‘Urmitella timonensis’ gen. nov., sp. nov., *‘Blautia marasmi’* sp. nov., *‘Lachnoclostridium pacaense’* sp. nov., *‘Bacillus marasmi’* sp. nov. and *‘Anaerotruncus rubiinfantis’* sp. nov., isolated from stool samples of undernourished African children

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Abstract

We report here the main characteristics of five new species *‘Urmitella timonensis’* strain Marseille-P2918^T (CSUR P2918), *‘Blautia marasmi’* strain Marseille-P2377^T (CSUR P2377), *‘Lachnoclostridium pacaense’* strain Marseille-P3100^T (CSUR P3100), *‘Bacillus marasmi’* strain Marseille-P3556^T (CSUR P3556) and *‘Anaerotruncus rubiinfantis’* strain MT15^T (CSUR P2276), which were isolated recently from stool samples taken from undernourished children in Niger and Senegal using microbial culturomics.

Keywords: Culturomics, human gut microbiota, malnutrition, new species, taxonogenomics
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In 2016, as a part of culturomics study of the human microbiome, we isolated five bacterial strains from stool samples of patients in Senegal and Niger with malnutrition (marasmus and kwashiorkor) [6,7] that could not be 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) [1,2,4]. The patients signed informed consent; the study was validated by the ethics committee of the Institut Federatif de Recherche IFR48 under number 09-022.

In this study, we will describe these five bacterial strains, which were deposited in the open Collection de Souches de l’Unité des Rickettsies (CSUR, WDCM 875) under reference numbers P2918 (*‘Urmitella timonensis’* strain Marseille-P2918^T) [8], P2377 (*‘Blautia marasmi’* strain Marseille-P2377^T), P3100 (*‘Lachnoclostridium pacaense’* strain Marseille-P3100^T), P3556 (*‘Bacillus marasmi’* strain Marseille-P3556^T) and P2276 (*‘Anaerotruncus rubiinfantis’* strain MT15^T), respectively.

Two strains, Marseille-P2918^T and Marseille-P3100^T, were isolated from a 3.3-month-old Senegalese girl with clinical aspects of marasmus (70 cm, 7 kg, weight-for-height Z score −1.75). Strain Marseille-P2377^T was isolated from a 2.5-month-old Senegalese boy with severe acute malnutrition (64 cm, 3 kg, weight-for-height Z score −8.9). Meanwhile, strain Marseille-P3556^T was isolated from a Nigerian child with clinical aspects of marasmus, but anthropometric criteria were not available for this child. Strain MT15^T was isolated from a 13-month-old Nigerian girl with oedematous severe acute malnutrition, also known as kwashiorkor (72 cm, 8 kg, presence of oedema).

The following conditions for initial growth existed for each strain: Strain Marseille-P2918^T was isolated after 7 days in blood culture bottle + sheep’s blood, in anaerobe condition, 37°C; strain Marseille-P2377^T was isolated after 3 days in blood

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culture bottle, in anaerobe condition, 37°C; strain Marseille-P3100^T was isolated after 3 days of culture in COS medium (Columbia blood agar with sheep's blood medium), anaerobe condition, 37°C; strain Marseille-P3556^T was isolated after 1 day in blood culture bottle + sheep's blood + rumen, aerobic condition, 37°Cand strain MT15^T was isolated after 10 days in blood culture + sheep's blood, anaerobe condition, 37°C.

In this study, we described the colony morphologies of these five bacterial species. Colonies of strain Marseille-P2918^T were circular, smooth, very small and pale grey with a mean diameter of 0.2 to 0.5 mm. Colonies of the strain Marseille-P2377^T were white, circular, smooth, convex with intact edges with a larger diameter of 0.5 to 3 mm. Colonies of the strain Marseille-P3100^T were white, circular, smooth with intact edges and a mean diameter of 1 to 3 mm. Colonies of the strain Marseille-P3556^T were flat, smooth, small, circular and pale grey with a mean diameter of 0.5 to 2 mm. Colonies of the strain MT15^T were small, smooth with intact edges and a mean diameter of 1 to 3 mm.

All five bacterial strains were Gram-positive, rod-shaped for strain Marseille-P2918^T, Marseille-P3100^T, Marseille-P3556^T, MT15^T; and coccus-shaped for strain Marseille-P2377^T; polymorphic.

The 16S rRNA gene of these five strains were sequenced using fD1-rP2 primers as previously described using a 3130-XL sequencer (Applied Biosciences, Saint Aubin, France).

Strain Marseille-P2918^T exhibited a 93.25% 16S rRNA gene sequence identity with *Tissierella creatinini* strain BN11 (Gen-Bank accession no. NR_104805), the phylogenetically closest species with standing in nomenclature (Fig. 1), which putatively

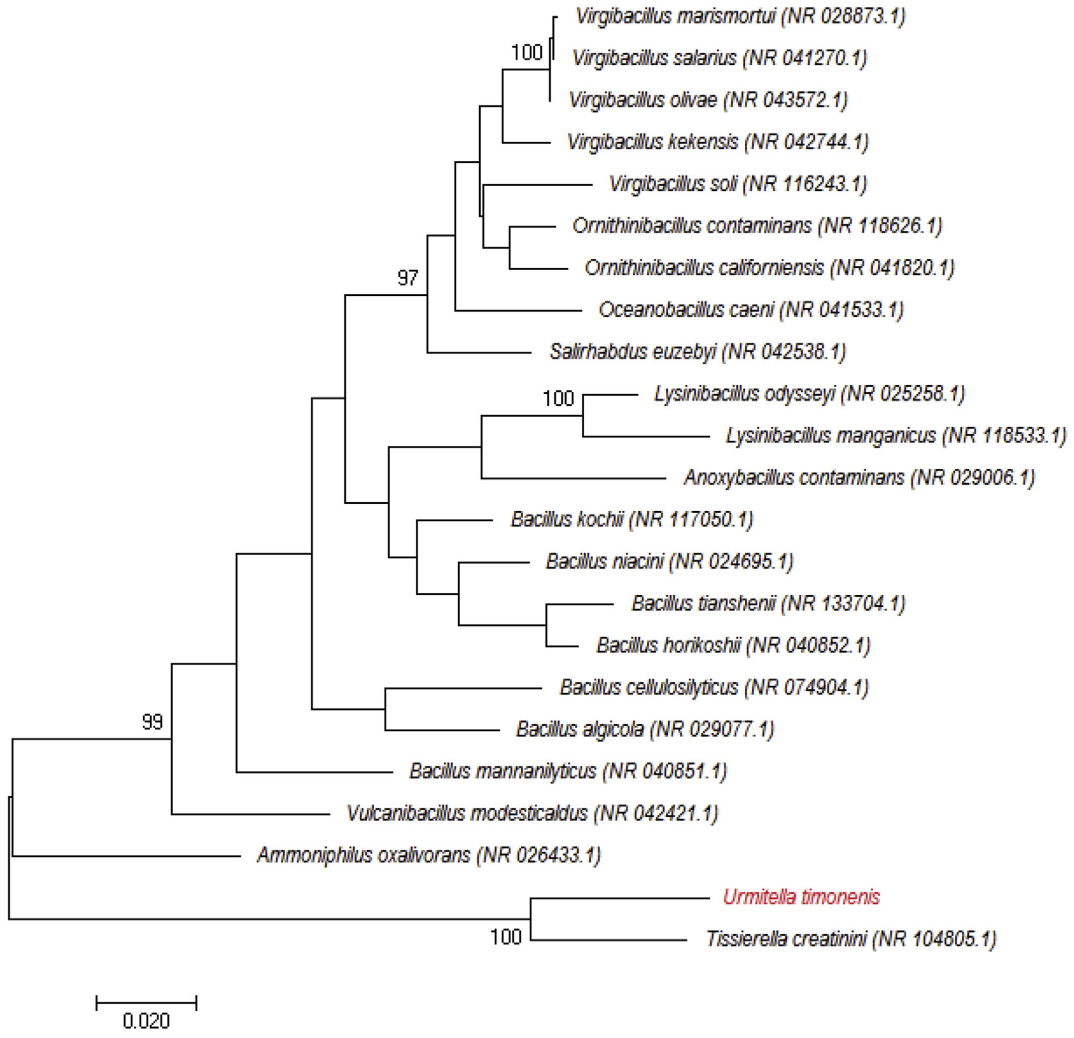

FIG. 1. Phylogenetic tree showing position of *'Urmitella timonensis'* strain Marseille-P2918^T relative to other phylogenetically close neighbours. Sequences were aligned using CLUSTALW, and phylogenetic inferences were obtained using maximum-likelihood method within MEGA software. Numbers at nodes are percentages of bootstrap values obtained by repeating analysis 500 times to generate majority consensus tree. Only bootstraps scores of at least 90% were retained. Scale bar indicates 2% nucleotide sequence divergence.