

Pseudomonas rhizoryzae sp. nov., isolated from rice

Xing Wang¹, Shan-wen He^{1,2}, He-Bao Guo¹, Kyu Kyu Thin¹, Ju-sheng Gao^{1,3}, Yao Wang^{1,2} and Xiao-Xia Zhang^{1,*}

Abstract

Two yellow-pigmented, Gram-stain-negative and rod-shaped bacterial strains, designated as RY24^T and ZYY160, were isolated from rice. Results of phylogenetic analysis based on 16S rRNA gene sequences showed that strains RY24^T and ZYY160 belonged to the genus *Pseudomonas*, and the 16S rRNA gene sequence similarity was 100 %. The DNA homology between the two strains was 99.7 %. The 16S rRNA and *rpoD* gene sequences of the two strains showed highest similarity values to *Pseudomonas oryzae* CGMCC 1.3392^T and *Pseudomonas psychrotolerans* DSM 15758^T (sharing 99.31 and 94.34 %, respectively). The major fatty acids of two strains were identified as summed feature 8 (C_{18:1}ω7c and/or C_{18:1}ω6c), C_{16:0} and summed feature 3 (C_{16:1}ω7c and/or C_{16:1}ω6c), and the major respiratory quinone was identified as ubiquinone Q-9, which are typical chemotaxonomic features of members of the genus *Pseudomonas*. The genomic DNA G+C contents of strains RY24^T and ZYY160 were determined to be 64.25 and 64.21 mol%, respectively. The DNA–DNA relatedness and average nucleotide identity values between the two strains and their closely related type strains were below 36 and 90 %, which supported that RY24^T and ZYY160 represent a novel species in the genus *Pseudomonas*. Phylogenetic and chemotaxonomic evidence, together with phenotypic characteristics, showed that the two isolates constitute a novel species of the genus *Pseudomonas*. The type strain is RY24^T (JCM 33201^T=ACCC 61555^T), for which the name *Pseudomonas rhizoryzae* sp. nov. is proposed.

The genus *Pseudomonas*, belonging to the family *Pseudomonadaceae* in the phylum *Proteobacteria*, was first proposed by Palleroni [1]. Membership of the genus has been growing rapidly and many novel species have been reported in recent years. The genus currently contains more than 200 validly named species (www.bacterio.net/pseudomonas.html). They play important roles in the environment, disease and food safety [2, 3]. For example, members of the genus *Pseudomonas* are not only able to produce interesting metabolic products of use in biotechnology, but are also well known to be active participants in the carbon cycle in nature [4–6]. The common characteristics of the genus are cells that are Gram-stain-negative and rod-shaped. Some of them produce different pigments [7]. Ubiquinone Q-9 is the major respiratory quinone and summed feature 8 (C_{18:1}ω7c and/or C_{18:1}ω6c) is the major fatty acid component in this genus. The DNA G+C content of this genus ranges from 58 to 69 mol% [8, 9]. In this

study, we isolated two novel strains belonging to the genus *Pseudomonas* from rice seeds and roots and their taxonomic assignment is described using a polyphasic approach.

Strain RY24^T was isolated from rice seeds of a long-term rice–rape rotation and ZYY160 was isolated from rice roots of a long-term rice–rice–*Astragalus* rotation. The sampling site is located in the Monitoring Experimental Station for Red Soil Eco-environment of the Ministry of Agriculture in Qiyang Country in Hunan Province, PR China (111° 52' 32" E 26° 45' 42" N; elevation, 150–170 m). Rice roots were sampled at tillering stage and seeds were collected at harvest time. Samples were used to isolate endophytic bacteria within 2 days. The treatment processes used for the roots and seeds was similar. Firstly, rice roots and seeds were washed in sterile tap water to remove surface soil and adherents. Subsequently, the surface of rice roots and seeds were sterilized by combining fresh sodium hypochlorite solution with ethanol

Author affiliations: ¹Key Laboratory of Microbial Resources Collection and Preservation, Ministry of Agriculture, Institute of Agricultural Resources and Regional Planning, Chinese Academy of Agricultural Sciences, Beijing 100081, PR China; ²College of Life Science, The Yangtze university, Jingzhou, 434025, PR China; ³Qiyang Agro-ecosystem of National Field Experimental Station, Institute of Agricultural Resources and Regional, Chinese Academy of Agricultural Sciences, Qiyang 426182, PR China.

*Correspondence: Xiao-Xia Zhang, zhangxiaoxia@caas.cn

Keywords: *Pseudomonas*; endophytic bacteria; rice seeds; rhizosphere.

Abbreviations: ANI, average nucleotide identity; DDH, DNA–DNA hybridization; GGDC, Genome-to-Genome Distance Calculator; R2A, Reasoner's 2A agar; TSA, tryptic soy agar; TSB, tryptic soy broth.

The GenBank/EMBL/DDBJ accession number for the 16S rRNA gene sequence of strain RY24^T is MK759856 and for strain ZYY160 it is MK775461. The whole genome sequences of strains RY24^T and ZYY160 have been deposited into GenBank/EMBL/DDBJ under accession numbers SULM00000000 and SULN00000000, respectively

Four supplementary figures are available with the online version of this article.