

Draft Genome Sequences of Six Lactobacillus pentosus Strains Isolated from Brines of Traditionally Fermented Spanish-Style Green Table Olives

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ABSTRACT Here, we report the genome sequences of six Lactobacillus pentosus strains isolated from traditional noninoculated Spanish-style green table olive brines. The total genome sizes varied between 3.77 and 4.039 Mbp. These genome sequences will assist in revealing the genes responsible for both technological and probiotic properties of these strains.

The genus *Lactobacillus* comprises many species indigenous to food-related habitats,
such as dairy products, meats, sausages, and fermented vegetables [\(1,](#page-1-0) [2\)](#page-1-1). Moreover, it has been shown that some species have potential probiotic properties, thus providing health benefits to human consumers [\(3,](#page-1-2) [4\)](#page-1-3). The Lactobacillus pentosus species is the main inhabitant in the economically important Spanish-style green olive fermentation process [\(5\)](#page-1-4). This lactic acid bacterium contributes to the preservation of olives by means of the large amounts of lactic acid produced, as well as by producing antibacterial compounds such us bacteriocins [\(6\)](#page-1-5). Spanish-style green olive fermentation usually relies on a spontaneous, traditional method in which both the olives and the environment are handled in order to favor the development of L. pentosus [\(7\)](#page-1-6). However, as the use of starter cultures is not a common practice in this fermentation, undesirable microorganisms frequently dominate the environment, and then spoilage of olives may occur. Thus, the genome sequences reported here will be highly valuable not only for designing appropriated starter cultures for olive fermentation but also for deepening the knowledge of potentially probiotic features shown by the L. pentosus species.

Here, the sequenced genomes of six L. pentosus strains (named L. pentosus IG2 to IG7) isolated from the brine of natural Spanish-style green olive fermentation are presented. The genomic DNA from the strains was extracted using a modification of the protein "salting-out" procedure [\(8\)](#page-1-7). Genome libraries were constructed using a TruSeq DNA PCR-free library preparation kit (Illumina, Inc.), with an insert size of 350 bp, and sequenced at Macrogen, Inc., (Seoul, Republic of Korea) by using an Illumina HiSeq platform with paired-end sequencing of 2 \times 101-bp read lengths. The genomes were assembled with Velvet 1.2.10 [\(9\)](#page-1-8), and parameters were optimized with VelvetOptimiser 2.2.5 [\(9\)](#page-1-8). The NCBI Prokaryotic Genome Annotation Pipeline (PGAP) was used to annotate the six strains, and the annotation was completed by use of the following protocol: protein-coding genes were predicted using Prodigal v 2.6.3, which is an accurate prokaryotic gene finder [\(10\)](#page-1-9), and then they were functionally annotated by Sma3s v2 using UniProt bacteria to get a higher sensitivity [\(11\)](#page-1-10). To annotate noncoding 2018 **Published** 3 May 2018 **Citation** Calero-Delgado B, Martín-Platero AM,

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genes, Infernal 1.1.2 [\(12\)](#page-1-11) was used with Rfam database 13.0 [\(13\)](#page-1-12). To estimate the number of plasmids appearing in each strain, the contig sequences were compared to all the plasmid sequences from *Lactobacillus* species available in the RefSeq database.

The total genome sizes ranged from 3.77 to 4.039 Mbp, thus representing the largest sequenced genomes among L. pentosus strains to date, with very similar $G+C$ contents, which varied from 45.78% to 45.98%. We found several plasmids per strain, ranging from 6 in the IG4 strain to 13 in the IG3 strain. The chromosomes encode from 84 to 92 tRNAs and from 5 to 9 rRNAs. These data show the high genome plasticity of this lactic acid bacterial species.

Accession number(s). The genome sequences of the six L. pentosus strains have been stored under NCBI BioProject number PRJNA436944, and genome information and the GenBank accession number for each strain are listed in [Table 1.](#page-1-13)

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REFERENCES

- 1. Hui YH (ed). 2012. Handbook of plant-based fermented food and beverage technology. CRC Press, Boca Raton, FL.
- 2. Wolfe BE, Dutton RJ. 2015. Fermented foods as experimentally tractable microbial ecosystems. Cell 161:49 –55. [https://doi.org/10.1016/j.cell.2015](https://doi.org/10.1016/j.cell.2015.02.034) [.02.034.](https://doi.org/10.1016/j.cell.2015.02.034)
- 3. Watson RR, Preedly VR (ed). 2016. Probiotics, prebiotics, and synbiotics: bioactive foods in health promotion. Academic Press, London, UK.
- 4. Benítez-Cabello A, Bautista-Gallego J, Garrido-Fernández A, Rantsiou K, Cocolin L, Jiménez-Díaz R, Arroyo-López FN. 2016. RT-PCR-DGGE analysis to elucidate the dominant bacterial species of industrial Spanish-style green table olive fermentations. Front Microbiol 7:1291. [https://doi.org/](https://doi.org/10.3389/fmicb.2016.01291) [10.3389/fmicb.2016.01291.](https://doi.org/10.3389/fmicb.2016.01291)
- 5. Domínguez-Manzano J, León-Romero Á, Olmo-Ruiz C, Bautista-Gallego J, Arroyo-López FN, Garrido-Fernández A, Jiménez-Díaz R. 2012. Biofilm formation on abiotic and biotic surfaces during Spanish style green table olive fermentation. Int J Food Microbiol 157:230 –238. [https://doi.org/10](https://doi.org/10.1016/j.ijfoodmicro.2012.05.011) [.1016/j.ijfoodmicro.2012.05.011.](https://doi.org/10.1016/j.ijfoodmicro.2012.05.011)
- 6. Ruiz-Barba JL, Jiménez-Díaz R. 2012. A novel Lactobacillus pentosuspaired starter culture for Spanish-style green olive fermentation. Food Microbiol 30:253–259. [https://doi.org/10.1016/j.fm.2011.11.004.](https://doi.org/10.1016/j.fm.2011.11.004)
- 7. Garrido Fernández A, García P, Brenes Balbuena M. 1995. Olive fermentations, p. 593-627. In Rehm HJ, Reed G (ed), Biotechnology: enzymes, biomass, food and feed. VCH, New York, NY.
- 8. Martín-Platero AM, Valdivia E, Maqueda M, Martínez-Bueno M. 2007. Fast, convenient, and economical method for isolating genomic DNA from lactic acid bacteria using a modification of the protein "salting-out" procedure. Anal Biochem 366:102–104. [https://doi.org/10.1016/j.ab.2007](https://doi.org/10.1016/j.ab.2007.03.010) [.03.010.](https://doi.org/10.1016/j.ab.2007.03.010)
- 9. Zerbino DR, Birney E. 2008. Velvet: algorithms for de novo short read assembly using de Bruijn graphs. Genome Res 18:821-829. [https://doi](https://doi.org/10.1101/gr.074492.107) [.org/10.1101/gr.074492.107.](https://doi.org/10.1101/gr.074492.107)
- 10. Hyatt D, Chen GL, LoCascio PF, Land ML, Larimer FW, Hauser LJ. 2010. Prodigal: prokaryotic gene recognition and translation initiation site identification. BMC Bioinformatics 11:119. [https://doi.org/10.1186/1471](https://doi.org/10.1186/1471-2105-11-119) [-2105-11-119.](https://doi.org/10.1186/1471-2105-11-119)
- 11. Casimiro-Soriguer CS, Muñoz-Mérida A, Pérez-Pulido AJ. 2017. Sma3s: a universal tool for easy functional annotation of proteomes and transcriptomes. Proteomics 17. [https://doi.org/10.1002/pmic.201700071.](https://doi.org/10.1002/pmic.201700071)
- 12. Nawrocki EP, Eddy SR. 2013. Infernal 1.1: 100-fold faster RNA homology searches. Bioinformatics 29:2933–2935. [https://doi.org/10.1093/](https://doi.org/10.1093/bioinformatics/btt509) [bioinformatics/btt509.](https://doi.org/10.1093/bioinformatics/btt509)
- 13. Kalvari I, Argasinska J, Quinones-Olvera N, Nawrocki EP, Rivas E, Eddy SR, Bateman A, Finn RD, Petrov AI. 2018. Rfam 13.0: shifting to a genomecentric resource for non-coding RNA families. Nucleic Acids Res 46: D335–D342. [https://doi.org/10.1093/nar/gkx1038.](https://doi.org/10.1093/nar/gkx1038)