



Complete Genome Sequence of *Micrococcus luteus* Strain SGAir0127, Isolated from Indoor Air Samples from Singapore

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ABSTRACT *Micrococcus luteus* strain SGAir0127 was isolated from indoor air samples collected in Singapore. The assembly, based on single-molecule real-time sequencing reads, resulted in two contigs, one chromosomal contig with a length of 2.57 Mbp and one nonchromosomal contig of 8.68 kbp. The genome has a total of 2,564 genes.

*M*icrococcus luteus, previously known as *Micrococcus lysodeikticus*, was first identified by Fleming (1) while discovering lysozyme. It is a Gram-positive, coccusshaped, aerobic bacterium arranged in clusters or tetrads belonging to the actinomycete branch of taxonomy. It survives and grows in a wide variety of environments, including oligotrophic environments (2), owing to its ability to biodegrade atypical substrates like carbofuran, naphthalene, environmental pesticides (3), and itaconate (4), as well as common substrates. *Micrococcus luteus* is also known to be opportunistically pathogenic (5) and putrefactive (6).

Micrococcus luteus strain SGAir0127 was isolated from indoor air samples collected in Singapore (global positioning system [GPS] coordinates 1.3451N, 103.6789E) using a single-stage Andersen impactor (SKC, USA) to impact air onto a Trypticase soy agar (TSA) plate (Becton, Dickinson, USA). After the initial incubation at 30°C, a pure culture of strain SGAir0127 was achieved by repeated restreaking on fresh TSA plates. A colony was then inoculated in lysogeny broth (LB) (Merck, USA) at 30°C overnight and used for DNA extraction. Genomic DNA was extracted using the Wizard Genomic DNA purification kit (Promega, USA), following the manufacturer's protocol. Library preparation was performed using the SMRTbell template prep kit 1.0 (Pacific Biosciences, USA), followed by single-molecule real-time (SMRT) sequencing on the Pacific Biosciences RS II platform.

The Hierarchical Genome Assembly Process (HGAP) version 3 (with default values) (7) from PacBio's SMRT Analysis 2.3.0 package assembled a total of 60,672 reads into two contigs. After Quiver polishing (with default values) (7), a chromosomal contig with a length of 2,570,740 bp and 201-fold coverage and a nonchromosomal contig with a length of 86,829 bp and 207-fold coverage were found. A positive GC skew of 72.9% was observed in the chromosomal contig, which is characteristic of actinomycetes (8).

Phyla-AMPHORA was run using MarkerScanner.pl with added -DNA flag and Marker-AlignTrim.pl and with options -WithReference and -OutputFormat phylip, followed by Phylotyping.pl, with default parameters (9). It was used for taxonomic identification and showed that the assembled genome has 99.5% marker identity with *Micrococcus luteus*. Microbial Species Identifier (MiSI) (10), based on the genome-wide average nucleotide identity (ANI) metric, was run against a database of 6,387 bacterial refseq genomes using ANICalculator with default parameters, a text filter for "type, synonym type, Citation Kutmutia SK, Drautz-Moses DI, Uchida A, Purbojati RW, Wong A, Kushwaha KK, Putra A, Premkrishnan BNV, Heinle CE, Vettath VK, Junqueira ACM, Schuster SC. 2019. Complete genome sequence of *Micrococcus luteus* strain SGAir0127, isolated from indoor air samples from Singapore. Microbiol Resour Announc 8:e00646-19. https://doi.org/10.1128/MRA .00646-19.

Editor Jason E. Stajich, University of California, Riverside

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Received 30 May 2019 Accepted 16 September 2019 Published 10 October 2019 proxytype," and a subsequent getorf -find 3 option. The results showed 97.8% identity to *Micrococcus luteus* NCTC 2665 with an alignment fraction of 0.60.

PROKKA version 1.12 (added parameters –force – compliant –addmrna –rfam) was used for gene annotation and predicted 2,495 protein-coding genes (11). The genome contained 6 rRNAs, 52 tRNAs, 1 transfer-messenger RNA (tmRNA), and 10 noncoding RNAs (ncRNAs), for a total of 69 RNA genes. In addition, a CRISPR element with 31 repeat units in the chromosomal contig and a CRISPR sequence with 6 repeat units in the nonchromosomal contig were also observed.

Rapid Annotations using Subsystems Technology (RAST) v2 (12–14), run with default parameters with an additional frameshift fix and ClassicRAST annotation scheme, showed the potential ability of this bacterium to metabolize a wide range of organic compounds, such as isoprenoids, aromatics, and carbohydrates, as well as inorganic ions/compounds. RAST identified 68 genes for stress response, including carbon starvation and oxidative stress as a survival strategy (15), as these are the obstacles most likely to be faced by bacteria in the air. Dormancy, persistence, and resuscitation genes were also annotated, but a reduced number was observed compared to other actinomycetes (16). Furthermore, the presence of fluoroquinolone resistance genes is an intriguing feature of this strain.

Data availability. The complete genome sequence of *Micrococcus luteus* strain SGAir0127 has been deposited in DDBJ/EMBL/GenBank under accession number CP025616, and the associated nonchromosomal contig was deposited under accession number CP025617. The Sequence Read Archive files are available under accession number SRR8948643.

ACKNOWLEDGMENTS

This work was supported by a Singapore Ministry of Education Academic Research Fund Tier 3 grant (MOE2013-T3-1-013).

We thank Anjali Bansal Gupta for her review of the manuscript.

REFERENCES

- 1. Alexander F, Edward WA. 1922. On a remarkable bacteriolytic element found in tissues and secretions. Proc R Soc Lond B Biol Sci 93:306–317.
- Greenblatt CL, Baum J, Klein BY, Nachshon S, Koltunov V, Cano RJ. 2004. *Micrococcus luteus*—survival in amber. Microb Ecol 48:120–127. https:// doi.org/10.1007/s00248-003-2016-5.
- Doddamani HP, Ninnekar HZ. 2001. Biodegradation of carbaryl by a Micrococcus species. Curr Microbiol 43:69–73. https://doi.org/10.1007/ s002840010262.
- Cooper RA, Itiaba K, Kornberg HL. 1965. The utilization of aconate and itaconate by *Micrococcus* sp. Biochem J 94:25–31. https://doi.org/10 .1042/bj0940025.
- Fosse T, Toga B, Peloux Y, Granthil C, Bertrando J, Sethian M. 1985. Meningitis due to *Micrococcus luteus*. Infection 13:280–281. https://doi .org/10.1007/bf01645439.
- 6. Hobbs G. 1986. Ecology of food microorganisms. Microb Ecol 12:15–30. https://doi.org/10.1007/BF02153219.
- Chin C-S, Alexander DH, Marks P, Klammer AA, Drake J, Heiner C, Clum A, Copeland A, Huddleston J, Eichler EE, Turner SW, Korlach J. 2013. Nonhybrid, finished microbial genome assemblies from long-read SMRT sequencing data. Nat Methods 10:563. https://doi.org/10.1038/nmeth .2474.
- Barka EA, Vatsa P, Sanchez L, Gaveau-Vaillant N, Jacquard C, Meier-Kolthoff JP, Klenk H-P, Clément C, Ouhdouch Y, van Wezel GP. 2016. Taxonomy, physiology, and natural products of actinobacteria. Microbiol Mol Biol Rev 80:1–43. https://doi.org/10.1128/MMBR.00019-15.
- Wang Z, Wu M. 2013. A phylum-level bacterial phylogenetic marker database. Mol Biol Evol 30:1258–1262. https://doi.org/10.1093/molbev/ mst059.
- Varghese NJ, Mukherjee S, Ivanova N, Konstantinidis KT, Mavrommatis K, Kyrpides NC, Pati A. 2015. Microbial species delineation using whole

genome sequences. Nucleic Acids Res 43:6761-6771. https://doi.org/10 .1093/nar/gkv657.

- Seemann T. 2014. Prokka: rapid prokaryotic genome annotation. Bioinformatics 30:2068–2069. https://doi.org/10.1093/bioinformatics/btu153.
- Aziz RK, Bartels D, Best AA, DeJongh M, Disz T, Edwards RA, Formsma K, Gerdes S, Glass EM, Kubal M, Meyer F, Olsen GJ, Olson R, Osterman AL, Overbeek RA, McNeil LK, Paarmann D, Paczian T, Parrello B, Pusch GD, Reich C, Stevens R, Vassieva O, Vonstein V, Wilke A, Zagnitko O. 2008. The RAST server: Rapid Annotations using Subsystems Technology. BMC Genomics 9:75. https://doi.org/10.1186/1471-2164-9-75.
- Overbeek R, Olson R, Pusch GD, Olsen GJ, Davis JJ, Disz T, Edwards RA, Gerdes S, Parrello B, Shukla M, Vonstein V, Wattam AR, Xia F, Stevens R. 2014. The SEED and the Rapid Annotation of microbial genomes using Subsystems Technology (RAST). Nucleic Acids Res 42:D206–D214. https://doi.org/10.1093/nar/gkt1226.
- Brettin T, Davis JJ, Disz T, Edwards RA, Gerdes S, Olsen GJ, Olson R, Overbeek R, Parrello B, Pusch GD, Shukla M, Thomason JA, Stevens R, Vonstein V, Wattam AR, Xia F. 2015. RASTtk: a modular and extensible implementation of the RAST algorithm for building custom annotation pipelines and annotating batches of genomes. Sci Rep 5:8365. https:// doi.org/10.1038/srep08365.
- McDougald D, Gong L, Srinivasan S, Hild E, Thompson L, Takayama K, Rice SA, Kjelleberg S. 2002. Defences against oxidative stress during starvation in bacteria. Antonie Van Leeuwenhoek 81:3–13. https://doi .org/10.1023/a:1020540503200.
- 16. Young M, Artsatbanov V, Beller HR, Chandra G, Chater KF, Dover LG, Goh E-B, Kahan T, Kaprelyants AS, Kyrpides N, Lapidus A, Lowry SR, Lykidis A, Mahillon J, Markowitz V, Mavromatis K, Mukamolova GV, Oren A, Rokem JS, Smith MCM, Young DI, Greenblatt CL. 2010. Genome sequence of the Fleming strain of *Micrococcus luteus*, a simple free-living actinobacterium. J Bacteriol 192:841–860. https://doi.org/10.1128/JB.01254-09.