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Draft Genome Sequence of the Polycyclic Aromatic Hydrocarbon-Degrading, Genetically Engineered Bioluminescent Bioreporter *Pseudomonas fluorescens* HK44▽

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Pseudomonas fluorescens **strain HK44 (DSM 6700) is a genetically engineered *lux*-based bioluminescent bioreporter. Here we report the draft genome sequence of strain HK44. Annotation of ~6.1 Mb of sequence indicates that 30% of the traits are unique and distributed over five genomic islands, a prophage, and two plasmids.**

Pseudomonas fluorescens HK44 is derived from *P. fluorescens* 18H, an isolate obtained from a manufactured gas plant soil heavily contaminated with polycyclic aromatic hydrocarbons (PAHs) and heavy metals (14). Strain HK44 was engineered to contain a naphthalene catabolic plasmid, pUTK21, mutagenized by transposon insertion of the *luxCDABE* (bioluminescent) gene cassette (7). Due to the naphthalene catabolic pathway carried on pUTK21, strain HK44 degrades a number of two- and three-ring PAHs (8) and produces bioluminescent light in response to naphthalene, salicylate, or 4-methyl salicylate. Strain HK44 has been used to model bioluminescence kinetics (6) and naphthalene pathway substrate range (15) and to examine PAH bioavailability in soils and sediments (3, 9). Additionally, it has been the subject of numerous applied studies, including investigation of the role of plant exudates in inducing phytoremediation (5), measurement of microbial growth and transport in sand and soil (13, 16–17), and environmental risk assessment and gene transfer in the vadose zone of soil lysimeters (12).

Whole-genome sequencing of strain HK44 was performed using a Roche 454 Life Sciences GS FLX system. Sequence data were generated from two prepared libraries, a shotgun and a 3-kb paired-end library, resulting in 250,726,215 and 57,244,093 reads, respectively, providing approximately 50-fold coverage. The shotgun sequences were assembled into 131 contigs using a Newbler version 2.3 assembler (Roche) containing a quality score of ≥40. These contigs were coupled into 21 scaffolds using paired-end information. Of the 21 scaffolds, four comprised sequences belonging to plasmids. One of these was identified as the recombinant pUTK21 and the remaining three as part of one or more cryptic plasmids. To arrange the assembled scaffolds, an optical map of strain HK44 was prepared at OpGen Technologies, Inc. (Madison, WI), using a BglII restriction digest (2, 11). The genome was annotated using the RAST server (1), the automated annotation pipeline of ORNL (4), and JGI's IMG/ER pipeline (http://img.jgi.doe.gov/er/doc/about_index.html).

Analysis of the unclosed draft genome sequence of strain HK44 shows that it has a genome size of 6,078,631 bp, a gene coding density of 84.82%, and a G+C content of 58.73 mol% and contains 5,720 protein-coding sequences, 3 rRNA operons, 56 tRNA genes, and one integrated phage. Among the 5,720 protein-coding genes, putative functions could be assigned to 4,558, with 1,617 being associated with KEGG (Kyoto Encyclopedia of Genes and Genomes) pathways. The HK44 genome exhibits certain characteristics that are typical of *P. fluorescens*, such as xenobiotics degradation, oxidative stress, and plant commensal relationship (5, 10). Surprisingly, 30% of strain HK44's genome is atypical of *P. fluorescens* and is distributed among five genomic islands, numerous smaller clusters, a prophage, the pUTK21 plasmid, and a potential cryptic plasmid. Additionally, strain HK44 encodes pathways conferring novel traits, including a previously undescribed aromatic carbon degradation pathway and polysaccharide production. Furthermore, several virulence factors not characteristic of other *P. fluorescens* strains were identified. Thus, the genome sequence of strain HK44 will provide further information on its environmental interaction and metabolic capabilities.

Nucleotide sequence accession numbers. The draft genome sequence of strain HK44 has been deposited in NCBI GenBank under accession number AFOY00000000, in the IMG database under identifier (ID) 2504643024, and in GOLD under ID Gi09651.

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