

## Phytophthora citrus diseases: current situation and management strategies in Tunisia

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In the Mediterranean areas, citrus is an important agricultural crop, with a production of about 10 million tons/year. Citrus fruits constitute a significant portion of the agricultural outputs in Tunisia. In 2019/2020, a land area of 29 thousand hectares was occupied by citrus. However, production in this region is affected by *Phytophthora* diseases. In spring 2023, a survey was conducted in Tunisian citrus orchards, to study the citrus farms characteristics (type of management, citrus varieties, geographic data), farmers' knowledge of the pathogens and management strategies applied to control *Phytophthora* spp. The results showed that the majority of the farmers were male, middle-aged adults and had a secondary education. Most of the citrus orchards were of medium area (59%) and the most common densities were 350, 360 and 400 ha. *Citrus sinensis* L. Osbeck and *C. limon* varieties planted on *C. aurantium* rootstock were the most used. Symptoms of *Phytophthora* citrus diseases consisted mainly in gummosis, dieback, wilting, and damping-off. Such information should be taken into consideration for a best management strategy of *Phytophthora* citrus diseases.

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## Endophytic, canker-inducing Botryosphaeriaceae causing flowering ash (*Fraxinus ornus*) dieback in central Italy

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A widespread dieback of flowering ash (*Fraxinus ornus* L.), with high mortality of young regeneration, was observed in some woody areas of central Italy (Tuscany). Symptoms appeared at the beginning of the growing season as typical sunken, light-brown cankers on the stem of young trees, which died as soon as the cankers girdled the stems. Many plants reacted vigorously to infection trying to callus over the lesions. Starting from 2018, a study was undertaken to clarify the etiology of the disease. Three botryosphaeriaceous fungi were isolated with high frequency from symptomatic plants: *Botryosphaeria dothidea*, *Diplodia fraxini* and *Neofusicoccum parvum*; a *Fusarium* sp. was also sometimes isolated. Artificial infection tests were carried out in July 2022 on two-year-old *Fraxinus ornus* seedlings with the four fungal taxa. All the botryosphaeriaceous fungi produced cankered lesions, with *B. dothidea* which reproduced the typical elongated cankers observed in the field. It is hypothesized that environmental stress was the main factor weakening trees and predisposing them to aggressive attack and colonization by pathogenic Botryosphaeriaceae. In fact, the areas with higher disease incidence and severity were those characterized by harsh environmental conditions, with low moisture, prolonged drought, especially on poor and gravelly soils.

## First report of *Phytophthora megasperma* and *P. pseudocryptogea* on *Rosmarinus officinalis* in Tuscany, Italy

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In January 2023, a severe dieback of *Rosmarinus officinalis* L. plants was observed in a rosemary plantation at Casole d'Elsa, Siena (Italy). Symptoms, including root rot, browning of the collar with discoloration of the internal tissue, leaf blight and stunting, closely resembled the typical root and crown rot syndrome induced by *Phytophthora* species. The lateral flow immunoassay test specific for *Phytophthora* species applied *in situ* on *R. officinalis* on symptomatic tissue samples gave positive results. In order to identify the associated *Phytophthora* taxa, roots, stem and soil samples were collected and taken to the laboratory. The baiting method was used in the laboratory for priming oomycetes. Only one of the two

cultured morphotypes, produced gametangia and oospores, confirming to be homothallic. The other one was heterothallic. DNA-based identification of these isolates revealed complete (100%) homology with respectively *Phytophthora megasperma* and *P. pseudocryptogea* for both rDNA-ITS and CoxI gene regions. This is the first report of *P. megasperma* and *P. pseudocryptogea* causing disease to *R. officinalis* in Italy. The pathogenic occurrence of these two harmful oomycetes on *R. officinalis*, could represent a new problem to the cultivation of this economically important aromatic plant species in the Mediterranean area.

### Microbiome innovations to support the ecological transition in agriculture

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Plant microbiomes are key components for ecosystem health in all terrestrial ecosystems including agriculture. The plant microbiota, which consist of bacteria, archaea, protists and fungi, is vertically transmitted by seeds and replenished horizontally from soil. All plants are holobionts and form a functional unit with its microbiome. Plant diversification and co-evolution shaped the plant microbiome and designed their specific composition and functional interplay including natural biocontrol of pathogens. Human activities in the Anthropocene, and especially intense agriculture, are linked to a significant shift of diversity and evenness of the plant microbiota. This shift is characterized by a decrease of host specificity and symbionts, and an increase of r-strategic microbes, pathogens, and hypermutators. Findings from plant microbiome research over the past 20 years clearly call for management of the microbiome and joint attention to the crop holobiont. Plant microbiomes can be managed either directly by applying (i) microbiome transplants, (ii) microbes with beneficial properties, or (iii) microbiota-active metabolites, or indirectly by changing environmental conditions in a way that microbiomes also shift their structure and function from dysbiosis into a healthy state. Examples for the different strategies for plant protection will be presented, and risk associated with the technology will be discussed. Beyond, the plant microbiome is connected across systems and crucial for human and planetary health

issues as well. This will be discussed in frame of the biodiversity crisis and the planetary boundary concept.

### Evaluation of soil quality and differences in microbial community structure in asymptomatic and symptomatic kiwifruit orchards

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*Actinidia chinensis* has been affected in Italy since 2012 by the Kiwifruit Vine Decline Syndrome (KVDS), which causes many plant and production losses. This study aims to characterize the aetiology of KVDS, using both traditional and molecular approaches, and to assess the impact of the disease on soil health and biodiversity. Root and rhizosphere soil samples were collected in 2022 from 5 kiwifruit orchards in Lazio, both from asymptomatic and symptomatic trees (n = 4 replicates), in two different seasons: spring and autumn. Isolations from symptomatic roots and baiting technique from rhizosphere samples confirmed the presence of potential kiwi pathogens, i.e.: *Cylindrocarpon* spp., in particular *Ilyonectria liriiodendri*, *I. robusta*, *Dactylonectria pauciseptata*, *D. ecuadoriensis*, and oomycetes, *Phytophthora vexans*, *P. chamaehyphon*, *P. litorale*, *P. helioides*, *Pythium dissotocum*, *Phytophthora plurivora*, and *P. acerina*. Pathogenicity tests of the most representative isolates are in progress at CREA-DC of Rome. Physico-chemical characteristics (pH, electrical conductivity, total nitrogen, total organic carbon, elemental contents, water-soluble carbon), enzyme activities (urease, phosphatase, and  $\beta$ -glucosidase), microbial activity (basal soil respiration), biomass and structure of the microbiome using phospholipid fatty acid (PLFAs) extraction were analyzed on soil collected from the rhizosphere of healthy and diseased kiwi plants to understand possible correlations with KVDS. Preliminary results showed well-defined diversity between