



Draft Genome Sequence of CO33, a Coffee-Infecting Isolate of Xylella fastidiosa

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The draft genome sequence of *Xylella fastidiosa* CO33 isolate, retrieved from symptomatic leaves of coffee plant intercepted in northern Italy, is reported. The CO33 genome size is 2,681,926 bp with a GC content of 51.7%.

Received 24 October 2015 Accepted 30 October 2015 Published 17 December 2015

Citation Giampetruzzi A, Loconsole G, Boscia D, Calzolari A, Chiumenti M, Martelli GP, Saldarelli P, Almeida RPP, Saponari M. 2015. Draft genome sequence of CO33, a coffeeinfecting isolate of *Xylella fastidiosa*. Genome Announc 3(6):e01472-15. doi:10.1128/genomeA.01472-15.

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ylella fastidiosa is a bacterium that colonizes the xylem vessels of host plants and the mouthparts of its insect vectors. This bacterium causes plant diseases of economic importance in a wide range of host plants (1). Seven complete and 12 draft X. fastidiosa genomes sequences are available (http://www.ncbi.nlm.nih.gov /genome/genomes/173?). In 2013, X. fastidiosa was detected in southern Italy, representing the first case of this quarantine bacterium becoming established in Europe (2). In July 2015, X. fastidiosa was also detected in Corsica, France. Due to its threat to European agriculture and the environment, the European Union (EU) strengthened customs control measures to limit importation of infected plant material. As a result several X. fastidiosa-infected coffee plants (Coffea arabica) originating from Central America were intercepted, mainly in the Netherlands (3). X. fastidiosa strains belonging to X. fastidiosa subsp. pauca and subsp. fastidiosa have been reported to infect this host (4-6). Therefore, importation of coffee plants from the Americas represents a potential reservoir of genetic and biological X. fastidiosa diversity.

Preliminary studies on the X. fastidiosa isolates recovered from these interceptions demonstrated that they belong to genetically distinct clades (7, 8). Isolate CO33, cultured from a coffee plant intercepted in northern Italy, represents a novel multilocus sequence typing profile, ST72 (G. Loconsole, personal communication). Isolates genetically related to CO33 were found in several coffee plants imported in October 2014 from Costa Rica through the Netherlands (European Food Safety Authority [EFSA] 2015). CO33 was cultured on BCYE medium and genomic DNA extracted using a commercial kit. A DNA library, paired-end sequenced with Illumina, resulted in 10,125,956 reads, representing 310-fold coverage of the expected X. fastidiosa genome. Reads were assembled de novo by EDENA, Velvet, and SOAPdenovo (9-11) with different k-mers. The best contig assemblies from each program were merged using CISA (12) and scaffolded with SSPACE (13) on the Orione instance of Galaxy (14). A final assembly of 96 scaffolds with sizes ranging from 204 to 406,234 bp and an average scaffold size of 27,936 bp was obtained. Scaffolds

less than 5 kb in size were also kept if found to include data homologous to *X. fastidiosa* species by BLASTN analysis.

The draft genome of *X. fastidiosa* isolate CO33 consisted of 2,681,926 bp with a GC content of 51.7%, in agreement with other sequenced isolates that have genome sizes ranging from 2.39 to 2.73 Mbp (15). The genome sequence was annotated through the NCBI Prokaryotic Genome Automatic Annotation Pipeline (PGAAP), which identified 11 rRNA genes, 49 tRNA loci, 2,436 genes, 2,301 protein-encoding genes, 1 noncoding RNA (ncRNA), and 79 repeat regions. Alignments of reads and BLASTN searching of contigs versus available genomes of *X. fastidiosa* revealed that CO33 is genetically related to isolates belonging to different subspecies of *X. fastidiosa*. Specifically, the highest number of CO33 reads mapped either with isolates of subsp. *sandyi* (isolate Ann-1). or of subsp. *morus* (isolate MUL0034), corroborating the genetic complexity of this plant pathogen bacterium and the role of homologous recombination on *X. fastidiosa* diversity.

Nucleotide sequence accession numbers. This whole-genome shotgun project has been deposited at DDBJ/EMBL/GenBank under the accession number LJZW00000000. The version described in this paper is version LJZW00000000.1.

ACKNOWLEDGMENTS

This work was supported by the Regione Puglia and conducted using equipment of "Rete di Laboratori Pubblici SELGE-Regione Puglia (cod. 14)."

REFERENCES

- 1. Hopkins DL, Purcell AH. 2002. *Xylella fastidiosa*: cause of Pierce's disease of grapevine and other emergent diseases. Plant Dis 86:1056–1066. http://dx.doi.org/10.1094/PDIS.2002.86.10.1056.
- Martelli GP, Boscia D, Porcelli F, Saponari M. 2015. The olive quick decline syndrome in south-east Italy: a threatening phytosanitary emergency. Eur J Plant Pathol 2015:1–9. http://dx.doi.org/10.1007/s10658-015 -0784-7.
- 3. EFSA. 2015. Categorisation of plants for planting, excluding seeds, according to the risk of introduction of *Xylella fastidiosa*. EFSA Journal 13:4061. http://dx.doi.org/10.2903/j.efsa.2015.4061.
- 4. Paradela-Filho O, Sugimori MH, Ribeiro IJA, Garcia A, Jr, Beretta

MJG, Harakawa R, Machado MA, Laranjeira FF, Rodrigues Neto J, Beriam LOS. 1997. Occurrence of *Xylella fastidiosa* in coffee plants in Brazil. Summa Phytopathol 23:46–49.

- Rocha JG, Zambolim L, Zambolim EM, Ribeiro do Vale FX, de Jesus WC, Jr, Filho AB. 2010. Quantification of yield loss due to coffee leaf scorch. Crop Protect 29:1100-1104. http://dx.doi.org/10.1016/ j.cropro.2010.04.011.
- Montero-Astúa M, Chacón-Díaz C, Aguilar E, Rodríguez CM, Garita L, Villalobos W, Moreira L, Hartung JS, Rivera C. 2008. Isolation and molecular characterization of *Xylella fastidiosa* from coffee plants in Costa Rica. J Microbiol 46:482–490. http://dx.doi.org/10.1007/s12275-008 -0072-8.
- Bergsma-Vlami M, van de Bilt JLJ, Tjou-Tam-Sin NNA, van de Vossenberg BTLH, Westenberg M. 2015. *Xylella fastidiosa* in coffea arabica ornamental plants imported from Costa Rica and Honduras in the Netherlands. J Plant Pathol 97:391–403. http://dx.doi.org/10.4454/ JPP.V9712.032.
- Legendre B, Mississipi S, Oliver V, Morel E, Crouzillat D, Durand K, Portier P, Poliakoff F, Jacques MA. 2014. Identification and characterisation of *Xylella fastidiosa* isolated from coffee plants in France. J Plant Pathol 96:S4.100. http://dx.doi.org/10.4454/jpp.v96i2SUP.3304.
- Hernandez D, Francois P, Farinelli L, Osteras M, Schrenzel J. 2008. De novo bacterial genome sequencing: millions of very short reads assembled on a desktop computer. Genome Res 18:802–809. http://dx.doi.org/ 10.1101/gr.072033.107.

- Zerbino DR, Birney E. 2008. Velvet: algorithms for de novo short read assembly using de Bruijn graphs. Genome Res 18:821-829. http:// dx.doi.org/10.1101/gr.074492.107.
- 11. Luo R, Liu B, Xie Y, Li Z, Huang W, Yuan J, He G, Chen Y, Pan Q, Liu Y, Tang J, Wu G, Zhang H, Shi Y, Liu Y, Yu C, Wang B, Lu Y, Han C, Cheung DW, Yiu SM, Peng S, Xiaoqian Z, Liu G, Liao X, Li Y, Yang H, Wang J, Lam T, Wang J. 2012. SOAPdenovo2: an empirically improved memory-efficient short-read *de novo* assembler. GigaScience 1:18. http://dx.doi.org/10.1186/2047-217X-1-18.
- Lin S, Liao Y. 2013. CISA: contig integrator for sequence assembly of bacterial genomes. PLoS One 8:e60843. http://dx.doi.org/10.1371/ journal.pone.0060843.
- 13. Boetzer M, Henkel CV, Jansen HJ, Butler D, Pirovano W. 2011. Scaffolding preassembled contigs using SSPACE. BioInformatics 27:578–579. http://dx.doi.org/10.1093/bioinformatics/btq683.
- Cuccuru G, Orsini M, Pinna A, Sbardellati A, Soranzo N, Travaglione A, Uva P, Zanetti G, Fotia G. 2014. Orione, a web-based framework for NGS analysis in microbiology. BioInformatics 30:1928–1929. http:// dx.doi.org/10.1093/bioinformatics/btu135.
- Varani AM, Monteiro-Vitorello CB, de Almeida LGPd, Souza RC, Cunha OL, Lima WC, Civerolo E, Van Sluys MA, Vasconcelos ATR. 2012. *Xylella fastidiosa* comparative genomic database is an information resource to explore the annotation, genomic features, and biology of different strains. Genet Mol Biol 35:149–152. http://dx.doi.org/10.1590/ S1415-47572012005000019.