

Whole-Genome Sequencing Analysis of Quorum-Sensing *Aeromonas hydrophila* Strain M023 from Freshwater

Wen-Si Tan,^a Wai-Fong Yin,^a Chien-Yi Chang,^{b,c}  Kok-Gan Chan^a

Division of Genetics and Molecular Biology, Institute of Biological Sciences, Faculty of Science, University of Malaya, Kuala Lumpur, Malaysia^a; Interdisciplinary Computing and Complex BioSystems (ICOS) Research Group, School of Computing Science, Newcastle University, Newcastle upon Tyne, United Kingdom^b; Centre for Bacterial Cell Biology, Medical School, Newcastle University, Newcastle upon Tyne, United Kingdom^c

***Aeromonas hydrophila* is a well-known waterborne pathogen that recently was found to infect humans. Here, we report the draft genome of a freshwater isolate from a Malaysian waterfall, *A. hydrophila* strain M023, which portrays *N*-acylhomoserine lactone-dependent quorum sensing.**

Received 20 December 2014 Accepted 7 January 2015 Published 19 February 2015

Citation Tan W-S, Yin W-F, Chang C-Y, Chan K-G. 2015. Whole-genome sequencing analysis of quorum-sensing *Aeromonas hydrophila* strain M023 from freshwater. *Genome Announc* 3(1):e01548-14. doi:10.1128/genomeA.01548-14.

Copyright © 2015 Tan et al. This is an open-access article distributed under the terms of the [Creative Commons Attribution 3.0 Unported license](https://creativecommons.org/licenses/by/3.0/).

Address correspondence to Kok-Gan Chan, kokgan@um.edu.my.

Bacteria communicate to modulate different physiological activities (1). This process is termed quorum sensing (QS), or bacteria cell-cell communication, which relies on the production and response of small diffusible molecules in relation to population density (2). *Aeromonas hydrophila* is ubiquitous in water medium and tends to be pathogenic to humans by causing gastroenteritis (3, 4). *A. hydrophila* can also cause major economic loss in aquaculture (5). In this study, *A. hydrophila* strain M023 was isolated from a waterfall sample. The whole-genome sequencing was conducted for better understanding of the quorum-sensing system of this strain.

Genomic DNA of strain M023 was extracted by using a MasterPure DNA purification kit (Epicentre, Inc., Madison, WI, USA) with its quality checked by NanoDrop spectrophotometer (Thermo Scientific, Waltham, MA, USA) and a Qubit version 2.0 fluorometer (Life Technologies, Carlsbad, CA, USA). Whole-genome shotgun sequencing of the purified DNA was then performed with the Illumina MiSeq platform (Illumina, Inc., CA, USA), which generated 5,031,085 paired-end reads. Next, the sequences were trimmed and *de novo* assembled with CLC Genomic Workbench version 5.1 (CLC Bio, Denmark). The assembly of trimmed reads (817,464 quality reads) generated a total of 183 contigs and an N_{50} of approximately 67,516.

The draft genome of the strain M023 isolate comprises 4,914,534 bases with an average coverage of 30.5-fold with 61% of G+C content. Next, gene prediction was conducted with the prokaryote gene prediction algorithm by using Prodigal version 2.60 (6), while the amount of tRNAs and rRNAs were predicted using tRNAscan SE version 1.21 (7) and RNAmmer (8), respectively. The strain was then annotated using RAST (9). The open reading frames of strain M023 were predicted at a total of 4,338. There are 6 copies of 5S rRNAs and a single copy each of 23S rRNA and 16S rRNA, while a total of 101 tRNAs were found in the genomes of strain M023.

From the annotation results, the *luxI* and *luxR* homologues of strain M023 were predicted to be located at contig 9. The whole-

genome sequence allows better understanding of the genetic makeup of *A. hydrophila* to determine the link between quorum sensing and pathogenicity and production of virulence factors (10, 11).

Nucleotide sequence accession numbers. This whole-genome shotgun project has been deposited at DDBJ/EMBL/GenBank under the accession number [JSWA00000000](https://www.ncbi.nlm.nih.gov/nuclink/JSWA00000000). The version described in this paper is the first version, JSWA01000000.

ACKNOWLEDGMENTS

This work was supported by the University of Malaya via High Impact Research Grants (UM C/625/1/HIR/MOHE/CHAN/01, A-000001-50001, and UM C/625/1/HIR/MOHE/CHAN/14/1, H-50001-A000027) awarded to K.-G.C.

REFERENCES

1. Tan WS, Yunos NYM, Tan PW, Mohamad NI, Adrian TGS, Yin WF, Chan KG. 2014. Freshwater-borne bacteria isolated from a Malaysian rainforest waterfall exhibiting quorum sensing properties. *Sensors* 14:10527–10537. <http://dx.doi.org/10.3390/s140610527>.
2. Fuqua WC, Winans SC, Greenberg EP. 1994. Quorum sensing in bacteria: the LuxR-LuxI family of cell density-responsive transcriptional regulators. *J Bacteriol* 176:269–275.
3. Williams P, Winzer K, Chan WC, Camara M. 2007. Look who's talking: communication and quorum sensing in the bacterial world. *Philos Trans R Soc Lond B Biol Sci* 362:1119–1134. <http://dx.doi.org/10.1098/rstb.2007.2039>.
4. Chong TM, Koh CL, Sam CK, Choo YM, Yin WF, Chan KG. 2012. Characterization of quorum sensing and quorum quenching soil bacteria isolated from Malaysian tropical montane forest. *Sensors* 12:4846–4859. <http://dx.doi.org/10.3390/s120404846>.
5. Yunos NYM, Tan WS, Mohamad NI, Tan PW, Adrian TGS, Yin WF, Chan KG. 2014. Discovery of *Pantoea rodasi* strain ND03 that produces *N*-(3-oxo-hexanoyl)-L-homoserine lactone. *Sensors* 14:9145–9152. <http://dx.doi.org/10.3390/s140509145>.
6. Hyatt D, Chen GL, LoCascio PF, Land ML, Larimer FW, Hauser LJ. 2010. Prodigal: prokaryotic gene recognition and translation initiation site identification. *BMC Bioinformatics* 11: <http://dx.doi.org/10.1186/1471-2105-11-119>.

7. Lowe TM, Eddy SR. 1997. tRNAscan-SE: a program for improved detection of transfer RNA genes in genomic sequence. *Nucleic Acids Res* 25: 955–964. <http://dx.doi.org/10.1093/nar/25.5.0955>.
8. Lagesen K, Hallin P, Rødland EA, Staerfeldt HH, Rognes T, Ussery DW. 2007. RNAmmer: consistent and rapid annotation of ribosomal RNA genes. *Nucleic Acids Res* 35:3100–3108. <http://dx.doi.org/10.1093/nar/gkm160>.
9. 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. <http://dx.doi.org/10.1186/1471-2164-9-75>.
10. Lynch MJ, Swift S, Kirke DF, Keevil CW, Dodd CER, Williams P. 2002. The regulation of biofilm development by quorum sensing in *Aeromonas hydrophila*. *Environ Microbiol* 4:18–28. <http://dx.doi.org/10.1046/j.1462-2920.2002.00264.x>.
11. Chan KG, Puthucheary SD, Chan XY, Yin WF, Wong CS, Too WS, Chua KH. 2011. Quorum sensing in *Aeromonas* species isolated from patients in Malaysia. *Curr Microbiol* 62:167–172. <http://dx.doi.org/10.1007/s00284-010-9689-z>.