



First report of *Pantoea ananatis* in japonica rice varieties in Turkey

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Seed samples from twenty japonica rice (*Oryza sativa* subsp. *japonica*) cultivars were obtained from rice farmers from different regions (Hakkari, Kastamonu, Samsun, Sinop and Edirne Province) in Turkey during the 2016 and 2017 growing seasons. The seeds were surface-disinfected with 1% sodium hypochlorite for 2 min and washed in sterile distilled water. The seed samples (10 g for each cultivar) were placed in 10 ml of PBS buffer (0.01 M, pH 7.4) supplemented with a drop of Tween 20 and shaken at 175 rpm for 2 h at 28 °C. From each sample suspension 50 µl was plated onto nutrient agar (NA) and incubated at 28 °C for 48 h. The isolate named PaTo34a1, with hypersensitive reaction on tobacco (*Nicotiana tabacum* cv. “Samsun-NN”) plants by injection infiltration technique was selected based on colony morphology and biochemical characteristics. Other isolates, showing no hypersensitivity were eliminated. Isolate PaTo34a1 was motile, rod-shaped, gram-negative and facultatively anaerobic. Its fresh colonies were circular (1–2 mm diameter), yellow, convex with entire edges and smooth after 24 h at 28 °C on NA. PaTo34a1 showed positive results for catalase, β-galactosidase and Voges-Proskauer tests, produced acid from D-melibiose, L-arabinose and inositol and negative for reduction of nitrate to nitrite and production of arginine dihydrolase. Physiological and biochemical characterisation confirmed that PaTo34a1 belonged to the genus *Pantoea* (Deletoile et al. 2009). The PaTo34a1 16S rDNA gene fragment was amplified using the primers 27F (5' AGAGTTTGATC(AC)TGGCTCAG3') and 1492R (5' ACGG(CT)TACCTTGTTACGAC TT3') (Weisburg et al., 1991). BLASTN analysis of a 1500 bp 16S rDNA gene sequence (GenBank Accession No. MH549218) showed 100% identity to *Pantoea ananatis* strain CSA35 (KM091726) and

P. ananatis strain 37 (KM091724). Pathogenicity was verified on four-week-old rice plants (cv. “Osmancik”). Inoculated plants were kept at 26 ± 1 °C, 70% relative humidity in a greenhouse and 4000 lx light in a 5 day/night. Symptoms on the inoculated leaves were yellowing with brown stripes on both halves of the leaf blade. Sequence analysis of PCR product of the 16S rRNA gene from isolate PaTo34a1 from inoculated plants showed 100% nucleotide identity with the original bacterial isolates and confirmed as *P. ananatis*. To the best of our knowledge, this study describes the first finding of *P. ananatis* in seed of japonica rice in Turkey. The importance of the presence of this pathogen and the disease it may cause in the field in Turkey is unknown, as it is in other countries, e.g. Brazil and Korea (Carrer Filho et al., 2018, Lee et al., 2010) and remains to be investigated.

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