSAS/DPRS/CNERS, 31 March 2015).

In 2017, we isolated from skin swabs a bacterial strain that could not be identified by our MALDI-TOF MS screening on a Microflex spectrometer (Bruker Daltonics, Bremen, Germany) [4]. The 16S rRNA gene was sequenced using fD1-rP2 primers as previously

described using a 3130-XL sequencer (Applied Biosciences, Saint-Aubin, France) [5]. Strain Marseille-P4330 exhibited a 98.61% sequence identity with *Citricoccus nitrophenolicus* strain NPPI (GenBank accession no. NR_112632) (Fig. 1). Because the strain showed a similarity of 16S rRNA gene sequence <98.65% with its phylogenetically closest species with standing in nomenclature, we here propose the creation of a new species [6].

Strain Marseille-P4330 was isolated in a skin swab from a 19-year-old man living in Dielmo, Senegal, Western Africa. The growth occurred in Columbia agar culture media with colistin and nalidixic acid (bioMérieux, Marcy l'Etoile, France) and 5% sheep's blood (bioMérieux). The initial agar-grown colonies were obtained after 24 to 48 hours' incubation at 37°C. Colonies were yellowish, circular, shiny, smooth and convex with regular edges and presented a mean diameter of 1.2 mm. The strain was an aerobic, Gram-positive, spore-forming and motile coccus-shaped bacterium, with positive catalase reaction and negative oxidase reaction.

Thus, we propose to classify strain Marseille-P4330 as a new species of the genus *Citricoccus* within the family *Micrococcaceae*, phylum *Actinobacteria* (GenBank accession no. LT960590).

Description of new species '*Citricoccus massiliensis*'

'*Citricoccus massiliensis*' had yellowish, circular, shiny, smooth and convex colonies with a mean diameter of 1.2 mm. The

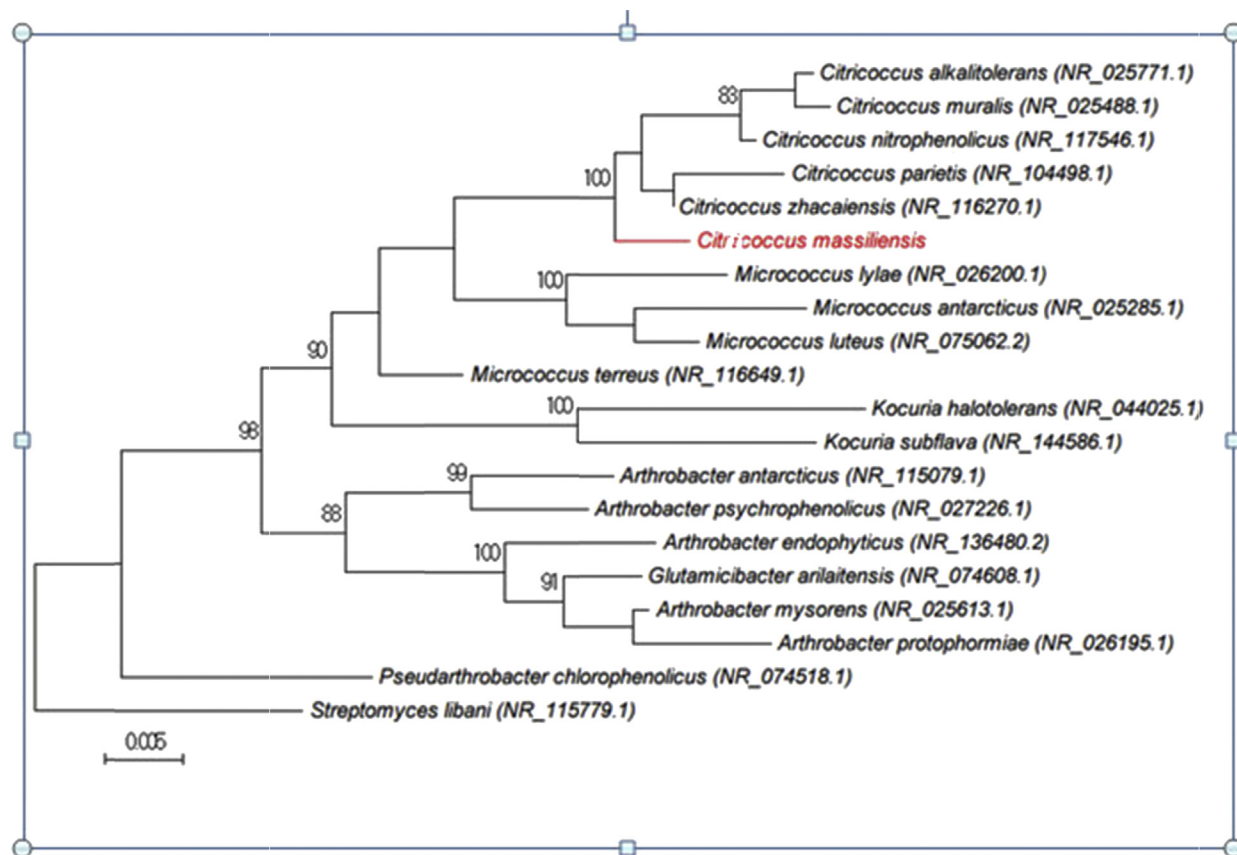


FIG. 1. Phylogenetic tree showing position of '*Citricoccus massiliensis*' Marseille-P4330 relative to other phylogenetically close neighbours. 16S rRNA gene sequences were aligned using CLUSTAL W, and phylogenetic inferences were obtained using maximum-likelihood method within MEGA software. Numbers at nodes are percentages of bootstrap values obtained by repeating analysis 500 times to generate majority consensus tree. Scale bar indicates 0.5% of nucleotide sequence divergence.

strain was an aerobic, Gram-positive, spore-forming and motile coccus-shaped bacterium, with positive catalase reaction and negative oxidase reaction. Strain Marseille-P4330^T (= CSUR P4330) is the type strain of the new species '*Citricoccus massiliensis*' (ma.ssi.li.en'sis, L. masc. adj. *massiliensis*, pertaining to Massilia, the Roman name of Marseille, where the strain was isolated).

Nucleotide sequence accession number

The 16S r-RNA gene sequence was deposited in GenBank under accession number LT960590.

Deposit in a culture collection

Strain Marseille-P4330 was deposited in the Collection de souches de l'Unité des Rickettsies (CSUR, WDCM 875) under number P4330.

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Conflict of interest

None declared.

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