

Draft Genome Sequence of *Enterobacter ludwigii* NCR3, a Heavy Metal-Resistant Rhizobacterium

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We report here the draft genome of *Enterobacter ludwigii* NCR3, a Gram-negative bacterium isolated from the *Carpobrotus rossii* (Haw.) Schwantes rhizosphere. The analysis of the ~4.8-Mb draft genome shows that this strain harbors several genes associated with heavy metal resistance and plant growth-promoting activity, suggesting its potential application in microbe-assisted phytoremediation.

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A promising approach to optimize the synergistic effect of plants and microorganisms during the phytoremediation of heavy metal (HM)-contaminated sites consists of coupling phytoextraction with soil bioaugmentation (1). The addition of microorganisms can change the bioavailability of metals and simultaneously promote plant growth results in an enhanced metal uptake by plants (2). Therefore, being able to identify microorganisms with the potential to assist the plant-mediated HM environmental remediation is fundamental to improving the process efficiency.

Due to their ability to increase HM mobilization in soils, stimulate plant growth, and influence HM accumulation in plant tissues, species belonging to the family *Enterobacteriaceae* have been previously proposed as suitable candidates to support bioaugmentation-assisted HM remediation (3). Here, we report the draft genome of *Enterobacter ludwigii* NCR3, a plant growth-promoting bacterium isolated from the rhizosphere of the heavy metal hyperaccumulator *Carpobrotus rossii* (Haw.) Schwantes and grown in a cadmium-contaminated environment (4). When tested for HM resistance, NCR3 showed a MIC of 50, 150, and 800 mg·L⁻¹ for cadmium, copper and zinc, respectively (4), suggesting tolerance for a wide range of heavy metals.

Genomic DNA was isolated using a DNeasy blood and tissue kit (Qiagen) and converted to sequencing libraries using the Nextera XT DNA library preparation kit (Illumina). Libraries were normalized and pooled before sequencing on an Illumina MiSeq with 2 × 300 paired-end reads. For each isolate, the A5-miseq pipeline (5) was used to perform read trimming and correction, contig assembly, crude scaffolding, misassembly correction, and final scaffolding. No putative misassemblies were detected with this method. Genome sequencing resulted in high-coverage assemblies of the 4,779,466-bp genome (115-fold median coverage; 54.7% G+C content), which should represent most of the functional annotated genes and allow for comparative studies. The final number of contigs was 23 with an *N*₅₀ value of 771,694 bp.

The RAST annotation system (6) showed that the *E. ludwigii* NCR3 genome contains 4,498 coding sequences. Consistent with Liu et al. (4), the genome harbors several predicted genes linked to HM resistance. These include the *ars* (*arsRDABC*) operon, as well as genes encoding for the proteins CopC and CopD associated with the copper resistance operon *copABCD*, and the cobalt-zinc-cadmium resistance protein CzcA. Several antibiotic resistance operons were also found (e.g., *mdtABCD*). Interestingly, we identified genes related to organic acids biosynthesis, suggesting the ability of *E. ludwigii* NCR3 to change the metal's bioavailability and contribute to plant-mediated metal uptake (7). The presence of genes encoding for siderophores (enterobactin and aerobactin) and auxin biosynthesis also confirms that NCR3 possesses characteristics relevant for a beneficial plant-microbe interaction.

Accession number(s). This whole-genome shotgun project has been deposited at DDBJ/ENA/GenBank under the accession number [MCGF000000000](https://www.ncbi.nlm.nih.gov/nuclink/MCGF000000000). The version described in this paper is the first version, MCGF01000000.

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REFERENCES

1. Koul S, Gauba P. 2014. Bioaugmentation-a strategy for cleaning up soil. *J Civil Eng Environ Technol* 1:68–70.
2. Rajkumar M, Sandhya S, Prasad MN, Freitas H. 2012. Perspectives of plant-associated microbes in heavy metal phytoremediation. *Biotechnol Adv* 30:1562–1574. <http://dx.doi.org/10.1016/j.biotechadv.2012.04.011>.
3. Płociniczak T, Sinkkonen A, Romantschuk M, Piotrowska-Seget Z. 2013. Characterization of *Enterobacter intermedius* MH8b and its use for the enhancement of heavy metals uptake by *Sinapis alba* L. *Appl Soil Ecol* 63:1–7. <http://dx.doi.org/10.1016/j.apsoil.2012.09.009>.
4. Liu W, Wang Q, Wang B, Hou J, Luo Y, Tang C, Franks AE. 2015. Plant growth-promoting rhizobacteria enhance the growth and Cd uptake of *Sedum plumbizincicola* in a Cd-contaminated soil. *J Soils Sediments* 15: 1191–1199. <http://dx.doi.org/10.1007/s11368-015-1067-9>.

5. Coil D, Jospin G, Darling AE. 2015. A5-miseq: an updated pipeline to assemble microbial genomes from Illumina MiSeq data. *Bioinformatics* 31:587–589. <http://dx.doi.org/10.1093/bioinformatics/btu661>.
6. 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. 2008. The RAST server: Rapid Annotations using Subsystems Technology. *BMC Genomics* 9:75. <http://dx.doi.org/10.1186/1471-2164-9-75>.
7. Iqbal J, Ahemad M. 2015. Recent advances in bacteria-assisted phytoremediation of heavy metals from contaminated soil, p. 401–423. *In* Chandra R (ed), *Advances in biodegradation and bioremediation of industrial waste*, 1st ed. CRC Press, Boca Raton, FL.