

Genome Sequence of *Stenotrophomonas maltophilia* Strain SmAs1, Isolated From the Asian Malaria Mosquito *Anopheles stephensi*

Grant L. Hughes,^a Juan Antonio Raygoza Garay,^{b,c} Vikas Koundal,^{b,c} Jason L. Rasgon,^{c,d} Michael M. Mwangi^{b,c}

Department of Pathology and Institute for Human Infections and Immunity, University of Texas Medical Branch, Galveston, Texas, USA^a; Department of Biochemistry and Molecular Biology, Center for Infectious Disease Dynamics, Pennsylvania State University, University Park, Pennsylvania, USA^b; Huck Institutes of the Life Sciences, Pennsylvania State University, University Park, Pennsylvania, USA^c; Department of Entomology, Center for Infectious Disease Dynamics, Pennsylvania State University, University Park, Pennsylvania, USA^d

G.L.H. and J.A.R.G. contributed equally to this article.

An isolate of *Stenotrophomonas maltophilia* was cultured from the Asian malaria vector *Anopheles stephensi*. Here, we present the annotated draft genome sequence of this *S. maltophilia* strain. This genomic resource will facilitate further characterization of bacteria associated with mosquitoes.

Received 19 January 2016 Accepted 28 January 2016 Published 10 March 2016

Citation Hughes GL, Raygoza Garay JA, Koundal V, Rasgon JL, Mwangi MM. 2016. Genome sequence of *Stenotrophomonas maltophilia* strain SmAs1, isolated from the Asian malaria mosquito *Anopheles stephensi*. *Genome Announc* 4(2):e00086-16. doi:10.1128/genomeA.00086-16.

Copyright © 2016 Hughes et al. This is an open-access article distributed under the terms of the [Creative Commons Attribution 4.0 International license](https://creativecommons.org/licenses/by/4.0/).

Address correspondence to Jason L. Rasgon, jlr54@psu.edu.

Mosquitoes are known to harbor a diverse microbiome (1–4), which can influence many host phenotypes (5). Importantly, from a public health perspective, bacterial microbes are known to influence the ability of *Anopheles* mosquitoes to transmit both viral and apicomplexan pathogens (6–8), and microbial control approaches to controlling arthropod-borne disease are gaining considerable attention. While our appreciation of the influence of the microbiome on mosquitoes is expanding, there are limited genomic resources for bacteria that associate with the Asian malaria mosquito. Here, we report the draft genome sequence of a *Stenotrophomonas maltophilia* isolate, cultured from laboratory-reared *Anopheles stephensi* mosquitoes.

A homogenate of surface-sterilized *A. stephensi* (Liston strain) was used to inoculate LB agar plates, and a single colony was isolated and confirmed by 16S rRNA gene sequencing to be *S. maltophilia*. Genomic DNA was extracted using a Qiagen Blood and Tissue kit following the recommendation for bacteria. The sequencing was done in a 500-cycle run on an Illumina MiSeq at the Pennsylvania State University Genomics Core Facility. The DNA library was prepared using a Nextera XT DNA library preparation kit and had an insert size of 400 bp. The 250-bp paired-end reads were initially assembled using MIRA version 4.0, and the assembly was refined using DNASTar SeqMan Pro version 12.0. This resulted in a total of 32 contigs with a combined length of 4.0 Mbp, an N_{50} statistic of 166,372 bp, a median read coverage of 66×, and an average G+C content of 66%. The annotation was done using the RAST pipeline (9–11), followed by manual curation, yielding 3,625 protein-coding genes and 67 RNA genes.

One species of *Stenotrophomonas*, namely, *S. maltophilia*, is an emerging global multidrug-resistant opportunistic human pathogen (12). The RAST pipeline identified genes with homology to resistance proteins: multidrug/efflux pumps, 49; heavy metal-resistance proteins, 48; beta-lactamases, 10; acriflavin-resistance proteins, 3; macrolide efflux pumps, 3; ami-

noglycoside acetyltransferases, 2; aminoglycoside phosphotransferases, 1; fosmidomycin-resistance proteins, 1; and fusaric acid-resistance proteins, 1.

Stenotrophomonas spp. appear to associate with multiple insect species (13–16). The bacterial genome sequences in this and other papers (17, 18) will provide further resources to examine bacterial interactions within insects.

Nucleotide sequence accession number. This whole-genome shotgun project has been deposited at GenBank under the accession number [LFKU000000000](https://www.ncbi.nlm.nih.gov/nuclink/LFKU000000000).

ACKNOWLEDGMENTS

This work was supported by NIH grants AI111175 and AI116636 to J.L.R., the University of Texas Rising Star Award and a Brown Foundation grant to G.L.H., and start-up funds provided by the Pennsylvania State University to M.M.M.

FUNDING INFORMATION

This work, including the efforts of Jason L. Rasgon, was funded by HHS | National Institutes of Health (NIH) (AI116636 and AI111175). This work, including the efforts of Grant L. Hughes, was funded by University of Texas Medical Branch (UTMB). This work, including the efforts of Grant L. Hughes, was funded by Brown Foundation (Brown Foundation, Inc.). This work, including the efforts of Michael Mwangi, was funded by Pennsylvania State University (PSU).

The funders had no role in study design, data collection and interpretation, or the decision to submit the work for publication.

REFERENCES

- Osei Poku J, Mbogo CM, Palmer WJ, Jiggins FM. 2012. Deep sequencing reveals extensive variation in the gut microbiota of wild mosquitoes from Kenya. *Mol Ecol* 21:5138–5150. <http://dx.doi.org/10.1111/j.1365-294X.2012.05759.x>.
- Hughes GL, Dodson BL, Johnson RM, Murdock CC, Tsujimoto H, Suzuki Y, Patt AA, Cui L, Nossa CW, Barry RM, Sakamoto JM, Hornett

- EA, Rasgon JL. 2014. Native microbiome impedes vertical transmission of *Wolbachia* in *Anopheles* mosquitoes. *Proc Natl Acad Sci USA* 111: 12498–12503. <http://dx.doi.org/10.1073/pnas.1408888111>.
3. Wang Y, Gilbreath TM, Kukutla P, Yan G, Xu J. 2011. Dynamic gut microbiome across life history of the malaria mosquito *Anopheles gambiae* in Kenya. *PLoS One* 6:e24767. <http://dx.doi.org/10.1371/journal.pone.0024767>.
4. Gimonneau G, Tchioffo MT, Abate L, Boissière A, Awono-Ambéné PH, Nsango SE, Christen R, Morlais I. 2014. Composition of *Anopheles coluzzii* and *Anopheles gambiae* microbiota from larval to adult stages. *Infect Genet Evol* 28:715–724. <http://dx.doi.org/10.1016/j.meegid.2014.09.029>.
5. Coon KL, Vogel KJ, Brown MR, Strand MR. 2014. Mosquitoes rely on their gut microbiota for development. *Mol Ecol* 23:2727–2739. <http://dx.doi.org/10.1111/mec.12771>.
6. Carissimo G, Pondeville E, McFarlane M, Dietrich I, Mitri C, Bischoff E, Antoniewski C, Bourgouin C, Failloux A-B, Kohl A, Vernick KD. 2015. Antiviral immunity of *Anopheles gambiae* is highly compartmentalized, with distinct roles for RNA interference and gut microbiota. *Proc Natl Acad Sci USA* 112:E176–E185. <http://dx.doi.org/10.1073/pnas.1412984112>.
7. Hughes GL, Koga R, Xue P, Fukatsu T, Rasgon JL. 2011. *Wolbachia* infections are virulent and inhibit the human malaria parasite *Plasmodium falciparum* in *Anopheles gambiae*. *PLoS Pathog* 7:e1002043. <http://dx.doi.org/10.1371/journal.ppat.1002043>.
8. Cirimotich CM, Dong Y, Clayton AM, Sandiford SL, Souza-Neto JA, Mulenga M, Dimopoulos G. 2011. Natural microbe-mediated refractoriness to *Plasmodium* infection in *Anopheles gambiae*. *Science* 332: 855–858. <http://dx.doi.org/10.1126/science.1201618>.
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. 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. <http://dx.doi.org/10.1093/nar/gkt1226>.
11. Brettin T, Davis JJ, Disz T, Edwards RA, Gerdes S, Olsen GJ, Olson R, Overbeek R, Parrello B, Pusch GD, Shukla M, Thomason JA III, Stevens R, Vonstein V, Wattam AR, Xia F. 2015. RASTik: a modular and extensible implementation of the RAST algorithm for building custom annotation pipelines and annotating batches of genomes. *Sci Rep* 5:8365. <http://dx.doi.org/10.1038/srep08365>.
12. Brooke JS. 2012. *Stenotrophomonas maltophilia*: an emerging global opportunistic pathogen. *Clin Microbiol Rev* 25:2–41. <http://dx.doi.org/10.1128/CMR.00019-11>.
13. Indiragandhi P, Anandham R, Madhaiyan M, Kim G-H, Sa T. 2008. Cross-utilization and expression of outer membrane receptor proteins for siderophore uptake by diamondback moth *Plutella xylostella* (Lepidoptera: *Plutellidae*) gut bacteria. *FEMS Microbiol Lett* 289:27–33. <http://dx.doi.org/10.1111/j.1574-6968.2008.01350.x>.
14. Chung SH, Rosa C, Scully ED, Peiffer M, Tooker JF, Hoover K, Luthe DS, Felton GW. 2013. Herbivore exploits orally secreted bacteria to suppress plant defenses. *Proc Natl Acad Sci USA* 110:15728–15733. <http://dx.doi.org/10.1073/pnas.1308867110>.
15. Dickey A, Trease A, Jara-Cavieles A, Kumar V. 2014. Estimating bacterial diversity in *Scirtothrips dorsalis* (Thysanoptera: Thripidae) via next generation sequencing. *Fla Entomol* 97:362–366.
16. Gouveia C, Asensi MD, Zahner V, Rangel EF, de Oliveria SMP. 2008. Study on the bacterial midgut microbiota associated to different Brazilian populations of *Lutzomyia longipalpis* (Lutz & Neiva) (Diptera: Psychodidae). *Neotrop Entomol* 37:597–601. <http://dx.doi.org/10.1590/S1519-566X2008000500016>.
17. Raygoza Garay JA, Hughes GL, Koundal V, Rasgon JL, Mwangi MM. 2016. Genome sequence of *Elizabethkingia anophelis* strain EaAs1, isolated from the Asian malaria mosquito *Anopheles stephensi*. *Genome Announc* 4(2):e00084–16. <http://dx.doi.org/10.1128/genomeA.00084-16>.
18. Hughes GL, Raygoza Garay JA, Koundal V, Rasgon JL, Mwangi MM. 2016. Genome sequences of *Staphylococcus hominis* strains ShAs1, ShAs2, and ShAs3, isolated from the Asian malaria mosquito *Anopheles stephensi*. *Genome Announc* 4(2):e00085–16. <http://dx.doi.org/10.1128/genomeA.00085-16>.