AMERICAN SOCIETY FOR MICROBIOLOGY | genomeAnnouncements™

PROKARYOTES

Draft Genome Sequence of *Epilithonimonas* sp. FP211-J200, Isolated from an Outbreak Episode on a Rainbow Trout (*Oncorhynchus mykiss*) Farm

Manuel Ayala,[a,b] Cristopher Segovia,[a,c] Rodrigo Rojas,[d,e] Claudio Miranda,[d,e] Javier Santander[a,c]

Marine Microbial Pathogenesis and Vaccinology Laboratory, Department of Ocean Sciences, Memorial University of Newfoundland, St. John's, Canada[a]; Faculty of Biological Sciences, Universidad Nacional de San Marcos, Lima, Perú[b]; Faculty of Sciences, Universidad Mayor, Huechuraba, Chile[c]; Departamento de Acuicultura, Universidad Católica del Norte, Coquimbo, Chile[d]; Centro AquaPacífico, Coquimbo, Chile[e]

ABSTRACT Here, we report the draft genome sequence of *Epilithonimonas* sp. FP211-J200, isolated from rainbow trout head kidney cells. The size of the genome is 4,110,772 bp, with a G+C content of 37.1%. The *Epilithonimonas* sp. FP211-J200 genome has genes related to tetracycline and β-lactam resistance. This is the first reported *Epilithonimonas* species genome isolated from a fish host.

Received 30 July 2017 **Accepted** 7 August 2017 **Published** 14 September 2017

Citation Ayala M, Segovia C, Rojas R, Miranda C, Santander J. 2017. Draft genome sequence of *Epilithonimonas* sp. FP211-J200, isolated from an outbreak episode on a rainbow trout (*Oncorhynchus mykiss*) farm. Genome Announc 5:e00819-17. https://doi.org/10.1128/genomeA.00819-17.

Address correspondence to Javier Santander, jsantander@mun.ca.

The genus *Epilithonimonas* (1) belongs to the phylum *Bacteriodetes*, family *Flavobacteriaceae*, which also includes the *Flavobacterium* and *Chryseobacterium* genera. Currently, five species of *Epilithonimonas* have been described (2), including *E. tenax* (1), *E. ginsengisoli* (3), *E. lactis* (4), *E. xixisoli* (5), and *E. psychrotolerans* (6). *Epilithonimonas* sp. strains have been isolated from different environments, including soil (3, 6), freshwater (1, 5), and milk (4). Members of the genus *Epilithonimonas* are chemoorganotrophs and most likely play a role in natural carbon cycles in low-salinity ecosystems, such as soil and freshwater (7). However, associations of *Epilithonimonas* spp. with animals like fish are unknown. Here, we report the draft genome sequence of *Epilithonimonas* sp. FP211-J200, a yellow-pigmented strain isolated from rainbow trout (*Oncorhynchus mykiss*) head kidney cells during a flavobacteriosis outbreak in a freshwater aquaculture facility at X Region, Chile.

Epilithonimonas sp. FP211-J200 was routinely grown in tryptone-yeast extract-salt (TYES) (8) with aeration (180 rpm) at 28°C. The genomic DNA was extracted according to Wilson (9) and purified using silica (10). Sequencing was performed using the next-generation sequencer (NGS) Illumina MiSeq platform (Universidad Mayor, Center for Genomics and Bioinformatics, Huechuraba, Chile) and paired-end libraries. Low-quality sequences were examined by FastQC version 0.10.1 (11). The sequences were trimmed and assembled using the CLC Genomics Workbench 9.0.1 (Qiagen) *de novo* tool, resulting in 83 contigs over 1 kb, with an N_{50} value of 98,567 bp. The total length of the draft genome of the *Epilithonimonas* sp. FP211-J200 was 4,110,772 bp, with a G+C content of 37.01%.

The assembled sequences were annotated by the National Center for Biotechnology Information (NCBI) Prokaryotic Genome Annotation Pipeline (PGAP [https://www.ncbi.nlm.nih.gov/genome/annotation_prok/]). The tRNA genes were detected by tRNAscan-SE version 1.3 (12) and the rRNA with RNAmmer (13). A total of 3,882 coding sequences (CDSs), 88 pseudogenes, 1 complete rRNA operon (5S-16S-23S), 41 tRNAs, and 2 noncoding RNAs (ncRNAs) were predicted by the pipeline.

Genes encoding proteins for resistance to antibiotics were identified using the Comprehensive Antibiotic Resistance Database (CARD) (14). *Epilithonimonas* sp. FP211-J200 presented genes potentially responsible for antibiotic resistance. We found three types of

multidrug efflux transporters, including *acrB*, *mexD*, and *adeB* (15). Also, we found the flavin-dependent monooxygenase *tetX* gene responsible for resistance to all clinical relevant tetracyclines (16). Additionally, we found the β-lactamase TLA-2, also present in *Chryseobacterium gleum* (17), and the metallo-β-lactamase GOB-1, also present in *Chryseobacterium meningoseptica* (18). Also, we identified the *crfA* gene, which is related to florfenicol resistance.

We identified several genes related to iron acquisition and virulence, including hemolysin, hemolysin III, ferritin, ferric siderophore ABC transporter substrate-binding protein, ferredoxin, hemin receptor, and the Fur transcriptional regulator, suggesting that *Epilithonimonas* sp. FP211-J200 might have pathogenesis potential.

Phylogenetic reconstruction using 16S rRNA showed that the *Epilithonimonas* sp. FP211-J200 strain is closely related to the genus *Chryseobacterium*. *In silico* DNA-DNA hybridization (http://cbrc.kaust.edu.sa/dna_hybridization/index.html) showed that strain FP211-J200 is different from other *Epilithonimonas* spp. and *Chryseobacterium* spp. The strain most closely related to FP211-J200 was *E. ginsengisoli*. This is the first reported *Epilithonimonas* sp. genome isolated from a fish host.

Accession number(s). The whole-genome shotgun project (BioProject PRJNA310285) has been deposited at DDBJ/EMBL/GenBank under the accession number LSHB00000000. The version described in this paper is version LSHB01000000.

ACKNOWLEDGMENTS

This work was funded by grant no. 1140330 from FONDECYT-Chile and R&D-Ignite Project no. 5404.2113.10 from the Research & Development Corporation (RDC) of Newfoundland and Labrador.

We thank Carolina Sanchez (Center for Genomics and Bioinformatics, Universidad Mayor) and Mario Moreno (Center for Genomics and Bioinformatics, Universidad Mayor) for their assistance at the sequencing facility and M. Ignacia Diaz (FONDECYT 1140330) for her logistic support.

REFERENCES

1. O'Sullivan LA, Rinna J, Humphreys G, Weightman AJ, Fry JC. 2006. Culturable phylogenetic diversity of the phylum "*Bacteroidetes*" from river epilithon and coastal water and description of novel members of the family *Flavobacteriaceae*: *Epilithonimonas tenax* gen. nov., sp. nov. and *Persicivirga xylanidelens* gen. nov., sp. nov. Int J Syst Evol Microbiol 56:169–180. https://doi.org/10.1099/ijs.0.63941-0.
2. Euzéby JP. 1997. List of bacterial names with standing in nomenclature: a folder available on the Internet. Int J Syst Bacteriol 47:590–592. https://doi.org/10.1099/00207713-47-2-590.
3. Hoang VA, Kim YJ, Ponnuraj SP, Nguyen NL, Hwang KH, Yang DC. 2015. *Epilithonimonas ginsengisoli* sp. nov., isolated from soil of a ginseng field. Int J Syst Evol Microbiol 65:122–128. https://doi.org/10.1099/ijs.0.065466-0.
4. Shakéd T, Hantsis-Zacharov E, Halpern M. 2010. *Epilithonimonas lactis* sp. nov., isolated from raw cow's milk. Int J Syst Evol Microbiol 60:675–679. https://doi.org/10.1099/ijs.0.012575-0.
5. Feng H, Zeng Y, Huang Y. 2014. *Epilithonimonas xixisoli* sp. nov., isolated from wetland bank-side soil. Int J Syst Evol Microbiol 64:4155–4159. https://doi.org/10.1099/ijs.0.065771-0.
6. Ge L, Zhao Q, Sheng H, Wu J, An L. 2015. *Epilithonimonas psychrotolerans* sp. nov., isolated from alpine permafrost. Int J Syst Evol Microbiol 65:3777–3781. https://doi.org/10.1099/ijsem.0.000489.
7. Kirchman DL, Yu L, Cottrell MT. 2003. Diversity and abundance of uncultured *Cytophaga*-like bacteria in the Delaware estuary. Appl Environ Microbiol 69:6587–6596. https://doi.org/10.1128/AEM.69.11.6587-6596.2003.
8. Cain KD, Lafrentz BR. 2007. Laboratory maintenance of *Flavobacterium psychrophilum* and *Flavobacterium columnare*. Curr Protoc Microbiol Chapter 13:Unit 13B.1. https://doi.org/10.1002/9780471729259.mc13b01s6.
9. Wilson K. 2001. Preparation of genomic DNA from bacteria. Curr Protoc Mol Biol Chapter 2:Unit 2.4. https://doi.org/10.1002/0471142727.mb0204s56.
10. Boom R, Sol C, Beld M, Weel J, Goudsmit J, Wertheim-van Dillen P. 1999. Improved silica-guanidiniumthiocyanate DNA isolation procedure based on selective binding of bovine alpha-casein to silica particles. J Clin Microbiol 37:615–619.
11. Andrews S. 2010. FastQC: a quality control tool for high throughput sequence data. Available online at: http://www.bioinformatics.babraham.ac.uk/projects/fastqc.
12. 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.
13. 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. https://doi.org/10.1093/nar/gkm160.
14. McArthur AG, Waglechner N, Nizam F, Yan A, Azad MA, Baylay AJ, Bhullar K, Canova MJ, De Pascale G, Ejim L, Kalan L, King AM, Koteva K, Morar M, Mulvey MR, O'Brien JS, Pawlowski AC, Piddock LJ, Spanogiannopoulos P, Sutherland AD, Tang I, Taylor PL, Thaker M, Wang W, Yan M, Yu T, Wright GD. 2013. The comprehensive antibiotic resistance database. Antimicrob Agents Chemother 57:3348–3357. https://doi.org/10.1128/AAC.00419-13.
15. Sun J, Deng Z, Yan A. 2014. Bacterial multidrug efflux pumps: mechanisms, physiology and pharmacological exploitations. Biochem Biophys Res Commun 453:254–267. https://doi.org/10.1016/j.bbrc.2014.05.090.
16. Volkers G, Palm GJ, Weiss MS, Wright GD, Hinrichs W. 2011. Structural basis for a new tetracycline resistance mechanism relying on the TetX monooxygenase. FEBS Lett 585:1061–1066. https://doi.org/10.1016/j.febslet.2011.03.012.
17. Girlich D, Poirel L, Schlüter A, Nordmann P. 2005. TLA-2, a novel Ambler class A expanded-spectrum beta-lactamase. Antimicrob Agents Chemother 49:4767–4770. https://doi.org/10.1128/AAC.49.11.4767-4770.2005.
18. Bellais S, Aubert D, Naas T, Nordmann P. 2000. Molecular and biochemical heterogeneity of class B carbapenem-hydrolyzing beta-lactamases in *Chryseobacterium meningosepticum*. Antimicrob Agents Chemother 44:1878–1886. https://doi.org/10.1128/AAC.44.7.1878-1886.2000.