



## Sphingomonas taxi, Isolated from Cucurbita pepo, Proves to Be a DDE-Degrading and Plant Growth-Promoting Strain

## Nele Eevers,<sup>a</sup> Jonathan D. Van Hamme,<sup>b</sup> Eric M. Bottos,<sup>b</sup> Nele Weyens,<sup>a</sup> Jaco Vangronsveld<sup>a</sup>

Centre for Environmental Sciences, Hasselt University, Diepenbeek, Belgium<sup>a</sup>; Department of Biological Sciences, Thompson Rivers University, Kamloops, British Columbia, Canada<sup>b</sup>

The draft genome of *Sphingomonas taxi*, a strain of the *Sphingomonadaceae* isolated from *Cucurbita pepo* root tissue, is presented. This Gram-negative bacterium shows 2,2-bis(*p*-chlorophenyl)-1,1-dichloroethylene (DDE)-degrading potential and plant growth-promoting capacities. An analysis of its 3.9-Mb draft genome will enhance the understanding of DDE-degradation pathways and phytoremediation applications for DDE-contaminated soils.

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Address correspondence to Jaco Vangronsveld, jaco.vangronsveld@uhasselt.be.

DT (2,2-bis[*p*-chlorophenyl]-1,1,1-trichloroethane) (1) is an agricultural and gardening pesticide that has been used since 1943 (2). When exposed to soil conditions, DDT degrades to 2,2-bis(*p*-chlorophenyl)-1,1-dichloroethylene (DDE). These products are categorized as persistent organic pollutants (POPs) (3) and threaten human and wildlife health because of their bioaccumulative and hormone-disrupting properties (4). DDE phytoremediation using *Cucurbita pepo*, a DDE accumulator (5), in combination with suitable endophytes, may resolve problems associated with DDE-contaminated soils.

The *Sphingomonas taxi* strain described here was isolated from *Cucurbita pepo* root tissue that was exposed to 100 mg·liter<sup>-1</sup> of DDE during cultivation. Partial 16S rRNA gene sequencing and phenotypic profiling identified the strain as *Sphingomonas taxi*, with the closest related 16S rRNA sequence (87%) being from strain ATCC 55669 (GenBank accession no. CP009571.1).

To better characterize the isolate, DNA was extracted, and the whole-genome shotgun sequence was prepared on an IonTorrent PGM, as described by Evers et al. (6).

In all, 1.29 million reads with a mean length of 270 bases generated 349 Mb of data in Torrent Suite version 4.2.1. Assembly using SPAdes version 3.1.0 (7, 8) (uniform coverage mode; *k*-mers 21, 33, 55, 77, 99) yielded 109 contigs >1,000 bp, giving a consensus length of 3,941,497 bp at a 51× coverage (largest contig, 231,989 bp;  $N_{50}$ , 75,931 bp). The contigs were ordered with Mauve (9), using the *Sphingomonas* taxi ATCC 55669 genome (accession no. CP00951.1) as a reference. The PGAP (NCBI) pipeline was used for annotation (10). The *S. taxi* genome consists of a single circular chromosome (67.15% GC content), which includes 534 pseudogenes, 3 rRNAs (5S, 16S, 23S), 48 tRNAs, and 1 noncoding RNA (ncRNA), and 3,634 coding genes that were arranged into 378 pathways using Pathway Tools (11, 12).

In experiments testing DDE-degrading capacities, *Sphingomonas taxi* showed increased growth when exposed to 50 mg·liter<sup>-1</sup> DDE in comparison to control conditions. This result is in agreement with the presence of halogenases, dioxygenases, and hydro-

lases (13-16). The presence of pathways related to the degradation of phenylacetate, octane, acrylonitrile, toluene, phenylmercury acetate, and naphthalene, as well as for the detoxification of arsenate and superoxide radicals, make this Sphingomonas taxi strain a suitable candidate for the phytoremediation of soils polluted with mixed contaminants. Genes coding for plant growth-promoting capacities are also present, confirming results from phenotypic tests: 1-aminocyclopropane-1-carboxylate deaminase activity, siderophore production, auxin biosynthesis, and phosphorous solubilization. Interestingly, pathways for nitrogen and carbon dioxide fixation are present, although we have not confirmed these traits. The strain also shows a capacity for hemicellulose and cellulose degradation, a useful trait for facilitating entrance into plant roots during inoculation. This combination of characteristics makes Sphingomonas taxi a promising strain for the phytoremediation of soils contaminated with DDE and other contaminants.

**Nucleotide sequence accession numbers.** This whole-genome shotgun project has been deposited at DDBJ/EMBL/GenBank under the accession number JXTP00000000. The version described in this paper is version JTXP01000000.

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## REFERENCES

- Thomas JE, Ou LT, All-Agely A. 2008. DDE remediation and degradation. Rev Environ Contam Toxicol 194:55–70. http://dx.doi.org/10.1007/ 978-0-387-74816-0\_3.
- Turusov V, Rakitsky V, Tomatis L. 2002. Dichlorodiphenyltrichloroethane (DDT): ubiquity, persistence, and risks. Environ Health Perspect 110: 125–128.
- White JC. 2010. Inheritance of p,p'-DDE phytoextraction ability in hybridized *Cucurbita pepo* cultivars. Environ Sci Technol 44:5165–5169. http://dx.doi.org/10.1021/es100706t.

- 4. Colles A, Koppen G. 2007. DDE serumwaarden in landelijke regio's en de Albertkanaalzone boven het Vlaamse referentiegemiddelde: enquete naar de oorzaak. De Biomonitor, digitale nieuwsbrief van het medisch milieukundig netwerk (VITO). http://www.milieu-en-gezondheid.be/nieuwsbrief/biomonitor%2017/DDE.html.
- 5. White JC, Parrish ZD, Isleyen M, Gent MPN, Iannucci-Berger W, Eitzer BD, Mattina MJI. 2005. Uptake of weathered *p*,*p*'-DDE by plant species effective at accumulating soil elements. Microchem J 81:148–155. http://dx.doi.org/10.1016/j.microc.2005.01.010.
- Eevers N, Van Hamme JD, Bottos EM, Weyens N, Vangronsveld J. 2015. Draft genome sequence of *Enterobacter aerogenes*, a DDE-degrading and plant growth-promoting strain isolated from *Cucurbita pepo*. Genome Announc 3(2):e00317-15. http://dx.doi.org/10.1128/ genomeA.00317-15.
- Bankevich A, Nurk S, Antipov D, Gurevich AA, Dvorkin M, Kulikov AS, Lesin VM, Nikolenko SI, Pham S, Prjibelski AD, Pyshkin AV, Sirotkin AV, Vyahhi N, Tesler G, Alekseyev MA, Pevzner PA. 2012. SPAdes: a new genome assembly algorithm and its applications to singlecell sequencing. J Comput Biol 19:455–477. http://dx.doi.org/10.1089/ cmb.2012.0021.
- 8. Gurevich A, Saveliev V, Vyahhi N, Tesler G. 2013. QUAST: quality assessment tool for genome assemblies. Bioinformatics 29:1072–1075. http://dx.doi.org/10.1093/bioinformatics/btt086.
- Rissman AI, Mau B, Biehl BS, Darling AE, Glasner JD, Perna NT. 2009. Reordering contigs of draft genomes using the Mauve Aligner. Bioinformatics 25:2071–2073. http://dx.doi.org/10.1093/bioinformatics/btp356.
- 10. Angiuoli SV, Gussman A, Klimke W, Cochrane G, Field D, Garrity G,

Kodira CD, Kyrpides N, Madupu R, Markowitz V, Tatusova T, Thomson N, White O. 2008. Toward an online repository of standard operating procedures (SOPs) for (meta)genomic annotation. OMICS 12:137–141. http://dx.doi.org/10.1089/omi.2008.0017.

- 11. Caspi R, Altman T, Billington R, Dreher K, Foerster H, Fulcher CA, Holland TA, Keseler IM, Kothari A, Kubo A, Krummenacker M, Latendresse M, Mueller LA, Ong Q, Paley S, Subhraveti P, Weaver DS, Weerasinghe D, Zhang P, Karp PD. 2014. The MetaCyc database of metabolic pathways and enzymes and the BioCyc collection of pathway/ genome databases. Nucleic Acids Res 42:D459–D471. http://dx.doi.org/ 10.1093/nar/gkt1103.
- Karp PD, Paley S, Romero P. 2002. The Pathway Tools software. Bioinformatics 18(Suppl 1):S225–S232. http://dx.doi.org/10.1093/ bioinformatics/18.suppl\_1.S225.
- Quensen JF, Mueller SA, Jain MK, Tiedje JM. 1998. Reductive dechlorination of DDE to DDMU in marine sediment microcosms. Science 280: 722–724. http://dx.doi.org/10.1126/science.280.5364.722.
- Hay AG, Focht DD. 1998. Cometabolism of 1,1-dichloro-2,2-bis(4chlorophenyl)ethylene by *Pseudomonas acidovorans* M3GY grown on biphenyl. Appl Environ Microbiol 64:2141–2146.
- Nadeau LJ, Menn FM, Breen A, Sayler GS. 1994. Aerobic degradation of 1,1,1-trichloro-2,2-bis(4-chlorophenyl)ethane (DDT) by Alcaligenes eutrophus A5. Appl Environ Microbiol 60:51–55.
- Hay AG, Focht DD. 2000. Transformation of 1,1-dichloro-2,2-(4-chlorophenyl)ethane (DDD) by *Ralstonia eutropha* strain A5. FEMS Microbiol Ecol 31:249-253. http://dx.doi.org/10.1111/j.1574 -6941.2000.tb00690.x.