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# Whole-Genome Sequence of *Stenotrophomonas maltophilia* D457, a Clinical Isolate and a Model Strain

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Stenotrophomonas maltophilia is an opportunistic pathogen with an environmental origin, and it is an increasingly relevant cause of nosocomial infections. Here we present the whole-genome sequence of *S. maltophilia* strain D457, a clinical isolate that is being used as a model for studying antibiotic resistance in this bacterial species.

tenotrophomonas maltophilia is a free-living bacterial species with biotechnological relevance for different applications, including the production of molecules of economic value, the biodegradation of pollutants, and use of the organism for the biological control of plant infections (12). In addition, S. maltophilia is an increasingly relevant human opportunistic pathogen that is involved in infections at hospitals and in patients with cystic fibrosis (5, 21). One of the most worrisome properties of S. maltophilia consists of its low susceptibility to most antibiotics currently in use for the treatment of infections (13). This lack of susceptibility is due at least in part to the presence in its genome of genes encoding antibiotic-inactivating enzymes, efflux pumps (1, 2, 6), and other proteins that contribute to the intrinsic resistance, such as SmQnr (15, 18). The strain D457 (3) is a clinical isolate that has been used for studying the presence of elements contributing to resistance in S. maltophilia and the regulation of these elements (2, 8, 9, 14–17).

In this article, we present the full sequence and the annotation of the genome of S. maltophilia D457. The genome was sequenced using the 454 GS FLX system with single ends and 3-kb paired ends (64 and 147 Mb, with 370,000 and 700,000 reads before assembly, respectively). All reads were assembled de novo by using the MIRA software (http://chevreux.org/projects\_mira.html). Assembly was revised using the Gap4 software from the Staden package (20). The genome of S. maltophilia K279a (6) and information for paired ends were used as references to design PCR amplifications that, after Sanger sequencing, served to close the genome gaps and to solve the repeat regions. Coding sequences were initially annotated in the RAST server (4) followed by a manual curation. Pseudogenes were confirmed by assembly inspection. Hypothetical coding genes smaller than 250 nucleotides without similarity with other S. maltophilia strains were removed. Noncoding RNA genes were annotated by using several methods (7, 10, 19). Repeat sequences were analyzed to identify transposable elements and for mapping the inverted repeats at the end of the IS

The chromosome comprises 4,769,156 bp, with a G+C content of 66.8%. It contains 4,209 genes, of which 4,101 are coding genes and 108 are noncoding RNA genes (13 rRNA, 71 tRNA, and 24 other RNAs). In addition, 30 pseudogenes (29 protein-coding genes and 1 tRNA) were identified. Six types of transposable elements were identified that comprised 19 complete and 3 defective forms. Only two of them were detected in other sequenced *S*.

maltophilia strains (ISSmaD4 and Tn5044). Among the predicted coding sequences of D457, we found that more than 200 genes were not shared with the other strains of *S. maltophilia* with completely sequenced genomes (6, 11, 22). Notably, most of them encoded hypothetical proteins and transposases, which indicates that the core genome of *S. maltophilia* is large. No chromosomal rearrangements were detected when we compared the sequence to those of other published strains, except for the insertion of phages or other horizontally transferred genes.

**Nucleotide sequence accession number.** The results of this whole-genome shotgun project have been deposited at the European Nucleotide Archive (ENA) under accession number HE798556.

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#### REFERENCES

- Alonso A, Martinez JL. 2000. Cloning and characterization of SmeDEF, a novel multidrug efflux pump from *Stenotrophomonas maltophilia*. Antimicrob. Agents Chemother. 44:3079–3086.
- 2. Alonso A, Martinez JL. 2001. Expression of multidrug efflux pump SmeDEF by clinical isolates of *Stenotrophomonas maltophilia*. Antimicrob. Agents Chemother. 45:1879–1881.
- 3. Alonso A, Martinez JL. 1997. Multiple antibiotic resistance in *Stenotrophomonas maltophilia*. Antimicrob. Agents Chemother. 41:1140–1142.
- Aziz RK, et al. 2008. The RAST server: rapid annotations using subsystems technology. BMC Genomics 9:75. doi:10.1186/1471-2164-9-75.

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- Brooke JS. 2012. Stenotrophomonas maltophilia: an emerging global opportunistic pathogen. Clin. Microbiol. Rev. 25:2–41.
- Crossman LC, et al. 2008. The complete genome, comparative and functional analysis of *Stenotrophomonas maltophilia* reveals an organism heavily shielded by drug resistance determinants. Genome Biol. 9:R74. doi: 10.1186/gb-2008-9-4-r74.
- Griffiths-Jones S, et al. 2005. Rfam: annotating non-coding RNAs in complete genomes. Nucleic Acids Res. 33:D121–D124.
- Hernandez A, et al. 2009. Structural and functional analysis of SmeT, the repressor of the *Stenotrophomonas maltophilia* multidrug efflux pump SmeDEF. J. Biol. Chem. 284:14428–14438.
- Hernandez A, Ruiz FM, Romero A, Martinez JL. 2011. The binding of triclosan to SmeT, the repressor of the multidrug efflux pump SmeDEF, induces antibiotic resistance in *Stenotrophomonas maltophilia*. PLoS Pathog. 7:e1002103. doi:10.1371/journal.ppat.1002103.
- 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.
- Rocco F, De Gregorio E, Colonna B, Di Nocera PP. 2009. Stenotrophomonas maltophilia genomes: a start-up comparison. Int. J. Med. Microbiol. 299:535–546.
- 12. Ryan RP, et al. 2009. The versatility and adaptation of bacteria from the genus *Stenotrophomonas*. Nat. Rev. Microbiol. 7:514–525.
- Sanchez MB, Hernandez A, Martinez JL. 2009. Stenotrophomonas maltophilia drug resistance. Future Microbiol. 4:655–660.
- 14. Sanchez MB, Hernandez A, Rodriguez-Martinez JM, Martinez-Martinez L, Martinez JL. 2008. Predictive analysis of transmissible quino-

- lone resistance indicates *Stenotrophomonas maltophilia* as a potential source of a novel family of Qnr determinants. BMC Microbiol. **8**:148. doi:10.1186/1471-2180-8-148.
- 15. Sanchez MB, Martinez JL. 2010. SmQnr contributes to intrinsic resistance to quinolones in *Stenotrophomonas maltophilia*. Antimicrob. Agents Chemother. 54:580–581.
- Sanchez P, Alonso A, Martinez JL. 2002. Cloning and characterization of SmeT, a repressor of the *Stenotrophomonas maltophilia* multidrug efflux pump SmeDEF. Antimicrob. Agents Chemother. 46:3386–3393.
- Sanchez P, Alonso A, Martinez JL. 2004. Regulatory regions of smeDEF in *Stenotrophomonas maltophilia* strains expressing different amounts of the multidrug efflux pump SmeDEF. Antimicrob. Agents Chemother. 48: 2274–2276.
- 18. Shimizu K, et al. 2008. Smqnr, a new chromosome-carried quinolone resistance gene in Stenotrophomonas maltophilia. Antimicrob. Agents Chemother. 52:3823–3825.
- Silva FJ, Belda E, Talens SE. 2006. Differential annotation of tRNA genes with anticodon CAT in bacterial genomes. Nucleic Acids Res. 34:6015– 6022.
- Staden R, Beal KF, Bonfield JK. 2000. The Staden package, 1998. Methods Mol. Biol. 132:115–130.
- Turrientes MC, et al. 2010. Polymorphic mutation frequencies of clinical and environmental *Stenotrophomonas maltophilia* populations. Appl. Environ. Microbiol. 76:1746–1758.
- 22. Zhu B, et al. 2012. Genome sequence of *Stenotrophomonas maltophilia* RR-10, isolated as an endophyte from rice root. J. Bacteriol. 194:1280–1281.

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