Non-contiguous finished genome sequence and description of *Nosocomiicoccus massiliensis* sp. nov.

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*Nosocomiicoccus massiliensis* strain NP2 sp. nov. is the type strain of a new species within the genus *Nosocomiicoccus*. This strain, whose genome is described here, was isolated from the fecal flora of an AIDS-infected patient living in Marseille, France. *N. massiliensis* is a Gram-positive aerobic coccus. Here we describe the features of this organism, together with the complete genome sequence and annotation. The 1,645,244 bp long genome (one chromosome but no plasmid) contains 1,738 protein-coding and 45 RNA genes, including 3 rRNA genes.

**Introduction**

*Nosocomiicoccus massiliensis* strain NP2 (CSUR P246 = DSM 26222) is the type strain of *N. massiliensis* sp. nov. This bacterium is a Gram-positive, non-spore-forming, indole negative, aerobic and motile coccus that was isolated from the stool of an AIDS-infected patient living in Marseille (France) and is part of a "culturomics" study aiming at cultivating all species within human feces [1,2].

The current prokaryote species classification, known as polyphasic taxonomy, is based on a combination of genomic and phenotypic properties [3]. With each passing year, the number of completely sequenced genomes increases geometrically while the cost of such techniques decreases. More than 4,000 bacterial genomes have been published and approximately 15,000 genome projects are anticipated to be completed in the near future [4]. We recently proposed to integrate genomic information in the taxonomic framework and description of new bacterial species [5-22].

Here we present a summary classification and a set of features for *N. massiliensis* sp. nov. strain NP2 (CSUR P246 = DSM 26222), together with the description of the complete genomic sequence and its annotation. These characteristics support the circumscription of the species *N. massiliensis*. The genus *Nosocomiicoccus* Alves *et al.* 2008 was created on the basis of 16S rRNA gene sequence and phenotypic analyses within the family *Staphylococcaceae* [23]. To date, this genus is comprised of a single species, *N. ampullae*, which was isolated from the surface of saline bottles used for washing wounds in hospital wards [23].

**Classification and features**

A stool sample was collected from an HIV-infected patient living in Marseille (France). The patient gave an informed and signed consent. This study and the assent procedure were approved by the ethics committee of the IFR48 (Marseille, France) under reference 09-022. The fecal specimen was preserved at -80°C after collection. Strain NP2 (Table 1) was isolated in January 2012 by aerobic cultivation on 5% sheep blood agar (BioMerieux, Marcy l’Étoile, France) at 37°C, after 14-days of preincubation of the stool sample in a blood culture bottle supplemented with 5 ml of sterile ovine rumen fluid. This strain exhibited a 97% nucleotide sequence similarity with *N. ampullae* [23] and a range of 92-94% nucleotide sequence similarity to the most closely related members of the genus *Jeotgalicoccus* [34] (Figure 1). These values were lower than the 98.7% 16S rRNA gene sequence threshold recommended by Stackebrandt and Ebers to delineate a new species without carrying out DNA-DNA hybridization [35].
**Table 1. Classification and general features of *Nosocomiicoccus massiliensis* strain NP2<sup>T</sup>**

| MIGS ID | Property              | Term                  | Evidence code<sup>a</sup> |
|---------|-----------------------|-----------------------|---------------------------|
|         | Domain                | *Bacteria*            | TAS [24]                  |
|         | Phylum                | *Firmicutes*          | TAS [25-27]               |
|         | Class                 | *Bacilli*             | TAS [28,29]               |
| Current classification | Order          | *Bacillales*          | TAS [30,31]               |
|         | Family                | *Staphylococcaceae*   | TAS [28,32]               |
|         | Genus                 | *Nosocomiicoccus*     | TAS [23]                  |
|         | Species               | *Nosocomiicoccus massiliensis* | IDA             |
|         | Type strain           | NP2<sup>T</sup>       |                           |
|         | Gram stain            | Positive              | IDA                       |
|         | Cell shape            | Cocci                 | IDA                       |
|         | Motility              | Motile                | IDA                       |
|         | Sporulation           | Nonsporulating        | IDA                       |
|         | Temperature range     | Mesophile             | IDA                       |
|         | Optimum temperature   | 37°C                  | IDA                       |
| MIGS-6.3| Salinity              | Unknown               | IDA                       |
| MIGS-22 | Oxygen requirement    | Aerobic               | IDA                       |
|         | Carbon source         | Unknown               | NAS                      |
|         | Energy source         | Unknown               | NAS                      |
| MIGS-6  | Habitat               | Human gut             | IDA                       |
| MIGS-15 | Biotic relationship   | Free living           | IDA                       |
| MIGS-14 | Pathogenicity         | Unknown               |                           |
|         | Biosafety level       | 2                     |                           |
|         | Isolation             | Human feces           |                           |
| MIGS-4  | Geographic location   | France                | IDA                       |
| MIGS-5  | Sample collection time| January 2012          | IDA                       |
| MIGS-4.1| Latitude              | 43.296482             | IDA                       |
| MIGS-4.2| Longitude             | 5.36978               | IDA                       |
| MIGS-4.3| Depth                 | Surface               | IDA                       |
| MIGS-4.4| Altitude              | 0 m above sea level   | IDA                       |

Evidence codes - IDA: Inferred from Direct Assay; TAS: Traceable Author Statement (i.e., a direct report exists in the literature); NAS: Non-traceable Author Statement (i.e., not directly observed for the living, isolated sample, but based on a generally accepted property for the species, or anecdotal evidence). These evidence codes are from the Gene Ontology project [33]. If the evidence is IDA, then the property was directly observed for a live isolate by one of the authors or an expert mentioned in the acknowledgements.
Figure 1. Phylogenetic tree highlighting the position of *Nosocomiicoccus massiliensis* strain NP2\(^T\) relative to a selection of type strains of validly published type strains within the *Staphylococcaceae* family. GenBank accession numbers are indicated in parentheses. Sequences were aligned using CLUSTALW, and phylogenetic inferences obtained using the maximum-likelihood method within MEGA program. Numbers at the nodes are percentages of bootstrap values obtained by repeating the analysis 500 times to generate a majority consensus tree. *Marinococcus halophilus* was used as the outgroup. The scale bar represents a 2\% nucleotide sequence divergence.
**Figure 2.** Gram staining of *N. massiliensis* strain NP2<sup>T</sup>

**Figure 3.** Transmission electron microscopy of *N. massiliensis* strain NP2<sup>T</sup>, using a Morgani 268D (Philips) at an operating voltage of 60kV. The scale bar represents 900 nm.
Different growth temperatures (25, 30, 37, 45°C) were tested. Growth was observed between 25 and 45°C, with optimal growth at 37°C after 24 hours of incubation. Colonies were 1 mm in diameter on blood-enriched Columbia agar. Growth of the strain was tested on 5% sheep blood agar, under anaerobic and microaerophilic conditions using GENbag anaer and GENbag microaer systems, respectively (BioMerieux), and under aerobic conditions, with or without 5% CO₂. The strain optimal growth was obtained aerobically, weak growth was observed in microaerophilic but no growth was observed under anaerobic atmospheres. Gram staining showed Gram-positive coccus. The motility test was positive. Cells grown on agar are Gram-positive cocci (Figure 2) and have a mean diameter of 0.72 µm as determined by electron microscopy (Figure 3).

Strain NP2ᵀ exhibited catalase but no oxidase activities. Using an API 20NE strip (BioMerieux, Marcy l’Etoile), negative reactions were obtained for nitrate reduction, urease, indole production, glucose fermentation, arginine dihydrolase, β-galactosidase, glucose, arabinose, mannose, mannitol, N-acetylglucosamine, maltose, gluconate, caprate, adipate, malate, citrate, phenyl-acetate and cytochrome oxidase. Substrate oxidation and assimilation was examined with an API 50CH strip (BioMerieux) at the optimal growth temperature but sugar fermentation reactions and assimilation were not observed. N. massiliensis strain NP2ᵀ was susceptible to amoxicillin, imipenem, rifampicin, vancomycin doxycycline and gentamicin but resistant to trimethoprim/sulfamethoxazole, metronidazole and ciprofloxacin. When compared with representative species from the family Staphylococcaceae, N. massiliensis strain NP2ᵀ exhibited the phenotypic differences detailed in Table 2.

Matrix-assisted laser-desorption/ionization time-of-flight (MALDI-TOF) MS protein analysis was carried out as previously described [36] using a Microflex spectrometer (Bruker Daltonics, Leipzig, Germany). Twelve individual colonies were deposited on a MTP 384 MALDI-TOF target plate (Bruker). The twelve NP2ᵀ spectra were imported into the MALDI BioTyper software (version 2.0, Bruker) and analyzed by standard pattern matching (with default parameter settings) against the main spectra of 4, 706 bacteria, including spectra from one validly published species of Nosocomiicoccus, used as reference data in the BioTyper database. A score enabled the presumptive identification and discrimination of the tested species from those in a database: a score ≥ 2 with a validly published species enabled the identification at the species level; and a score < 1.7 did not enable any identification. For strain NP2ᵀ, no significant score was obtained, suggesting that our isolate was not a member of any known species (Figures 4 and 5).

**Genome sequencing information**

**Genome project history**

The organism was selected for sequencing on the basis of its phylogenetic position and 16S rRNA similarity to other members of the genus Nosocomiicoccus, and is part of a “culturomics” study of the human digestive flora aiming at isolating all bacterial species within human feces. It was the first genome of a Nosocomiicoccus species and the first genome of Nosocomiicoccus massiliensis sp. nov. A summary of the project information is shown in Table 3. The Genbank accession number is CAVG00000000 and consists of 154 contigs. Table 3 shows the project information and its association with MIGS version 2.0 compliance [37].

**Growth conditions and DNA isolation**

N. massiliensis sp. nov. strain NP2ᵀ (≈ CSURP246 = DSM 26222), was grown aerobically on M17 agar medium at 37°C. Five Petri dishes were spread and resuspended in 3x100µl of G2 buffer (EZ1 DNA Tissue kit, Qiagen). A first mechanical lysis was performed by glass powder on the Fastprep-24 device (Sample Preparation system, MP Biomedicals, USA) for 2×20 seconds. DNA was treated with 2.5 µg/µL of lysozyme (30 minutes at 37°C) and extracted using the BioRobot EZ 1 Advanced XL (Qiagen). The DNA was concentrated and purified on a Qiamp kit (Qiagen). The yield and the concentration of DNA was 69.3 ng/µl as measured by using Quant-it Picogreen kit (Invitrogen) on the Genios Tecan fluorometer.

**Genome sequencing and assembly**

DNA (5 µg) was mechanically fragmented for the paired-end sequencing, using a Covaris device (Covaris Inc., Woburn, MA,USA) with an enrichment size of 3-4 kb. The DNA fragmentation was visualized through an Agilent 2100 BioAnalyzer on a DNA Labchip 7500 which yielded an optimal size of 3.4 kb. The library was constructed using a 454 GS FLX Titanium paired-end rapid library protocol. Circularization and nebulization were performed and a pattern of optimal size of 589 bp was generated. PCR amplification was performed for 17 cycles followed by double size selection. The single-

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stranded paired-end library was quantified using a Quant-it Ribogreen Kit (Invitrogen) using a Genios Tecan fluorometer. The library concentration equivalence was calculated as $1.42 \times 10^{10}$ molecules/µL. The library was stored at -20°C until further use.

Table 2. Differential characteristics of Nosocomiicoccus species*

| Properties                  | N. massiliensis | N. ampullae | J. psychrophilus | M. caseolyticus | S. pseudointermedius | S. albus |
|-----------------------------|-----------------|-------------|-------------------|-----------------|-----------------------|---------|
| Cell diameter (µm)          | 0.72            | na          | 0.6-1.1           | 1.1-2           | 1.0-1.5               | 1.0-2.0 |
| Oxygen requirement          | aerobic         | aerobic     | anaerobic         | Facultative anaerobic | aerobic               | aerobic |
| Pigment production          | +               | +           | +                 | +               | +                     | +       |
| Gram stain                  | +               | +           | +                 | +               | +                     | +       |
| Salt requirement            | –               | +           | +                 | +               | na                    | +       |
| Motility                    | +               | –           | –                 | –               | –                     | –       |
| Peptidoglycan type          | na              | L-Lys-Gly$_4$-L-Ser(Gly) | L-Lys-Gly$_3$-L-Ala(Gly) | L-Lys-Gly$_4$-L-Ser-teichoic acid | na | L-Lys-Gly$_5$ |
| Endospore formation         | –               | –           | –                 | –               | –                     | –       |

Production of

- Acid phosphatase: –, –, +, +, +, +
- Catalase: +, +, +, +, +, +
- Oxidase: –, +, +, +, +, –
- Nitrate reductase: –, –, –, +, +, +
- Urease: –, –, –, –, +, +
- a-galactosidase: –, –, na, –, +, –
- N-acetyl-glucosamine: –, –, na, na, +, –

Acid from

- L-Arabinose: –, w, –, +, –, +
- Ribose: –, –, –, +, +, +
- Mannose: –, –, –, +, +, +
- Mannitol: –, –, –, +, w, +
- Sucrose: –, –, w, na, +, +
- D-glucose: –, –, –, +, +, +
- D-fructose: –, –, –, +, +, +
- D-maltose: –, –, –, +, +, +
- D-lactose: –, –, –, +, +, –

Hydrolysis of gelatin

G+C content (mol%) 36.4 33.5 42 36.5 37.5 43.88

Habitat

- human gut
- surface of used saline bottles
- fermented seafood
- raw cow milk and dairy products
- skin and mucosal surfaces of most healthy dogs
- subterranean brine sample

na = data not available; w = weak

*Nosocomiicoccus massiliensis strain NP2*, Nosocomiicoccus ampullae strain TRF-1, Jeotgalicoccus psychrophilus strain YKJ-115, Macrococcus caseolyticus strain JCSC5402, Staphylococcus pseudointermedius strain ED 99 and Salinicoccus albus strain DSM 19776
Figure 4. Reference mass spectrum from *N. massiliensis* strain NP2<sup>T</sup>. Spectra from 12 individual colonies were compared and a reference spectrum was generated.

Figure 5. Gel view comparing *N. massiliensis* sp. nov strain NP2 and other *Staphylococcus* species. The gel view displays the raw spectra of loaded spectrum files arranged in a pseudo-gel like look. The x-axis records the m/z value. The left y-axis displays the running spectrum number originating from subsequent spectra loading. The peak intensity is expressed by a Gray scale scheme code. The color bar and the right y-axis indicate the relation between the color a peak is displayed with and the peak intensity in arbitrary units. Displayed species are indicated on the left.
For the shotgun sequencing, DNA (500 ng) was mechanically fragmented using a Covaris device (Covaris Inc.) as described by the manufacturer. The DNA fragmentation was visualized using an Agilent 2100 BioAnalyzer on a DNA LabChip 7500 which yielded an optimal size of 1.7 kb. The library was constructed using the GS Rapid library Prep kit (Roche) and quantified using a TBS 380 mini fluorometer (Turner Biosystems, Sunnyvale, CA, USA). The library concentration equivalence was calculated as $2.8 \times 10^9$ molecules/µL. The library was stored at -20°C until further use.

The shotgun library was clonally amplified with 1 and 2 cpb in two emPCR reactions each, and the paired-end library was amplified with 0.5 cpb in three emPCR reactions using the GS Titanium SV emPCR Kit (Lib-L) v2 (Roche). The yields of the emPCR were 68 and 9.8%, respectively, for the shotgun library, and 11.29% for the paired-end library. These yields fall into the expected 5 to 20% range according to Roche protocol.

For each library, approximately 790,000 beads for a quarter region were loaded on the GS Titanium PicoTiterPlate PTP kit and sequenced with the GS FLX Titanium Sequencing Kit XLR70 (Roche). The run was performed overnight and analyzed on a cluster using the gsRunBrowser and Newbler assembler (Roche). For the shotgun sequencing, 188,659 passed-filter wells were obtained. The sequencing generated 129.3 Mb with a length average of 685 bp. For the paired-end sequencing, 106,675 passed-filter wells were obtained. The sequencing generated 35 Mb with an average length of 262 bp. The passed-filter sequences were assembled using Newbler with 90% identity and 40 bp as overlap. The final assembly identified 12 scaffolds and 154 contigs (> 1,500 bp) and generated a genome size of 1.65 Mb, which corresponds to a coverage of 94.97 genome equivalents.

### Genome annotation

Open Reading Frames (ORFs) were predicted using Prodigal [38] with default parameters but the predicted ORFs were excluded if they were spanning a sequencing gap region. The predicted bacterial protein sequences were searched against the GenBank database [39] and the Clusters of Orthologous Groups (COG) databases using BLASTP. The tRNAscanSE tool [40] was used to find tRNA genes, whereas ribosomal RNAs were found by using RNAmer [41] and BLASTn against the GenBank database. Lipoprotein signal peptides and numbers of transmembrane helices were predicted using SignalP [42] and TMHMM [43] respectively. ORFans were identified if their BLASTP E-value was lower than $1e^{-03}$ for alignment length greater than 80 amino acids. If alignment lengths were smaller than 80 amino acids, we used an E-value of $1e^{-05}$. Such parameter thresholds have already been used in previous works to define ORFans. To estimate the mean level of nucleotide sequence similarity at the genome level between *N. massiliensis* and three other members of the family *Staphylococcaceae* (Table 6), we used the Average Genomic Identity of Orthologous gene Sequences (AGIOS) home-made software. Briefly, this software combines the Proteinortho software (version 1.4) [44] for detecting orthologous proteins between genomes compared two by two, then retrieves the corresponding genes and determines the mean percentage of nucleotide sequence identity among orthologous ORFs using the Needleman-Wunsch global alignment algorithm.

*Nosocomiicoccus massiliensis* strain NP2T was compared to *Macrococcus caseolyticus* strain JCSC5402.
(GenBank accession number NC_011999), *Staphylococcus pseudointermedius* strain ED 99 (NC_017568), and *Salinicoccus albus* strain DSM 19776 (ARQJ00000000). Artemis [45] was used for data management and DNA Plotter [46] was used for visualization of genomic features. The Mauve alignment tool was used for multiple genomic sequence alignment and visualization [47].

**Genome properties**

The genome of *N. massiliensis* strain NP2\(^T\) is 1,645,244 bp long (1 chromosome, but no plasmid) with a 36.40% G + C content (Figure 6 and Table 4). Of the 1,783 predicted genes, 1,738 were protein-coding genes, and 45 were RNAs. Three rRNA genes (one 16S rRNA, one 23S rRNA and one 5S rRNA) and 42 predicted tRNA genes were identified in the genome. A total of 1,350 genes (75.71%) were assigned a putative function. Two hundred forty-six genes were identified as ORFans (13.79%). The remaining genes were annotated as hypothetical proteins. The properties and the statistics of the genome are summarized in Table 4 and Table 5. The distribution of genes into COGs functional categories is presented in Table 5.

![Figure 6](http://standardsingenomics.org)
**Table 4.** Nucleotide content and gene count levels of the genome

| Attribute                              | Value   | % of total |
|----------------------------------------|---------|------------|
| Genome size (bp)                       | 1,645,244 |
| DNA coding region (bp)                 | 1,479,861 | 89.94     |
| DNA G+C content (bp)                   | 5,98,869  | 36.4       |
| Number of replicons                    | 1       |            |
| Extrachromosomal elements              | 0       |            |
| Total genes                            | 1,783   | 100        |
| RNA genes                              | 45      | 2.52       |
| rRNA operons                           | 1       |            |
| Protein-coding genes                   | 1,738   | 97.47      |
| Genes with function prediction         | 1,511   | 84.74      |
| Genes assigned to COGs                 | 1,350   | 75.71      |
| Genes with peptide signals             | 84      | 4.71       |
| Genes with transmembrane helices       | 425     | 23.83      |
| CRISPR repeats                         | 0       |            |

*aThe total is based on either the size of the genome in base pairs or the total number of protein coding genes in the annotated genome.

**Table 5.** Number of genes associated with the 25 general COG functional categories

| Code | Value | % of total | Description                                                                 |
|------|-------|------------|-----------------------------------------------------------------------------|
| J    | 144   | 8.29       | Translation                                                                 |
| A    | 0     | 0          | RNA processing and modification                                             |
| K    | 89    | 5.12       | Transcription                                                              |
| L    | 111   | 6.39       | Replication, recombination and repair                                       |
| B    | 1     | 0.06       | Chromatin structure and dynamics                                            |
| D    | 21    | 1.12       | Cell cycle control, mitosis and meiosis                                     |
| Y    | 0     | 0          | Nuclear structure                                                          |
| V    | 36    | 2.07       | Defense mechanisms                                                         |
| T    | 39    | 2.24       | Signal transduction mechanisms                                              |
| M    | 80    | 4.60       | Cell wall/membrane biogenesis                                               |
| N    | 3     | 0.17       | Cell motility                                                              |
| Z    | 0     | 0          | Cytoskeleton                                                               |
| W    | 0     | 0          | Extracellular structures                                                   |
| U    | 23    | 1.32       | Intracellular trafficking and secretion                                     |
| O    | 59    | 3.39       | Posttranslational modification, protein turnover, chaperones                |
| C    | 94    | 5.41       | Energy production and conversion                                            |
| G    | 65    | 3.74       | Carbohydrate transport and metabolism                                       |
| E    | 114   | 6.56       | Amino acid transport and metabolism                                         |
| F    | 55    | 3.16       | Nucleotide transport and metabolism                                         |
| H    | 73    | 4.20       | Coenzyme transport and metabolism                                           |
| I    | 46    | 2.65       | Lipid transport and metabolism                                              |
| P    | 108   | 6.21       | Inorganic ion transport and metabolism                                      |
| Q    | 28    | 1.61       | Secondary metabolites biosynthesis, transport and catabolism                |
| R    | 185   | 10.64      | General function prediction only                                            |
| S    | 137   | 7.88       | Function unknown                                                           |
| -    | 388   | 22.32      | Not in COGs                                                                |

*aThe total is based on the total number of protein coding genes in the annotated genome.
Genome comparison of *Nosocomiicoccus massiliensis* with *Macrococcus caseolyticus*, *Staphylococcus pseudointermedius* and *Salinicoccus albus*

We compared the genome of *N. massiliensis* strain NP2³, with those of *M. caseolyticus* strain JCSC5402 (GenBank accession number NC_011999) and *S. pseudointermedius* strain ED 99 (NC_017568), and *S. albus* strain DSM 19776 (ARQJ00000000). The draft genome of *N. massiliensis* is smaller in size than those of *M. caseolyticus*, *S. pseudointermedius* and *S. albus* (1.6, 2.2, 2.5 and 2.6 Mb, respectively). The G+C content of *B. massiliensis* is comparable to that of *M. caseolyticus* (36.40 and 36.56%, respectively) and lower than that of *S. pseudointermedius* and *S. albus* (37.56 and 43.88%, respectively). The gene content of *N. massiliensis* is lower than those of *M. caseolyticus* *S. pseudointermedius* and *S. albus* (1,783, 2,113, 2,435 and 2,770, respectively). The ratio of genes per Mb of *N. massiliensis* is larger to those of *M. caseolyticus*, *S. pseudointermedius* and *S. albus* (1,080, 956, 947 and 1,049, respectively). However, the distribution of genes into COG categories was not entirely similar in the four genomes (Figure 7).

The nucleotide sequence identity ranged from 64.75 to 69.80% among the genera. Table 6 summarizes the numbers of orthologous genes and the average percentage of nucleotide sequence identity between the different genomes studied.

![Figure 7. Distribution of functional classes of predicted genes on Nosocomiicoccus massiliensis (colored in blue), Macrococcus caseolyticus (colored in green), Staphylococcus pseudointermedius (colored in red) and Salinicoccus albus (colored in yellow) chromosomes according to the clusters of orthologous groups of proteins.](http://standardsingenomics.org)
**Nosocomiicoccus massiliensis** sp. nov.

### Table 6. The numbers of orthologous protein shared between genomes

|                      | Nosocomiicoccus massiliensis | Macroccocus caseolyticus | Salinicoccus albus | Staphylococcus pseudointermedius |
|----------------------|------------------------------|--------------------------|--------------------|----------------------------------|
| **Nosocomiicoccus massiliensis** | 1,742                        | 995                      | 1,003              | 954                              |
| **Macroccocus caseolyticus**      |                              | 67.50                    | 2,216              | 1,167                            |
| **Salinicoccus albus**            | 66.22                        | 65.46                    | 2,680              | 1,135                            |
| **Staphylococcus pseudointermedius** | 67.48                        | 69.80                    | 64.75              | 2,351                            |

Upper right triangle- average percentage similarity of nucleotides corresponding to orthologous protein shared between genomes

Lower left triangle- orthologous protein shared between genomes

Bold- numbers of proteins per genome

### Conclusion

On the basis of phenotypic, phylogenetic and genomic analyses, we formally propose the creation of **Nosocomiicoccus massiliensis** sp. nov. that contains the strain NP2\(^T\). This bacterium strain has been isolated from the fecal flora of an AIDS-infected patient living in Marseille, France. Several other undescribed bacterial species were also cultivated from different fecal samples through diversification of culture conditions [5-22], thus suggesting that the human fecal flora of humans remains partially unknown.

**Description of Nosocomiicoccus massiliensis** sp. nov. **Nosocomiicoccus massiliensis** (mas.si.li.en’sis. L. masc. adj. massiliensis of Massilia, the Roman name of Marseille, France, where the type strain was isolated).

Colonies are 1 mm in diameter on blood-enriched Columbia agar. Cells are cocci-shaped with a mean diameter of 0.72 µm. Optimal growth is achieved aerobically and weak growth was observed microaerophilic condition. No growth is observed in anaerobic conditions. Growth occurs between 25 and 45°C, with optimal growth observed at 37°C. Cells stain Gram-positive, are non-endospore forming and are motile. Cells are negative for nitrate reduction, urease, indole production, glucose fermentation, arginine dihydrolase, β-galactosidase, glucose, arabinose, mannose, mannitol, N-acetyl-glucosamine, maltose, gluconate, caprate, adipate, malate, citrate, phenyl-acetate and cytochrome oxidase. Cells are susceptible to amoxicillin, imipenem, rifampicin, vancomycin doxycycline and gentamicin but resistant to trimethoprim/sulfamethoxazole, metronidazole and ciprofloxacin. The G+C content of the genome is 36.40%. The 16S rRNA and genome sequences are deposited in Genbank under accession numbers JX424771 and CAVG00000000, respectively.

The type strain NP2\(^T\) (= CSUR P246 = DSM 26222) was isolated from the fecal flora of an AIDS-infected patient living in Marseille, France.

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