

Draft Genome Sequences of 10 *Microbacterium* spp., with Emphasis on Heavy Metal-Contaminated Environments

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***Microbacterium* spp. isolated from heavy metal (HM)-contaminated environments (soil and plants) can play a role in mobilization processes and in the phytoextraction of HM. Here, we report the whole-genome sequences and annotation of 10 *Microbacterium* spp. isolated from both HM-contaminated and -noncontaminated compartments.**

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Microbacterium spp. have been isolated from diverse environments, including soil, plants, water, and humans (1–4). They belong to the class *Actinobacteria*, known for the production of a rich spectrum of secondary metabolites, including organic acids, antibiotics, siderophores, and other chelators (5). Interestingly, several *Microbacterium* spp. have shown to be resistant to different HM and to influence the mobility of HM in contaminated soils (6–8). The influence on HM mobilization might be due to the secretion of specific metabolites, making these bacteria suitable candidates for not only the understanding of metal mobilization processes but also the improvement of phytoextraction techniques limited mainly by the plant biomass production and the rate of metal uptake (9, 10). Bacteria isolated from HM-contaminated environments can overcome these limitations by promoting plant growth and increasing HM availability. Therefore, we decided to sequence a set of *Microbacterium* sp. genomes of organisms isolated from different HM-contaminated sites in Europe and strains obtained from the DSMZ collection (<http://www.dsmz.de/catalogues/catalogue-microorganisms.html>). Based on related publications (11–15), we conclude that the DSMZ strains do not derive from HM-polluted environments (Table 1).

Genomic DNA was extracted with a phenol-chloroform extraction protocol (16). The Nextera XT kit (Illumina, San Diego, CA) was used for library preparation, and whole-genome sequencing was performed using Illumina MiSeq (MiSeq reagent kit version 3). Raw reads were screened for PhiX contamination using Bowtie2 (17). Adapter and quality trimming were performed in Trimmomatic-0.32 (18). Overlapping reads were subsequently merged using FLASH (19), and long single reads and paired-end reads were assembled with SPAdes 3.1.0 (20). The assembly quality was estimated in QUAST 2.3 (21), and quality control of the mapping data was performed in Qualimap 1.0 (22). PhyloSift version 1.0.1 (23) was used to verify the genome completeness, assessing a list of 40 highly conserved single-copy marker genes, all of which were identified in each assembly. The phylogenetic analysis of the genomes showed no contamination, but a substantial

number of viral sequences were detected (10 to 40% of the genome content), which was also confirmed by PHAST (24) for most genomes (Table 1). Moreover, we detected viral sequences in already-published *Microbacterium* sp. genomes in similar percentages.

Genome annotation (summarized in Table 1) was performed in Prokka (25), incorporating Prodigal 2.60, Aragorn, and Barrnap for the prediction of open reading frames (ORFs), tRNAs, and rRNAs, respectively. rRNA detection was confirmed with RNAmmer 1.2 (26). Using antiSMASH 2.0 (27), we detected gene clusters involved in the production of terpenoids in all sequenced strains, except the colorless isolate ARN176. Clusters involved in the production of polyketides and nonribosomal peptides were found in all strains but in DSM 18659 and in DSM 18659 and BEL163, respectively. A deeper comparison of the *Microbacterium* genes involved in HM resistance and mobilization is currently being performed and will be published in a subsequent report.

Nucleotide sequence accession numbers. The nucleotide sequences have been deposited at the DDBJ/EMBL/GenBank under the accession numbers provided in Table 1.

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TABLE 1 Summary of isolate origins and genomic features

| <i>Microbacterium</i> sp. ^a | Isolate | Source | Viral sequence % estimated with PhyloSift | No. of rRNAs in Barnap/ total no. ^b | No. of tRNAs | G+C content (%) | L_{50}^d | N_{50}^d | No. of contigs | Total bp | Coverage (×) | Accession no. |
|---|-----------|---|---|--|-----------------|--------------------|------------|------------|-------------------|----------|--------------|---------------|
| From HM | | | | | | | | | | | | |
| <i>M. azadirachtae</i> NR_116502 | DSM 23848 | Rhizosphere of neem (<i>Azadirachta indica</i>) | 28 ^c | 3/12 | 49 | 3,751 | 70.45 | 12 | 97,758 | 86 | 4,037,586 | 49.69 ± 16.7 |
| <i>M. foliorum</i> NR_025368 | DSM 12966 | Phyllosphere of grasses seedlings | 10 | 5/11 | 47 | 3,324 | 68.7 | 6 | 151,883 | 46 | 3,558,318 | 74.75 ± 24.7 |
| <i>M. episengisoli</i> NR_041516 | DSM 18659 | Soil of ginseng field | 24 ^e | 3/3 | 45 | 2,963 | 70.22 | 8 | 145,068 | 80 | 3,047,504 | 71.53 ± 29.4 |
| <i>M. ketosireducens</i> NR_024638 | DSM 12510 | Soil | 26 | 3/12 | 49 | 3,481 | 70.27 | 10 | 115,384 | 57 | 3,922,598 | 56.55 ± 21.1 |
| <i>M. trichothecenolyticum</i> NR_044937 | DSM 8608 | Soil | 23 ^e | 3/15 | 48 | 4,074 | 70.16 | 8 | 220,592 | 41 | 4,524,308 | 132.14 ± 42.4 |
| From HM contaminated environment | | | | | | | | | | | | |
| <i>M. oxydans</i> JX185498 | BEL4b | Rhizosphere of <i>Brassica napus</i> (Lommel Field, Belgium) | 25 | 3/9 | 49 | 3,589 | 68.27 | 3 | 408,688 | 26 | 3,807,916 | 189.68 ± 47.9 |
| <i>M. oxydans</i> JX185498 | BEL163 | Roots of <i>Salix viminalis</i> (Lommel Field, Belgium) | 28 ^e | 4/16 | 44 | 3,545 | 68.03 | 6 | 164,861 | 30 | 3,692,080 | 153.17 ± 50.4 |
| <i>M. hydrocarbonoxydans</i> | SA335 | Rhizosphere of <i>Alyssum serpyllifolium</i> subsp. <i>lusatanicum</i> (Portugal, Samlil, Trás-os-Montes) | 17 ^e | 5/11 | 55 | 3,663 | 68.52 | 3 | 545,806 | 10 | 3,933,585 | 165.98 ± 38.9 |
| <i>Microbacterium</i> sp. AB042083 | SA39 | Rhizosphere of <i>Alyssum serpyllifolium</i> subsp. <i>lusatanicum</i> (Portugal, Samlil, Trás-os-Montes) | 40 ^e | 4/13 | 50 | 3,732 | 68.26 | 7 | 232,853 | 39 | 3,862,900 | 94.19 ± 27.4 |
| <i>M. azadirachtae</i> KF150486 | ARN176 | Soil (Arnoldstein, Austria) | 37 | 3/9 | 40 | 3,910 | 70.14 | 7 | 258,062 | 40 | 4,243,255 | 126.11 ± 36.7 |

^aThe number after the organism name refers to the 16S rRNA gene NCBI accession number of the closest described relative.^bNumber of rRNAs predicted by Barnap/number of rRNAs estimated based on the coverage of the contigs containing rRNA genes.^cORFs, open reading frames.^d L_{50} and N_{50} were calculated using QUAST. N_{50} is the contig length such that using longer or equal length contigs produces half (50%) the bases of the assembly. L_{50} is the minimal number of contigs that cover ≥50% of the total length.^ePresence of viral sequence confirmed by PHAST.