5-21-2020

Draft Genome Sequence of Lactobacillus mulieris UMB7784, Isolated from the Female Urinary Tract

Oleksandra Tsibere  
*Loyola University Chicago*

Taylor Miller-Ensminger  
*Loyola University Chicago*

Adelina Voukadinova  
*Loyola University Chicago*

Alan J. Wolfe  
*Loyola University Chicago*, awolfe@luc.edu

Catherine Putonti  
*Loyola University Chicago*, cputonti@luc.edu

Follow this and additional works at: https://ecommons.luc.edu/bioinformatics_facpub

Part of the Bioinformatics Commons, and the Biology Commons

---

**Recommended Citation**

Tsibere, Oleksandra; Miller-Ensminger, Taylor; Voukadinova, Adelina; Wolfe, Alan J.; and Putonti, Catherine. Draft Genome Sequence of Lactobacillus mulieris UMB7784, Isolated from the Female Urinary Tract. Microbiology Resource Announcements, 9, : , 2020. Retrieved from Loyola eCommons, Bioinformatics Faculty Publications, http://dx.doi.org/10.1128/MRA.00403-20

This Article is brought to you for free and open access by the Faculty Publications and Other Works by Department at Loyola eCommons. It has been accepted for inclusion in Bioinformatics Faculty Publications by an authorized administrator of Loyola eCommons. For more information, please contact ecommons@luc.edu.

This work is licensed under a Creative Commons Attribution 4.0 License.  
© Tsibere et al., 2020
Draft Genome Sequence of *Lactobacillus mulieris* UMB7784, Isolated from the Female Urinary Tract

Oleksandra Tsibere,a Taylor Miller-Ensminger,b Adelina Voukadinova,b Alan J. Wolfe,c Catherine Putontia,b,c,d

aDepartment of Biology, Loyola University Chicago, Chicago, Illinois, USA
bBioinformatics Program, Loyola University Chicago, Chicago, Illinois, USA
cDepartment of Microbiology and Immunology, Stritch School of Medicine, Loyola University Chicago, Maywood, Illinois, USA
dDepartment of Computer Science, Loyola University Chicago, Chicago, Illinois, USA

**ABSTRACT** We sequenced the genome of *Lactobacillus mulieris* UMB7784, isolated from the urinary tract. With a genome size of 1,695,489 bp and a GC content of 34.2%, the draft genome sequence presented here expands our understanding of lactobacilli in the female bladder.

*Lactobacillus* species are native members of the female urinary microbiota (1–3), and their presence is associated with a healthy urinary tract (4, 5). Some *Lactobacillus* species can inhibit the growth of uropathogens by releasing biosurfactants with antimicrobial properties (6, 7). One such species, *Lactobacillus jensenii*, releases biosurfactants that disrupt biofilms of pathogens, such as *Enterobacter aerogenes* and *Escherichia coli* (8). In March 2020, *L. jensenii* was split into two species—*L. jensenii* and *L. mulieris*—based upon average nucleotide identity (9). The type strain for the new species *L. mulieris* was isolated from a urine sample, and here, we present the genome sequence of another urinary isolate of this new lineage. *L. mulieris* UMB7784 was isolated from the catheterized urine of a woman with a recurrent urinary tract infection (UTI).

Catheterized urine samples were collected from women as part of a prior institutional review board (IRB)-approved study (University of California, San Diego, IRB no. 170077AW). *L. mulieris* UMB7784 was isolated using the expanded quantitative urinary culture (EQUC) protocol (10). The genus and species were identified by matrix-assisted laser desorption ionization–time of flight (MALDI-TOF) mass spectrometry, following the protocol previously detailed (10), prior to storage at −80°C. The isolate was streaked onto a Columbia nalidixic acid (CNA) agar plate and incubated for 24 hours at 35°C in 5% CO₂. A single colony was transferred to liquid MRS medium supplemented with 1 ml/liter of Tween 80 and grown for 48 hours at 35°C in 5% CO₂. DNA was extracted using the DNeasy blood and tissue kit. Some modifications were made to the manufacturer's Gram-positive procedure, as follows: 230 μl of lysis buffer (180 μl of 20 mM Tris-Cl, 2 mM sodium EDTA, and 1.2% Triton X-100 and 50 μl of lysozyme) was used (step 2), and the sample was incubated at 56°C for 10 minutes (step 5). DNA was quantified using a Qubit fluorometer. Sequencing was performed by the Microbial Genome Sequencing Center (MiGS) at the University of Pittsburgh. DNA was first enzymatically fragmented using an Illumina tagmentation enzyme, and indices were attached using PCR. Sequencing was conducted on the Illumina NextSeq 550 platform, producing 2,368,077 pairs of 150-bp reads. Raw reads were trimmed using Sickle v1.33 (https://github.com/najoshi/sickle) and assembled using SPAdes v3.13.0 with the “only-assembler” option for k values of 55, 77, 99, and 127 (11). The coverage was calculated using BBMap v38.47 (https://sourceforge.net/projects/bbmap/). The genome was annotated using PATRIC v3.6.3 (12) and the NCBI Prokaryotic Genome Annotation Pipeline.
The L. mulieris UMB7784 draft genome is 1,695,489 bp long in 42 contigs with a 345× genome coverage and an N₅₀ score of 67,456 bp. The genomic GC content is 34.2%. PGAP annotation identified 1,451 protein-coding genes, 53 tRNA genes, and 5 rRNA genes. PHASTER (14) analysis revealed 1 incomplete phage, and CRISPRCasFinder v1.1.2 (15) detected 1 CRISPR array with 40 spacer sequences. Since our strain was isolated from the urine of a woman with a UTI, further analysis will be useful for studying the antimicrobial properties of L. mulieris. Future work will expand our understanding of the prevalence and role(s) of this new species within the female urinary tract.

**Data availability.** This whole-genome shotgun project has been deposited in GenBank under the accession no. JAAVSF000000000. The version described in this paper is the first version, JAAVSF010000000. Raw sequence data are publicly available under accession no. SRR11441026.

**ACKNOWLEDGMENTS**

This work was conducted as part of Loyola University Chicago’s Department of Biology Graduate course. For prior patient recruitment, we acknowledge the Loyola University Education and Research Collaborative (LUEREC) and the patients who provided the samples for this study.

**REFERENCES**

1. Curtiss N, Balachandran A, Krska L, Peppiatt-Wildman C, Wildman S, DUCKETT J. 2017. A case controlled study examining the bladder microbiome in women with overactive bladder (OAB) and healthy controls. Eur J Obstet Gynecol Reprod Biol 214:31–35. https://doi.org/10.1016/j.ejogrb.2017.04.040.

2. Fok CS, Gao X, Lin H, Thomas-White KJ, Mueller ER, Wolfe AJ, Dong Q, Brubaker L. 2018. Urinary symptoms are associated with certain urinary microbes in urogynecologic surgical patients. Int Urogynecol J 29:1765–1771. https://doi.org/10.1007/s00192-018-3732-1.

3. Pearce MM, Hilt EE, Rosenfeld AB, Zilliox MJ, Thomas-White K, Fok C, Klettermènes S, Schreckenberger PC, Brubaker L, Gai X, Wolfe AJ. 2014. The female urinary microbiome: a comparison of women with and without urgency urinary incontinence. mBio 5:e01283-14. https://doi.org/10.1128/mbio.01283-14.

4. Fouts DE, Pieper R, Szpakowski S, Pohl H, Knoblach S, Suh M-J, Huang ST-J, Ljungberg I, Sprague BM, Lucas SK, Torralba M, Nelson KE, Groah SL. 2012. Integrated next-generation sequencing of 16S rDNA and metaproteomics differentiate the healthy urine microbiome from asymptomatic bacteriuria in neuropathic bladder associated with spinal cord injury. J Transl Med 10:174. https://doi.org/10.1186/1475-2876-10-174.

5. Govender Y, Gabriel I, Minassian V, Fichorova R. 2019. The current evidence on the association between the urinary microbiome and urinary incontinence in women. Front Cell Infect Microbiol 9:133. https://doi.org/10.3389/fcimb.2019.00133.

6. Vefradis MMC, van der Mei HC, Reid G, Busscher HJ. 1996. Physicochemical and biochemical characterization of biosurfactants released by Lactobacillus strains. Colloids Surf B Biointerfaces 8:51–61. https://doi.org/10.1016/0927-7757(96)01297-0.

7. Barrons R, Tassone D. 2008. Use of Lactobacillus probiotics for bacterial genitourinary infections in women: a review. Clin Ther 30:453–468. https://doi.org/10.1016/j.clinthera.2008.03.013.

8. Morais IMC, Cordeiro AL, Teixeira GS, Domingues VS, Nardi RMD, Monteiro AS, Alves RJ, Siqueira EP, Santos VL. 2017. Biological and physicochemical properties of biosurfactants produced by Lactobacillus jensenii P6A and Lactobacillus gasseri P65. Microb Cell Fact 16:155. https://doi.org/10.1186/s12934-017-0769-7.

9. Rocha J, Botelho J, Ksiezarek M, Perovic SU, Machado M, Carriço JA, Pimentel LL, Salsinha S, Rodríguez-Alcalá LM, Pintoado M, Ribeiro TG, Peixe L. 2020. Lactobacillus mulieris sp. nov., a new species of Lactobacillus delbrueckii group. Int J Syst Evol Microbiol 70:1522–1527. https://doi.org/10.1099/ijsem.0.003901.

10. Hilt EE, McKinley K, Pearce MM, Zilliox MJ, Mueller ER, Brubaker L, Gai X, Wolfe AJ, Schreckenberger PC. 2014. Urine is not sterile: use of enhanced urine culture techniques to detect resident bacterial flora in the adult female bladder. J Clin Microbiol 52:871–876. https://doi.org/10.1128/JCM.02786-13.

11. Banekvich A, Nurk S, Antipov D, Gurevich AA, Dvorkin M, Kulikov AS, Lesin VM, Nikolenko SI, Pham S, Pjibelski AD, Poshyvanyk D, Sidorenko V, Vyahhi N, Tesler G, Alekseyev MA, Pevzner PA. 2012. SPAdes: a new genome assembly algorithm and its applications to single-cell sequencing. J Comput Biol 19:455–477. https://doi.org/10.1089/cmb.2012.0021.

12. 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. RASTTk: a modular and extensible implementation of the RAST algorithm for building custom annotation pipelines and annotating batches of genomes. Sci Rep 5:8365. https://doi.org/10.1038/srep08365.

13. 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.

14. Arndt D, Grant JR, Marcu A, Sajed T, Pon A, Liang Y, Wishart DS. 2016. PHASTER: a better, faster version of the PHAST phage search tool. Nucleic Acids Res 44:W16–W21. https://doi.org/10.1093/nar/gkw387.

15. Couvin D, Bernheim A, Toffano-Nioche C, Touchon M, Michalk J, Néron B, Rocha EPC, Vergnaud G, Gautheret D, Pourcel C. 2018. CRISPRCasFinder, an update of CRISPRFinder, includes a portable version, enhanced performance and integrates search for Cas proteins. Nucleic Acids Res 46:W246–W251. https://doi.org/10.1093/nar/gky425.