genomeA
Journals.ASM.org

Complete Genome of the Attenuated Sparfloxacin-Resistant *Streptococcus agalactiae* Strain 138spar

Julia W. Pridgeon,[a] Dunhua Zhang,[a] Lee Zhang[b]

Aquatic Animal Health Research Unit, USDA, ARS, Auburn, Alabama, USA[a]; Genomics and Sequencing Laboratory, Department of Entomology and Plant Pathology, Auburn University, Auburn, Alabama, USA[b]

Through the selection of resistance to sparfloxacin, an attenuated *Streptococcus agalactiae* strain, 138spar, was obtained from its virulent parent strain, *S. agalactiae* 138P. The full genome of *S. agalactiae* 138spar is 1,838,126 bp. This genome will allow comparative genomics to identify genes associated with virulence, antibiotic resistance, or other characteristics.

Received 15 April 2014 **Accepted** 1 May 2014 **Published** 15 May 2014

Citation Pridgeon JW, Zhang D, Zhang L. 2014. Complete genome of the attenuated sparfloxacin-resistant *Streptococcus agalactiae* strain 138spar. Genome Announc. 2(3): e00431-14. doi:10.1128/genomeA.00431-14.

Address correspondence to Julia W. Pridgeon, Julia.Pridgeon@ars.usda.gov.

The Gram-positive bacterium *Streptococcus agalactiae* (group B streptococcus [GBS]) causes streptococcosis in fish (1), resulting in significant economic losses. For example, GBS was responsible for a massive disease outbreak in the Kuwait Bay in 2011, killing 2,500 metric tons of wild mullet (*Liza klunzingeri*) (2). In addition, large-scale streptococcal outbreaks occurred frequently in tilapia farms in China from 2009 to 2011, involving >95% of farms, with a cumulative mortality rate of 30 to 80% (3–5). GBS also causes meningitis (6–8), neonatal sepsis (9), and pneumonia (10) in humans. GBS is a major cause of bovine mastitis, a dominant health disorder affecting milk production (11, 12). A virulent GBS strain, *S. agalactiae* 138P, was isolated from infected Nile tilapia during a 2007 disease outbreak in Idaho. To develop an attenuated bacterial vaccine, a sparfloxacin resistance strategy was used to modify GBS strain 138P, resulting in the isolation of an attenuated GBS 138spar (13) that was 2,048-fold resistant to sparfloxacin. However, whether changes occurred at the genomic DNA level is unknown. Therefore, the whole-genome sequence of GBS 138spar was determined in this study.

The genome of GBS 138spar was sequenced using the Illumina 1500 HiSeq platform. BioNumerics (Applied Maths) was used to assemble a total of 2,743,316 sequence reads, with an average length of 100 bp. The whole genome of the virulent GBS strain 138P was reported recently (14). Using similar assembly methods as described previously (14), the whole genome of GBS 138spar was obtained, which is 1,838,126 bp in length. The RAST server (15) predicted 1,892 coding sequences, including all the subsystems reported in its parent strain, GBS 138P (14). RNAmmer (16) predicted 5 copies of 5S rRNA, 6 copies of 16S rRNA, and 6 copies of 23S rRNA, similar to those in the genome of GBS 138P and in the reference genome GBS 2-22 (GenBank accession no. FO393392). Compared to the whole genome of GBS 138P (14), GBS 138spar has a deletion >6 bp in 22 places. In addition, single-nucleotide changes (deletion, insertion, and point mutation) were found in 26 places in the GBS 138P genome compared to in the GBS 138spar genome.

Nucleotide sequence accession number. The complete genome sequence of *S. agalactiae* 138spar was deposited in GenBank under the accession no. CP007565.

ACKNOWLEDGMENTS

This study was supported by the USDA/ARS CRIS project 6420-32000-024-00D.

The use of trade, firm, or corporate names in this publication is for the information and convenience of the reader. Such use does not constitute an official endorsement or approval by the United States Department of Agriculture or the Agricultural Research Service of any product or service to the exclusion of others that may be suitable.

We thank Dehai Xu (USDA-ARS) and Sang-Jin Suh (Auburn University) for critical reviews of the manuscript. We thank Beth Peterman (USDA-ARS) for her technical support.

REFERENCES

1. **Liu G, Zhang W, Lu C.** 2012. Complete genome sequence of *Streptococcus agalactiae* GD201008-001, isolated in China from tilapia with meningoencephalitis. J. Bacteriol. **194:**6653. http://dx.doi.org/10.1128/JB.01788-12.
2. **Glibert PM, Landsberg JH, Evans JJ, Al-Sarawi MA, Faraj M, Al-Jarallah MA, Haywood A, Ibrahem S, Klesius P, Powell C, Shoemaker C.** 2002. A fish kill of massive proportion in Kuwait Bay, Arabian Gulf, 2001: the roles of bacterial disease, harmful algae, and eutrophication. Harmful Algae **1:**215–231. http://dx.doi.org/10.1016/S1568-9883(02)00013-6.
3. **Chen M, Wang R, Li LP, Liang WW, Li J, Huang Y, Lei AY, Huang WY, Gan X.** 2012. Screening vaccine candidate strains against *Streptococcus agalactiae* of tilapia based on PFGE genotype. Vaccine **30:**6088–6092. http://dx.doi.org/10.1016/j.vaccine.2012.07.044.
4. **Chen M, Li LP, Wang R, Liang WW, Huang Y, Li J, Lei AY, Huang WY, Gan X.** 2012. PCR detection and PFGE genotype analyses of streptococcal clinical isolates from tilapia in China. Vet. Microbiol. **159:**526–530. http://dx.doi.org/10.1016/j.vetmic.2012.04.035.
5. **Ye X, Li J, Lu M, Deng G, Jiang X, Tian Y, Quan Y, Jian Q.** 2011. Identification and molecular typing of *Streptococcus agalactiae* isolated from pond-cultured tilapia in China. Fish. Sci. **77:**623–632. http://dx.doi.org/10.1007/s12562-011-0365-4.
6. **Sanz-Rojas P, Cabeza-Osorio L, Hermosa C, Serrano-Heranz R.** 2013. Acute meningitis by *Streptococcus agalactiae* in an immunocompetent male. Rev. Esp. Quimioter. **26:**78–79. (In Spanish.).

7. **Reguera A, González MÁ, Sánchez A, Aguayo C.** 2011. *Streptococcus agalactiae* meningitis in an immunocompetent male. Enferm. Infecc. Microbiol. Clin. **29:**707–708. (In Spanish.) http://dx.doi.org/10.1016/j.eimc.2011.05.020.
8. **Martín Diaz RM, Ruiz-Giardín JM, Gonzalo Pascua S, Canora Lebrato J.** 2010. Meningitis by *S. agalactiae* in a non-pregnant immunocompetent woman. Rev. Clín. Esp. **210:**591–592. (In Spanish.) http://dx.doi.org/10.1016/j.rce.2010.05.013.
9. **Jawa G, Hussain Z, da Silva O.** 2013. Recurrent late-onset group B streptococcus sepsis in a preterm infant acquired by expressed breast milk transmission: a case report. Breastfeed Med. **8:**134–136. http://dx.doi.org/10.1089/bfm.2012.0016.
10. **Villena Ruiz MA, Olalla Sierra J, de la Torre Lima J, García-Alegría J.** 2009. *Streptococcus agalactiae* induced cavitated pneumonia. Rev. Clín. Esp. **209:**252–254. (In Spanish.) http://dx.doi.org/10.1016/S0014-2565(09)71245-7.
11. **Katholm J, Rattenborg E.** 2010. The surveillance program of *Streptococcus agalactiae* in Danish dairy herds, p 1989–2008. *In* Hillerton J (ed), Proceedings of the 5th IDF Mastitis Conference. 2010. Mastitis Res. Into Practice. N Z Veterinary Association Vet. Learn. Christchurch, NZ.
12. **Richards VP, Lang P, Bitar PD, Lefébure T, Schukken YH, Zadoks RN, Stanhope MJ.** 2011. Comparative genomics and the role of lateral gene transfer in the evolution of bovine adapted *Streptococcus agalactiae*. Infect. Genet. Evol. **11:**1263–1275. http://dx.doi.org/10.1016/j.meegid.2011.04.019.
13. **Pridgeon JW, Klesius PH.** 2013. Development of live attenuated *Streptococcus agalactiae* as potential vaccines by selecting for resistance to sparfloxacin. Vaccine **31:**2705–2712. http://dx.doi.org/10.1016/j.vaccine.2013.03.066.
14. **Pridgeon JW, Zhang D.** 2014. Complete genome sequence of a virulent *Streptococcus agalactiae* strain, 138P, isolated from diseased Nile tilapia. Genome Announc. **2**(2):e00295-14. http://dx.doi.org/10.1128/genomeA.00295-14.
15. **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.
16. **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.