

Metagenomic analysis of *Phytophthora* diversity in nurseries of potted ornamental species

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The genetic diversity of *Phytophthora* was investigated in soil and root samples of potted ornamental and fruit tree plants collected in nurseries located in Apulia and Calabria (Southern Italy) using metagenomic approaches based on *Phytophthora* genus specific primers. PCR amplicons containing the ITS1 region of the rDNA were sequenced using both a conventional cloning and Sanger sequencing approach and a 454 pyrosequencing protocol. All sequences were accurately analyzed with an appropriate software and used as barcode for species identification utilizing a validated ITS database. The cloning/Sanger sequencing approach enabled the identification of nine different *Phytophthora* taxa (*P. nicotianae*, *P. citrophthora*, *P. meadii*, *P. cinnamomi*, *P. parvispora*, *P. cambivora*, *P. niederhauserii*, *P. taxon Pgchlamido*, and *P. lateralis*), 3 phylotypes associated to “species complexes” (*P. citricola*, *P. cryptogea* and *P. pseudosyringae*) and three other phylotypes considered as unknown or non well identified *Phytophthora* taxa. The 454 pyrosequencing confirmed above results and provided a higher levels of accuracy enabling the detection of four additional species (*P. cactorum*, *P. psycrophila*, *P. palmivora* and *P. ramorum*) and a general higher level of diversity (number of detected genotypes) within analyzed samples. Data of the present study indicate the use of genus specific primers combined with next generation sequencing approaches as valuable tools to investigate *Phytophthora* diversity in different environments and pathosystems. Furthermore, the large number of genotypes and *Phytophthora* taxa detected in a limited geographic area confirms a primary role of nurseries in favoring the diffusion and the evolution of *Phytophthora* species.