

TITLE: Population structure analysis of *Pseudomonas putida* provides new insights into biotechnological applications and pathogenicity

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ABSTRACT: *Pseudomonas putida* is a Gram-negative rod-shaped bacterium found in diverse environments. The biotechnological application of *P. putida* to promote plant growth and bioremediate toxic compounds has been widely explored. However, *P. putida* have been associated with human infections, which raises the importance of a better understanding of this species. Here, we characterize, for the first time, the population structure of *P. putida*, aiming to understand genomic diversity and niche adaptation of the species. We downloaded all 8,763 *Pseudomonas* genomes from NCBI and used *P. putida* KT2440 as a *type* strain gather genomes from this species by using a 95% average nucleotide identity threshold. We performed a Multilocus Sequence Typing (MLST) analysis with eight housekeeping genes and extracted SNPs present in genes common to all isolates (i.e. the core genome) to assess genetic relatedness. We used the software STRUCTURE to evaluate the clonal distribution and RAxML to reconstruct the species phylogeny using core genome SNPs. Our results indicate that more than 50% of *P. putida* genomes deposited in NCBI are misclassified, especially those related with hospital outbreaks. We show that the population structure of *P. putida* is composed by at least seven Clonal Complexes (CC) and clinical isolates were found in CC1 and CC5. All isolates harbor phosphate solubilization genes (i.e. *pqq*). CC2 comprises rhizosphere isolates, including the plant-growth promoting *P. putida* BIRD-1. Regarding the resistance profiles, *P. putida* demonstrated a low frequency of resistance genes. Virulence profiles clearly separate the population based on presence and absence of genes involved in siderophore production. Virulence genes commonly found in *P. putida*, such as exotoxin A, phospholipase C, and elastase were absent in *P. putida*. Our results provide a foundation to understand *P. putida* genetic diversity, with important implications to its biosafety in agricultural and environmental applications.

Keywords: MLST, Clonal Complexes, PGPR, resistome

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