



Draft Genome Sequence of the Antimycin-Producing Bacterium *Streptomyces* sp. Strain SM8, Isolated from the Marine Sponge *Haliclona simulans*

Eduardo L. Almeida,^a Lekha M. Margassery,^a Jonathan Kennedy,^b Alan D. W. Dobson^{a,b}

^aSchool of Microbiology, University College Cork, Cork, Ireland ^bEnvironmental Research Institute, University College Cork, Cork, Ireland

ABSTRACT *Streptomyces* sp. strain SM8, isolated from *Haliclona simulans*, possesses antifungal and antibacterial activities and inhibits the calcineurin pathway in yeast. The draft genome sequence is 7,145,211 bp, containing 5,929 predicted coding sequences. Several secondary metabolite biosynthetic gene clusters are present, encoding known and novel metabolites, including antimycin.

Marine organisms are a rich source of novel secondary metabolites, with over 1,000 novel marine compounds discovered in 2015 alone (1). Sponges are a significant source of secondary metabolites, and more than 190 different metabolites have been isolated from the genus *Haliclona* alone (2). Many of the metabolites isolated from marine sponges are believed to be of microbial origin, suggesting symbiotic relationships between sponges and associated bioactive-producing microorganisms (3).

In a study of bacteria isolated from the marine sponge *Haliclona simulans* collected from the west coast of Ireland, up to 50% of bacteria were found to produce antibiotic activity against medically important pathogens, such as methicillin-resistant *Staphylococcus aureus* (MRSA) (4). In an initial screen, the *Streptomyces* sp. strain SM8 showed antibacterial and strong antifungal activities (4). Nuclear magnetic resonance (NMR) analysis of the active fractions proved that hydroxylated saturated fatty acids were the major components present in the antibacterial fractions (5). Subsequent screening showed that this strain also produced compounds that inhibited the calcineurin pathway in *Saccharomyces cerevisiae* (6). Metabolic profiling of the compounds produced by the organism identified antimycin as one of the main products with antifungal activity (5). The genome sequence of strain SM8 was determined to facilitate the identification of the range of bioactive compounds produced by the organism and of further heterologous expression of gene clusters encoding products of interest using the transformation-association recombination (TAR) cloning technique (7, 8).

Genomic DNA (gDNA) was obtained as previously described (5). The nucleotide sequence was generated from a fragment library using the GS FLX Titanium system (Roche), resulting in 229,280 reads and 94,668,678 bp. The assembly of the contigs was performed using i) GS De Novo Assembler v2.3 (Roche) software for the *de novo* assembly of reads and then ii) MeDuSa software v1.6 for the reference-based assembly of scaffolds (9), using as the reference the top 3 complete genomes that shared the highest 16S rRNA gene sequence similarities in NCBI's GenBank database, namely, *Streptomyces sampsonii* strain KJ40 (GenBank accession no. NZ_CP016824), *Streptomyces albus* strain SM254 (NZ_CP014485), and *S. albus* strain J1074 (NC_020990) (10–13). The quality of the final assembly was evaluated with QUAST v4.5 and checkM software, resulting in 11 scaffolds of >500 bp, a length of 7,145,211 bp, a G+C content of 73.33%, and an estimated genome completion of 96.11% (14, 15). The draft sequence was

Received 8 December 2017 Accepted 15 December 2017 Published 25 January 2018

Citation Almeida EL, Margassery LM, Kennedy J, Dobson ADW. 2018. Draft genome sequence of the antimycin-producing bacterium *Streptomyces* sp. strain SM8, isolated from the marine sponge *Haliclona simulans*. Genome Announc 6:e01535-17. https://doi.org/10.1128/ genomeA.01535-17.

Copyright © 2018 Almeida et al. This is an open-access article distributed under the terms of the Creative Commons Attribution 4.0 International license.

Address correspondence to Alan D. W. Dobson, a.dobson@ucc.ie.

annotated using the NCBI Prokaryotic Genome Annotation Pipeline (PGAP) v4.3, which predicted 5,929 coding sequences (CDSs), 3 rRNAs, and 61 tRNAs (16).

Analysis for the prediction of secondary metabolite gene clusters using antiSMASH v4.0.2 identified several gene clusters encoding polyketide synthases (PKS), nonribosomal peptide synthetases (NRPS), PKS/NRPS hybrids, terpene biosynthesis, and siderophores (17). The complete gene cluster for the biosynthesis of antimycin—a compound which was previously identified by mass spectrometry analysis (5)—was also identified using antiSMASH and manually curated. Knowledge of the genetic basis of secondary metabolism in *Streptomyces* sp. strain SM8 will lead to further characterization of the compounds responsible for the wide range of biological activities present.

Accession number(s). This whole-genome shotgun project has been deposited in DDBJ/ENA/GenBank under the accession no. AMPN00000000. The version described in this paper is the second version, AMPN02000000.

ACKNOWLEDGMENTS

This work was supported partially by the Brazilian National Council for Scientific and Technological Development (CNPq), the Beaufort Marine Research Award, part of the Sea Change Strategy, and the Strategy for Science Technology and Innovation (2006–2012), with the support of the Marine Institute under the Marine Research Sub-Programme of the National Development Plan 2007–2013.

Sequencing was carried out at the Centre for Genomic Research, University of Liverpool, Liverpool, United Kingdom.

REFERENCES

- Blunt JW, Copp BR, Keyzers RA, Munro MHG, Prinsep MR. 2017. Marine natural products. Nat Prod Rep 34:235–294. https://doi.org/10.1039/ c6np00124f.
- Yu S, Deng Z, Proksch P, Lin W. 2006. Oculatol, oculatolide, and A-nor sterols from the sponge *Haliclona oculata*. J Nat Prod 69:1330–1334. https://doi.org/10.1021/np0600494.
- Fortman JL, Sherman DH. 2005. Utilizing the power of microbial genetics to bridge the gap between the promise and the application of marine natural products. Chembiochem 6:960–978. https://doi.org/10.1002/cbic .200400428.
- Kennedy J, Baker P, Piper C, Cotter PD, Walsh M, Mooij MJ, Bourke MB, Rea MC, O'Connor PM, Ross RP, Hill C, O'Gara F, Marchesi JR, Dobson ADW. 2009. Isolation and analysis of bacteria with antimicrobial activities from the marine sponge *Haliclona simulans* collected from Irish waters. Mar Biotechnol 11:384–396. https://doi.org/10.1007/s10126-008-9154-1.
- Viegelmann C, Margassery LM, Kennedy J, Zhang T, O'Brien C, O'Gara F, Morrissey JP, Dobson AD, Edrada-Ebel R. 2014. Metabolomic profiling and genomic study of a marine sponge-associated *Streptomyces* sp. Mar Drugs 12:3323–3351. https://doi.org/10.3390/md12063323.
- Margassery LM, Kennedy J, O'Gara F, Dobson AD, Morrissey JP. 2012. A high-throughput screen to identify novel calcineurin inhibitors. J Microbiol Methods 88:63–66. https://doi.org/10.1016/j.mimet.2011.10.012.
- Kouprina N, Larionov V. 2006. TAR cloning: insights into gene function, long-range haplotypes and genome structure and evolution. Nat Rev Genet 7:805–812. https://doi.org/10.1038/nrg1943.
- Li Y, Li Z, Yamanaka K, Xu Y, Zhang W, Vlamakis H, Kolter R, Moore BS, Qian PY. 2015. Directed natural product biosynthesis gene cluster capture and expression in the model bacterium *Bacillus subtilis*. Sci Rep 5:9383. https://doi.org/10.1038/srep09383.
- Bosi E, Donati B, Galardini M, Brunetti S, Sagot MF, Lió P, Crescenzi P, Fani R, Fondi M. 2015. MeDuSa: a multi-draft based scaffolder. Bioinformatics 31:2443–2451. https://doi.org/10.1093/bioinformatics/btv171.

- Benson DA, Cavanaugh M, Clark K, Karsch-Mizrachi I, Lipman DJ, Ostell J, Sayers EW. 2017. GenBank. Nucleic Acids Res 45:D37–D42. https://doi .org/10.1093/nar/gkw1070.
- Zhang B, Zhu T. 2016. Data from "Complete genome sequence of Streptomyces sampsonii KJ40." GenBank https://www.ncbi.nlm.nih.gov/ nuccore/NZ_CP016824 (accession no. NZ_CP016824).
- Badalamenti J, Erickson J, Salomon C. 2016. Data from "Complete genome sequence of *Streptomyces albus* SM254, a potent antagonist of bat white-nose syndrome pathogen *Pseudogymnoascus destructans.*" GenBank https://www.ncbi.nlm.nih.gov/nuccore/NZ_CP014485 (accession no. NZ_CP014485).
- Zaburannyi N, Rabyk M, Ostash B, Fedorenko V, Luzhetskyy A. 2014. Data from "Insights into naturally minimised Streptomyces albus J1074 genome." GenBank https://www.ncbi.nlm.nih.gov/nuccore/NC_020990 (accession no. NC_020990).
- Gurevich A, Saveliev V, Vyahhi N, Tesler G. 2013. QUAST: quality assessment tool for genome assemblies. Bioinformatics 29:1072–1075. https://doi.org/10.1093/bioinformatics/btt086.
- Parks DH, Imelfort M, Skennerton CT, Hugenholtz P, Tyson GW. 2015. CheckM: assessing the quality of microbial genomes recovered from isolates, single cells, and metagenomes. Genome Res 25:1043–1055. https://doi.org/10.1101/gr.186072.114.
- Tatusova T, DiCuccio M, Badretdin A, Chetvernin V, Nawrocki EP, Zaslavsky L, Lomsadze A, Pruitt KD, Borodovsky M, Ostell J. 2016. NCBI prokaryotic genome annotation pipeline. Nucleic Acids Res 44:6614–6624. https://doi .org/10.1093/nar/gkw569.
- Blin K, Wolf T, Chevrette MG, Lu X, Schwalen CJ, Kautsar SA, Suarez Duran HG, de los Santos ELC, Kim HU, Nave M, Dickschat JS, Mitchell DA, Shelest E, Breitling R, Takano E, Lee SY, Weber T, Medema MH. 2017. antiSMASH 4.0—improvements in chemistry prediction and gene cluster boundary identification. Nucleic Acids Res 45:W36–W41. https://doi .org/10.1093/nar/gkx319.