forests

IMPACT FACTOR 2.9

CITESCORE 4.5

Article

Diversity of *Phytophthora* Communities across Different Types of Mediterranean Vegetation in a Nature Reserve Area

Mario Riolo, Francesco Aloi, Federico La Spada, Saverio Sciandrello, Salvatore Moricca, Elena Santilli, Antonella Pane and Santa Olga Cacciola

Special Issue
Forest Pathology and Entomology

Edited by
Prof. Dr. Salvatore Moricca and Prof. Dr. Tiziana Panzavolta

MDPI

https://doi.org/10.3390/f11080853

forests

MDPI

Article

Diversity of *Phytophthora* Communities across Different Types of Mediterranean Vegetation in a Nature Reserve Area

Mario Riolo [1,2,3,†], Francesco Aloi [1,4,†], Federico La Spada [1], Saverio Sciandrello [5], Salvatore Moricca [6], Elena Santilli [2], Antonella Pane [1,*] and Santa Olga Cacciola [1,*]

1 Department of Agriculture, Food and Environment, University of Catania, 95123 Catania, Italy; mario.riolo@unirc.it (M.R.); francesco.aloi@unipa.it (F.A.); federicolaspada@yahoo.it (F.L.S.)
2 Council for Agricultural Research and Agricultural Economy Analysis, Research Centre for Olive, Citrus and Tree Fruit-Rende CS (CREA-OFA), 87036 Rende, Italy; elena.santilli@crea.gov.it
3 Department of Agricultural Science, Mediterranean University of Reggio Calabria, 89122 Reggio Calabria, Italy
4 Department of Agricultural, Food and Forest Sciences, University of Palermo, 90128 Palermo, Italy
5 Department of Biological, Geological and Environmental Sciences, University of Catania, 95125 Catania, Italy; s.sciandrello@unict.it
6 Department of Agricultural, Food, Environmental and Forestry Science and Technology, University of Florence, 50144 Firenze, Italy; salvatore.moricca@unifi.it
* Correspondence: apane@unict.it (A.P.); olga.cacciola@unict.it (S.O.C.)
† These authors contributed equally to the study.

Received: 9 July 2020; Accepted: 4 August 2020; Published: 6 August 2020

Abstract: Research Highlights: Protected natural areas are a reservoir of *Phytophthora* species and represent the most suitable sites to study their ecology, being less disturbed by human activities than other environments. Background and Objectives: The specific objective of this study was to correlate the diversity and distribution of *Phytophthora* species with the vegetation in aquatic, riparian and terrestrial habitats within a protected area in Eastern Sicily, Southern Italy. Materials and Methods: Environmental samples (water and soil) were sourced from two streams running through the reserve and six different types of vegetation, including *Platano-Salicetum pedicellatae*, the *Sarcopoterium spinosum* community, *Myrto communis-Pistacietum lentisci*, *Pistacio-Quercetum ilicis*, *Oleo-Quercetum virgilianae* and a gallery forest dominated by *Nerium oleander* (Natura 2000 classification of habitats). *Phytophthora* species were recovered from samples using leaf baiting and were classified on the basis of morphological characteristics and sequencing of internal transcribed spacer (ITS) regions of ribosomal DNA (rDNA). Results: As many as 11 *Phytophthora* species, within five different ITS clades, were identified, including *P. asparagi*, *P. bilorbang*, *P. cryptogea*, *P. gonapodyides*, *P. lacustris*, *P. multivora*, *P. nicotianae*, *P. oleae*, *P. parvispora*, *P. plurivora* and *P. syringae*. No *Phytophthora* species were found in the *Sarcopoterium spinosum* comm. *Phytophthora asparagi*, *P. lacustris* and *P. plurivora* were the prevalent species in the other five plant communities, but only *P. plurivora* was present in all of them. Overall aquatic species from clade 6 (100 out of 228 isolates) were the most common; they were recovered from all five types of vegetation, streams and riparian habitats. *Phytophthora* populations found in the *Platano-Salicetum pedicellatae* and *Oleo-Quercetum virgilianae* show the highest diversity, while no correlation was found with the physicochemical characteristics of the soil. *Conclusions:* The vegetation type and the aquatic or terrestrial habitat were identified as major environmental factors correlated with the diversity of *Phytophthora* communities in this reserve.

Keywords: leaf baiting; rDNA ITS regions; soil; water; ITS clades; Mediterranean vegetation; ecology; soil inhabitants; aquatic species

1. Introduction

The genus *Phytophthora* (Pythiaceae, Peronosporales, Oomycota, Chromista) comprises to date over 180 described species while, according to a conservative estimate, the actual number of species in this genus is at least double, if not triple [1,2]. Many *Phytophthora* species, such as *Phytophthora infestans*, *P. cinnamomi* and *P. ramorum*, are destructive plant pathogens causing severe crop losses and tree decline worldwide [3–10]. Most plant pathogenic *Phytophthora* species are polyphagous, with a host range encompassing plants of different families [11–15], and are typically soil inhabitants, although a number of them that produce deciduous sporangia have partially or temporarily adapted to an aerial lifestyle [16]. A more restricted number of species in the phylogenetic ITS clade 6 recovered from water courses, lakes and irrigation basins are functionally more adapted to aquatic habitats [17–21]. In general, these aquatic *Phytophthora* species are weakly aggressive as plant pathogens and, consequently, it has been assumed they behave as saprotrophs in plant debris in water and as opportunistic pathogens in riparian habitats. However, their ecological role in ecosystems is not fully understood.

Human-mediated transport, mainly the trade of nursery plants, has been identified as a major pathway for the introduction of non-native *Phytophthora* species into new areas [22–26]. It was demonstrated, e.g., that there is a causal link between the ornamental plant industry and the introduction of the destructive oak pathogen *P. ramorum* in the wildland in North America [27]. The use of nursery plants for forest restoration and afforestation is a way to introduce and spread exotic *Phytophthora* species in natural habitats and forests [28–30]. In a survey of protected natural areas in Sicily, 13 out of 20 *Phytophthora* recovered species were putatively exotic and only seven could be considered endemic to Europe [31]. In many cases, *Phytophthora* species found in forests and natural or naturalized ecosystems included aggressive plant pathogens of cultivated plants, suggesting that these ecosystems may act, in turn, as potential sources and reservoirs of *Phytophthora* inoculum for agricultural crops [32]. As a consequence, monitoring of forest and natural ecosystems should be included in *Phytophthora* species surveillance and biosecurity schemes. The establishment of alien invasive *Phytophthora* species in natural ecosystems has destabilizing effects as it affects the ecosystem homeostasis and resilience. The invasion of natural and semi-natural ecosystems by these pathogens may endanger native and rare plant species and are a threat to the diversity of plant communities [27,32–34]. Hence, the knowledge of resident *Phytophthora* populations should be a prerequisite for a rational management strategy of protected natural areas (PNAs).

The refinement of baiting and sampling methods, together with rapid advances in molecular diagnostics and DNA-sequencing technology [35–54], facilitated the detection of *Phytophthora* in environmental samples and stimulated the study of *Phytophthora* communities in forest and natural ecosystems all over the world, including watercourses and still unexplored areas [55–67]. These surveys revealed the richness of *Phytophthora* diversity in these ecosystems and led to the discovery of an impressive number of cryptic new species in this genus. They contributed to a better understanding of the global diversity of *Phytophthora*, the geographic radiation pathways of single *Phytophthora* species and clades from their centers of origin, their lifestyle and, in particular, their reproductive behavior and adaptation to different environments [2]. Despite numerous surveys, the environmental factors shaping the *Phytophthora* populations and conditioning their compositional changes in the wild have been poorly investigated. However, PNAs, being less disturbed by human activities, are the most suitable context for studying the ecology of *Phytophthora* species. The main objective of this study was to investigate whether the diversity of *Phytophthora* species and their frequency and spatial distribution across a small nature reserve in Sicily (Southern Italy) are correlated with the type of vegetation and the preferential habitat.

2. Materials and Methods

2.1. Sampling Area

Sampling activities were carried out in the "Complesso Speleologico Villasmundo—S. Alfio" regional nature reserve (strict nature reserve), part of the Special Areas of Conservation (SAC) (ITA090024) "Cozzo Ogliastri" in the municipality of Melilli (Sicily, Italy) (Figure 1). The reserve is managed by the CUTGANA (University of Catania) and was established in 1998 in order to protect one of the most important karst systems of the Hyblean area. The reserve covers a surface of 72 hectares in the north-eastern sector of the Climiti Mountains, between the "Belluzza" and "Cugno di Rio" streams. Along the river "Cugno di Rio", there are cave entrances (A zone, the core zone of the reserve). The buffer zone of the reserve (B zone) hosts several natural ecosystems of great importance.

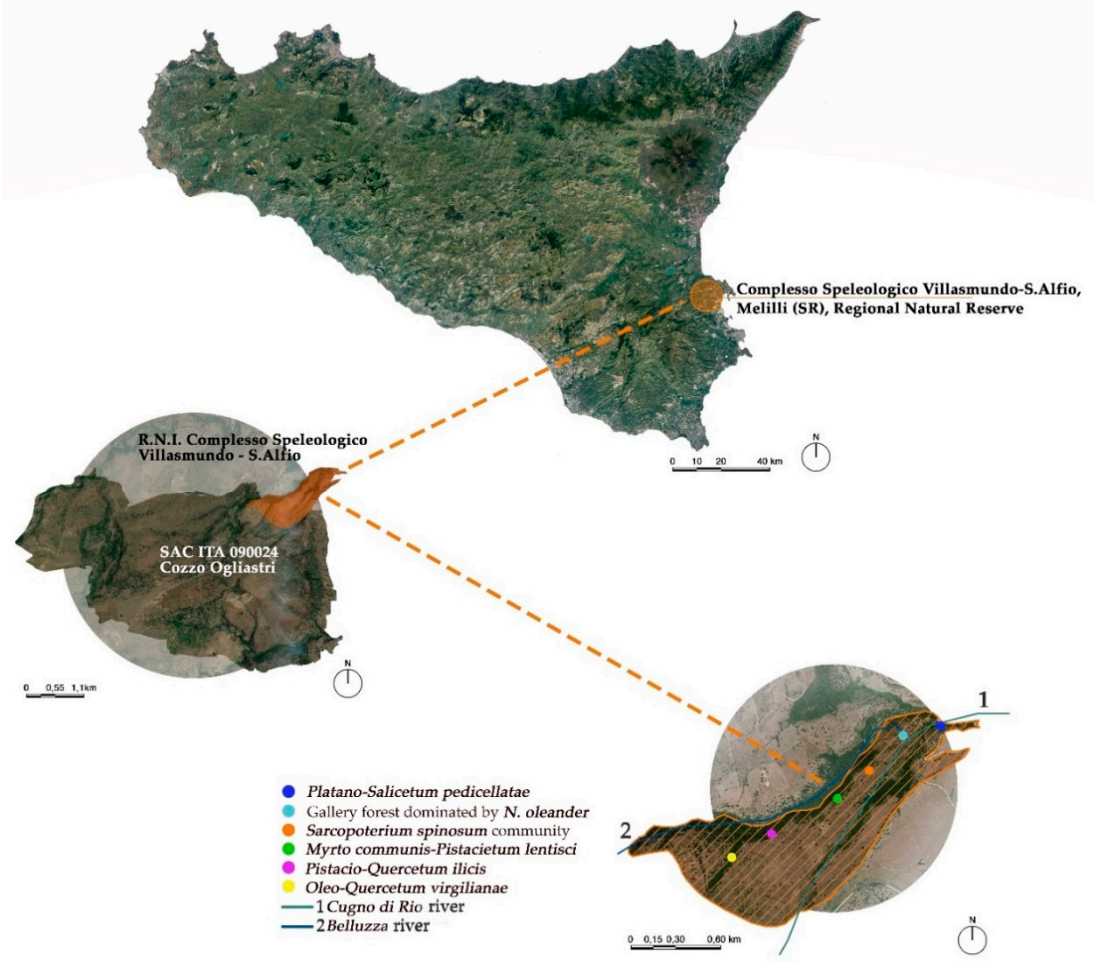


Figure 1. Geographical location of "Complesso Speleologico Villasmundo—S. Alfio" regional nature reserve (RNR) and plant communities within the reserve.

The plant communities and their respective habitats (according to the Habitats of Directive 92/43/EEC) where samples were collected are located in the B zone of the reserve and are described herein. (i) *Platano-Salicetum pedicellatae* Barbagallo, Brullo and Fagotto (Nature CODE 92C0 I): forests and woods, prevalently riparian, dominated by oriental plane (*Platanus orientalis* L.) and willow (*Salix pedicellata* Desf.), probably a relic of a more extended plane tree wood. (ii) *Sarcopoterium spinosum* community (Nature CODE 5420): low, thorny formations of hemispherical shrubs of the coastal thermo-Mediterranean zone of Aegean islands, of mainland Greece and the Ionian islands, of coastal Anatolia, much more widespread and diverse than the Western Mediterranean formations. (iii) *Myrto*

communis-Pistacietum lentisci (Molinier) Rivas-Martínez (Nature CODE 9320): thermo-Mediterranean woodland dominated by arborescent mastic (*Pistacia lentiscus* L.) and myrtle (*Myrtus communis* L.). (iv) *Pistacio-Quercetum ilicis* Brullo and Marcenò (Nature CODE 9340): Mediterranean oak stand characterized by holm oak (*Quercus ilex* L.). (v) *Oleo-Quercetum virgilianae* Brullo (Nature CODE: 91AA): mature climax community typified by southern live oak *(Quercus virgiliana* (Ten.) Ten.) in association with cork oak (*Q. suber* L.), holm oak (*Q. ilex* L.), carob (*Ceratonia siliqua* L.), wild olive (*Olea europaea* L. subsp. *sylvestris* (Mill.) Hegi)) and mastic (*P. lentiscus* L.). (vi) Gallery forest dominated by oleander (*Nerium oleander* L.) (Nature CODE 92CO I): thermo-Mediterranean community dominated by oleander (*N. oleander* L.) in association with willows (*Salix* spp.) and poplars (*Populus* spp.). Sampling activities were carried out during the autumn of 2015/2016 and 2017/2018. Plant nomenclature follows Pignatti [68], while the syntaxa classification follows Biondi et al. [69]. For the correlation between plant communities and habitat types, we referred to the Italian Interpretation Manual for the Habitats of Directive 92/43/EEC [70].

2.2. Sampling and Phytophthora Isolation

Twenty rhizosphere soil samples, including fine roots, were collected randomly from 20 mature trees and shrubs growing in all six plant communities (Table S1).

Soil sampling and isolation were performed in accordance with Jung et al. [31]: four soil cores were collected under each tree or shrub, 50–150 cm away from the stem base, and rhizosphere soil from all four cores was bulked together (about 1 L).

For each sample, subsamples of 400 mL were used for baiting tests that were performed in a walk-in growth chamber with 12 h natural daylight at 20 °C. Young leaves of *C. siliqua* and *Quercus* spp. floated over flooded soil were used as baits. After 24–48 h incubation, necrotic segments (2 × 2 mm) from symptomatic leaves were plated in Petri dishes onto selective PARPNH agar medium which consisted of 100 mL V8 juice (Campbell Grocery Products Ltd., Ashford, UK), 15 g agar, 3 g CaCO3, 200 mg ampicillin, 10 mg rifampicin, 25 mg pentachloronitrobenzene (PCNB), 50 mg nystatin, 50 mg hymexazol, and 1 L of deionised water [71]. Petri dishes were incubated at 20 °C in the dark. Outgrowing *Phytophthora* hyphae were transferred onto V8 juice agar (V8A) under the stereomicroscope. All the *Phytophthora* isolates were maintained on V8 agar in the dark at a temperature of 6 °C.

Additional isolations were performed directly from river water by using an in situ baiting technique. To this end, 10 non-wounded young leaves of *C. siliqua* and *Quercus* spp. were arranged in a mesh-bag styrofoam raft (25 × 30 cm) fixed to float on the water surface (Figure 2). In total, five mesh-bag-styrofoam rafts were placed: two on the surface of the Belluzza stream and three on the Cugno di Rio river, the two water courses crossing the reserve. The rafts were collected after 3 days. All obtained isolates were maintained on V8A and stored at 6 °C in the dark.

Figure 2. Mesh-bag styrofoam raft: (**A**) raft placed in Cugno di Rio river; (**B**) raft floating on the water surface of the Belluzza river.

2.3. Morphological Characterization of Isolates

Cultures of seven days, grown on V8A at 20 °C in the dark, were used to group all isolates into morphotypes on the basis of their colony growth patterns. For each host plant and plant community, the different morphological types have been labeled with progressive numbering (Roman numbering); then, isolates belonging to the same sampling hosts have been tagged with the relative type number.

Moreover, morphological features of chlamydospores, sporangia, oogonia, antheridia and hyphal swellings were carefully analyzed and compared with species descriptions in the literature [17,72].

2.4. Molecular Identification of Isolates

The DNA of the pure cultures of isolates obtained from soil and rafts was extracted by using PowerPlant® Pro DNA isolation Kit (MO BIO Laboratories, Inc., Carlsbad, CA, USA), following the manufacturer's protocol. The DNA was preserved at −20 °C. The identification of *Phytophthora* species was performed by the analysis of internal transcribed spacer (ITS) regions of ribosomal DNA (rDNA). DNA was amplified using forward primers ITS6 (5′-GAAGGTGAAGTCGTAACAAGG-3′) [73] and reverse primer ITS4 (5′-TCCTCCGCTTATTGATATGC-3′) [74]. The PCR amplification mix and thermocycler conditions were in accordance with Cooke et al. [73]. All PCRs were carried out in a 25 µL reaction mix containing PCR Buffer (1×), dNTP mix (0.2 mM), $MgCl_2$ (1.5 mM), forward and reverse primers (0.5 mM each), Taq DNA Polymerase (1 U) and 100 ng of DNA. The thermocycler conditions were as follows: 94 °C for 3 min; followed by 35 cycles of 94 °C for 30 s, 55 °C for 30 s, 72 °C for 30 s and then 72 °C for 10 min.

Amplicons were detected in 1% agarose gel and sequenced in both directions by an external service (Amsterdam, The Netherlands). Derived sequences were analyzed using FinchTV v.1.4.0 (Geospiza Inc., Seattle, WA, United States) [75]. For species identification, blast searches [76] in the *Phytophthora* Database [77], GenBank [78] and in a local database containing sequences of ex-type or key isolates from published studies were performed. Isolates were assigned to a species when their sequences were at least 99–100% identical to a reference isolate.

2.5. Analysis of Phytophthora Diversity

The *Phytophthora* diversity of soil samples sourced from the six plant communities was assessed by using the Shannon diversity (H = −Σpiln(pi)), the Pielou evenness (J = H/lnS) and the Simpson dominance (λ = 1/Σpi2) indices, where pi represents the frequency of each species and S the number of different species per plant community. Since the assumption of normal distribution was violated (the Shapiro–Wilk test was applied), the statistical differences in the diversity among sampling areas were assessed by the chi-square non-parametric test of Kruskal–Wallis followed by Dunn's multiple comparison post-hoc test (the R software [79] was used).

2.6. Soil Analysis and USDA Classification

An additional twenty rhizosphere soil samples were collected from plants within the selected sampling areas. The soil analysis was performed by a private laboratory (Progetto Ambiente & C. s.a.s., Catania, Italy) following the "official method of soil chemical analysis", in accordance with standard protocols defined by D.M. 13/09/1999, G.U. n°248, 21/10/99 and D.M. 25/03/2002, G.U. n°84, 10/04/2002. The following characteristics of the soil were determined: pH-H_2O, electrical conductivity at 25 °C, active limestone, organic matter content, nitrates and soil texture.

To define the soil texture of each sample, the USDA classification method [80] was used. The percentage of each soil component (sand, clay and silt) has been used in order to assign each sample to a textural class.

3. Results

Molecular analyses were performed on 228 isolates, of which there were 45 from rivers and 183 from the soil of the reserve. These isolates represented all morphotypes recovered in soil samples and baiting rafts. Morphological and ITS sequence analyses revealed the occurrence of multiple *Phytophthora* species in each type of plant community with the only exception of the *Sarcopoterium spinosum* vegetation, where no *Phytophthora* species was detected. ITS sequence analyses showed that all 228 isolates (65.9%) matched with 99–100% identity reference sequences of 11 known *Phytophthora* species belonging to five different ITS clades. The ITS sequences of isolates of this study were deposited at GenBank, and, since isolates of the same species were all the same, only some sequences were deposited in GenBank. The *Phytophthora* species recovered in the reserve, the host species and the accession numbers are given in Table S2.

Among the isolates, 100 belonged to species in ITS clade 6 (i.e., *P. bilorbang*, *P. asparagi*, *P. lacustris* and *P. gonapodydes*), 72 to species in ITS clade 2 (i.e., *P. multivora*, *P. oleae* and *P. plurivora*), 20 to species in ITS clade 8 (i.e., *P. cryptogea* and *P. syringae*) and 36 to *P. parvispora* and *P. nicotianae*, in ITS clades 7 and 1, respectively. The distribution of each *Phytophthora* species in each plant community type is shown in Table 1.

Table 1. Diagrammatic representation of the diversity and distribution of *Phytophthora* species recovered from six plant communities in the "Complesso Speleologico Villasmundo—S. Alfio" RNR. The proportion of isolates of each *Phytophthora* species recovered from each plant community is reported on the same row. The color intensity indicates the frequency of each species in each plant community (see legend).

Plant Community	*Phytophthora* Species Recovered in the Reserve										
	Clade 1		**Clade 2**			**Clade 6**			**Clade 7**	**Clade 8**	
	NIC	**MUL**	**OLE**	**PLU**	**ASP**	**LAC**	**GON**	**BIL**	**PAR**	**CRY**	**SYR**
Platano-Salicetum pedicellatae	16.7%	6.7%		3.3%		36.7%	3.3%	3.3%	6.7%	23.3%	
Sarcopoterium spinosum comm.											
Myrto communis-Pistacietum lentisci				34.4%	59.4%						6.25%
Pistacio-Quercetum ilicis				81.8%	4.5%	13.6%					
Oleo-Quercetum virgilianae	48%		8.7%	4.3%	35%					4.3%	
Gallery forest dominated by *N. oleander*				14.3%		71.4%		14.3%			

ASP = *P. asparagi*; **BIL** = *P. bilorbang*; **CRY** = *P. cryptogea*; GON = *P. gonapodyides*; LAC = *P. lacustris*; **MUL** = *P. multivora*; **NIC** = *P. nicotianae*; **OLE** = *P. oleae*; **PAR** = *P. parvispora*; **PLU** = *P. plurivora*; SYR = *P. syringae*. Underlined, the species that are considered native to Europe; in bold, species regarded as exotic [20,25,31]; the origin of *P. oleae* has not been established yet. % isolated species in each plant community: <25% 25–50% 50–75% >75%.

3.1. *Phytophthora* Diversity and Distribution in Different Plant Communities

With the only exception of the *Sarcopoterium spinosum* community, *Phytophthora* species from ITS clade 6 were isolated from all the plant communities (Figure 3). The most common *Phytophthora* species were aquatic species from ITS clade 6, such as *P. lacustris*, which were recovered from all river and riparian systems. *P. gonapodyides* and *P. bilorbang* were isolated from rhizosphere soil of willow while *P. lacustris* was recovered from river water and rhizosphere soil of mastic. With regard to species from clade 2, *P. oleae*, *P. multivora* and *P. plurivora*, they were detected in five out of six surveyed plant communities. In particular, *P. oleae* was recovered from rhizosphere soil of southern live oak, *P. multivora* from river water and *P. plurivora* from river water and rhizosphere soil of mastic, cork oak, southern live oak and holm oak. The clade 8 species *P. cryptogea* and *P. syringae* were found in three out of six plant communities. *P. cryptogea* was recovered from rhizosphere soil of willow, plane tree and southern live oak, while *P. syringae* was recovered only from rhizosphere soil of mastic. *Phytophtora parvispora* (ITS clade 7) was isolated exclusively from rhizosphere soil of mature willow trees in the *Platano-Salicetum pedicellatae* plant community. Finally, the ITS clade 1 species *P. nicotianae* was found in the *Platano-Salicetum pedicellatae* and *Oleo-Quercetum virigilianae* plant communities and was isolated from rhizosphere soil of plane, willow, southern live oak and cork oak (Table 1). *Phytophthora plurivora*, *P. asparagi*, *P. lacustris* and *P. nicotianae* were the prevalent species, accounting for 28%, 25%, 17% and 14% of all the isolates, respectively. Conversely, *P. syringae*, *P. gonapodyides*, *P. bilorbang*, *P. multivora*, *P. oleae* and *P. parvispora* were represented by less than 3% of isolates.

The structure of the community *of Phytophthora* species in soil samples differed significantly between the vegetation types. Among 11 *Phytophthora* species detected, only *P. plurivora* was found in all five plant communities. By contrast, *P. oleae* was detected exclusively in the *Oleo-Quercetum virgilianae*, *P. syringae* in the *Myrto Communis-Pistacietum lentisci*; *P. gonapodyides*, *P. bilorbang* and *P. parvispora* in the *Platanum-Salicetum pedicellatae*.

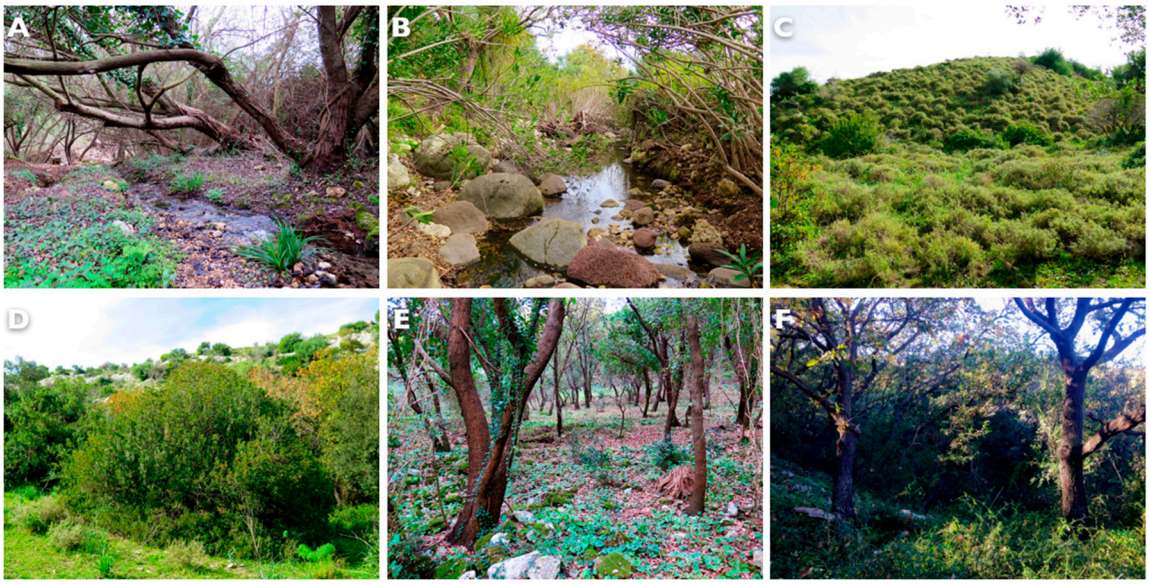
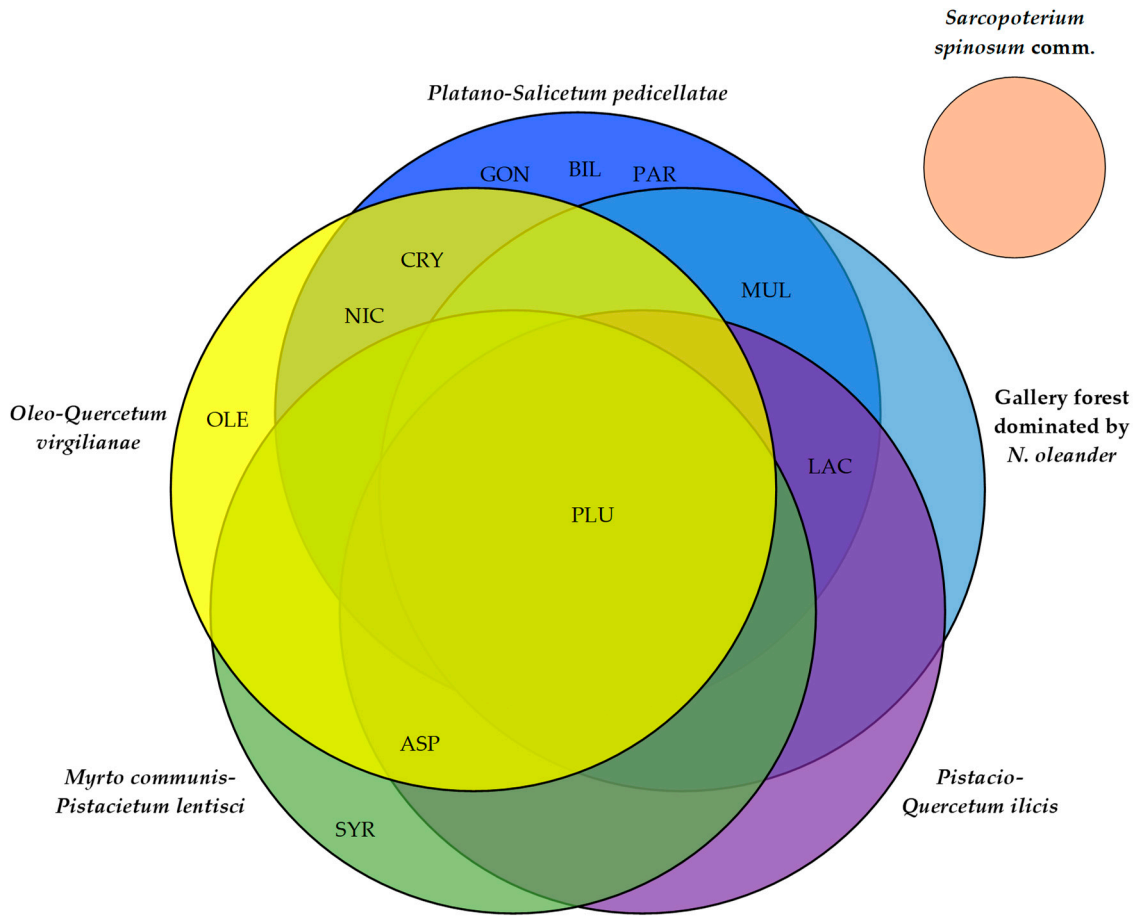


Figure 3. Plant communities: (**A**) *Platano-Salicetum pedicellatae*; (**B**) gallery forest dominated by *Nerium oleander*; (**C**) *Sarcopoterium spinosum* community; (**D**) *Myrto communis-Pistacietum lentisci*; (**E**) *Pistacio-Quercetum ilicis*; (**F**) *Oleo-Quercetum virgilianae*.

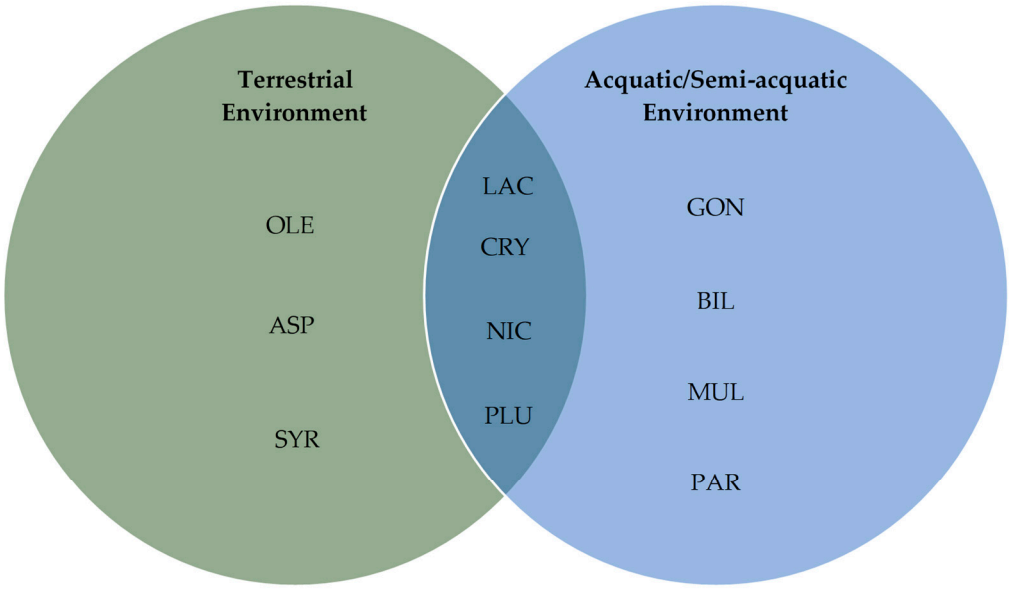
Overall, the sets of *Phytophthora* species from different plant communities strongly overlapped (Figure 4).



ASP= *P. asparagi*; BIL= *P. bilorbang*; CRY= *P. cryptogea*; GON= *P. gonapodyides*; LAC= *P. lacustris*; MUL= *P. multivora*; NIC= *P. nicotianae*; OLE= *P. oleae*; PAR= P. *parvispora*; PLU= *P. plurivora*; SYR= *P. syringae*

Figure 4. Venn diagram showing the distribution of *Phytophthora* species in six different plant communities in the "Complesso Speleologico Villasmundo—S. Alfio" RNR.

Comparing the different environments from which the species have been isolated, it can be observed that *P. cryptogea*, *P. lacustris*, *P. nicotianae* and *P. plurivora* were recovered from both terrestrial and aquatic or semi-aquatic environments, while *P. asparagi*, *P. oleae* and *P. syringae* were found exclusively in terrestrial environments and *P. bilorbang*, *P. gonapodyides*, *P. multivora* and *P. parvispora* only in aquatic or semi-aquatic environments. Overall, the sets of species from terrestrial habitats and from aquatic or semi-aquatic (riparian) habitats were distinct (Figure 5).



ASP= *P. asparagi*; BIL= *P. bilorbang*; CRY= *P. cryptogea*; GON= *P. gonapodyides*; LAC= *P. lacustris*; MUL= *P. multivora*; NIC= *P. nicotianae*; OLE= *P. oleae*; PAR= P. *parvispora*; PLU= *P. plurivora*; SYR= *P. syringae*

Figure 5. Venn diagram showing the distribution of *Phytophthora* species in terrestrial and aquatic or semi-aquatic environments in the "Complesso Speleologico Villasmundo—S. Alfio" RNR.

3.2. Analysis of Soil

Results of soil analyses are schematically summarized in Table 2. Values of soil pH from all plant communities were above 7.5 and not significantly different from each other. Concerning the electrical conductivity at 25 °C, moderately high values were found in soil samples from the *Platano-Salicetum pedicellatae* (1100 ± 48 µS/cm), *Sarcopoterium spinosum* comm. (973 ± 45 µS/cm) and *Myrto communis-Pistacietum lentisci* (894 ± 43 µS/cm) plant communities, high values in soil of the *Pistacio-Quercetum ilicis* (1414 ± 63 µS/cm) and significantly lower values in soil from the *Oleo-Quercetum virgilianae* (439 ± 36 µS/cm) plant community. As far as the active limestone is concerned, a high value was found in soil of the *Oleo-Quercetum virgilianae* (127 ± 5 g/Kg), while a relatively low value was recorded in soil from *Pistacio-Quercetum ilicis* (31 ± 2 g/Kg). The amount of nitrates was relatively high in soil from *Pistacio-Quercetum ilicis* and *Oleo-Quercetum virgilianae* plant communities (12.8 ± 1 and 11.59 ± 1 mg/Kg, respectively) and significantly lower in soil taken from *Platano-Salicetum pedicellatae*, *Sarcopoterium spinosum* comm. and *Myrto communis-Pistacietum lentisci*. Soils from all plant communities were rich in organic matter. The highest content of organic matter was found in soil from *Pistacio-Quercetum ilicis* (15%).

According to the USDA soil textural classification, soils from *Platano-Salicetum pedicellatae*, *Sarcopoterium spinosum* comm. and *Oleo-Quercetum virgilianae* were sandy clay loam; soil from *Pistacio-Quercetum ilicis* was clay loam and soil from the *Myrto communis-Pistacietum lentisci* was sandy clay. No obvious correlation was found between soil characteristics and *Phytophthora* species diversity.

Table 2. Soil characteristics in each plant community type of the "Complesso Speleologico Villasmundo—S. Alfio" RNR.

Soil Properties	Plant Community [a]				
	Platano-Salicetum pedicellatae	***Sarcopoterium spinosum* Comm.**	***Myrto communis-Pistacietum lentisci***	***Pistacio-Quercetum ilicis***	***Oleo-Quercetum virgilianae***
pH	7.5 ± 0.1	7.4 ± 0.1	7.3 ± 0.1	7.4 ± 0.1	7.6 ± 0.1
Electrical conductivity at 25 °C (µS/cm)	1.100 ± 48	973 ± 45	894 ± 43	1.414 ± 63	439 ± 36
Active limestone (g/Kg)	69 ± 3	86 ± 4	81 ± 4	31 ± 2	127 ± 5
Soil texture	Sandy clay loam	Sandy clay loam	Sandy clay	Clay loam	Sandy clay loam
Nitrates (mg/Kg)	6.7 ± 0.7	6.8 ± 0.6	4 ± 0.5	12.8 ± 1	11.59 ± 1
Organic matter (%)	5 ± 0.3	6 ± 0.5	8.2 ± 0.7	15 ± 2	6.1 ± 0.5

[a] Only rafts were placed in the gallery forest dominated by Nerium oleander and no soil sample was collected from this vegetation community.

3.3. Analysis of Phytophthora Diversity

The analysis of the diversity of *Phytophthora* populations from different plant communities showed a high variability of evenness (Table 3). Significantly higher values of Shannon and Pielou evenness diversity indices were observed in the *Phytophthora* populations from the *Platano-Salicetum pedicellatae* and *Oleo-Quercetum virgilianae* plant communities, while the evenness was moderate to low in *Phytophthora* populations from *Myrto communis-Pistacietum lentisci*, *Pistacio-Quercetum ilici* and the gallery forest dominated by *N. oleander*. In contrast, values of the Simpson dominance index were significantly higher in *Phytophthora* populations from the gallery forest dominated by *N. oleander* and *Pistacio-Quercetum ilicis*, intermediate in the *Phytophthora* population from *Myrto communis-Pistacietum lentisci* and significantly lower in the *Phytophthora* populations from *Platano-Salicetum pedicellatae* and *Oleo-Quercetum virgilianae* plant communities.

Table 3. Values of the diversity indices, Shannon diversity, evenness and Simpson dominance of *Phytophthora* populations from six different plant communities in the "Complesso Speleologico Villasmundo—S. Alfio" RNR. Data were analyzed with the Kruskal–Wallis test. Different letters indicate significant differences according to Dunn's multiple comparison test ($p \leq 0.01$).

Plant Communities	Diversity Indexes					
	Shannon Index		Pielou Evenness		Simpson Dominance	
Platano-Salicetum pedicellatae	1.707	a	0.821	a	0.229	c
Sarcopoterium spinosum comm.	-		-		-	
Myrto communis- Pistacietum lentisci	0.760	b	0.692	b	0.563	b
Pistacio-Quercetum ilicis	0.652	bc	0.593	c	0.649	a
Oleo-Quercetum virgilianae	1.205	a	0.749	ab	0.361	c
Gallery forest dominated by *N. oleander*	0.451	c	0.650	b	0.722	a

4. Discussion

As many as 11 *Phytophthora* species, including putatively endemic and exotic species as well as pathogens associated prevalently to agriculture, such as *P. nicotianae* [14,81], were found in the "Complesso Speleologico Villasmundo—S. Alfio" RNR, a relatively high number compared to the limited extension of the reserve (0.72 km^2). This can be explained with the quite recent establishment of the reserve (D. ARTA n° 616 04/11/1998), which, until 22 years ago, also comprised tree crops whose relics still survive and a high variability of plant communities included within the boundaries of the reserve, each occupying a different ecological niche and constituting a distinct ecosystem. The variability of ecosystems within the reserve is further increased by the presence of two streams, which favor the settlement of species with aquatic or semi-aquatic habitats along the banks and in the rhizosphere soil of the riparian vegetation.

Phytophthora plurivora, *P. asparagi* and *P. lacustris* were the most widespread species in the "Complesso Speleologico Villasmundo—S. Alfio" RNR, whereas the other species had a scattered distribution. Among the species recovered from the RNR, some, such as *P. cryptogea*, *P. gonapodyides*, *P. plurivora* and *P. syringae*, are common in natural and forest ecosystems throughout Europe [35,60,61,82], while others, like *P. bilorbang*, *P. oleae* and *P. parvispora*, occur more sporadically. Both *P. plurivora* and *P. gonapodyides* were reported in a previous survey of protected natural areas in Sicily [31]. In this study, *P. oleae* was isolated only from the rhizosphere soil of southern live oak (*Q. virgilianae* (Ten.) Ten.) in the *Oleo-Quecetum virgilianae* plant community. This was the first time that this recently described species was reported from a host plant other than olive (*O. europaea* L.) [83,84]. *Phytophthora bilorbang* is a prevalently aquatic species, but it has been occasionally reported as an opportunistic, aggressive plant pathogen [85]. *Phytophthora parvispora* (formerly *P. cinnamomi* var. *parvispora*), a species in clade 7a, is an aggressive plant pathogen with a prevalently terrestrial habitat. In this survey, it was recovered from the rhizosphere of willow (*S. pedicellata* Desf.) in a riparian semi-aquatic environment. In a previous study, it had been reported as a pathogen of strawberry tree (*Arbutus unedo*) in Sardinia (central Italy)

and described as a new species distinct from *P. cinnamomi* [86]. The first record of this species in Italy dates back to 2010 and was from ornamental plants in nursery [87].

Phytophthora gonapodyides is a species with a prevalently aquatic lifestyle but may be an aggressive opportunistic plant pathogen [88]. It inhibits seed germination and causes root rot and stem lesions in *Quercus robur* L. and *Q. ilex* L.; also, in association with other species of *Phytophthora*, it was recovered from declining oak stands even in xerophytic environments [71,89,90]. According to Erwin and Ribeiro [91], the damage caused by this species is underestimated and is overshadowed by that of other *Phytophthora* species, since it is traditionally regarded a minor pathogen. Presumably, the role of this and other *Phytophthora* species with a prevalently aquatic lifestyle in natural ecosystems is more complex than that of mere plant pathogens and deserves to be further investigated.

Phytophthora plurivora is a very polyphagous pathogen whose host range encompasses more than 80 woody host species including oaks (*Quercus* spp.), willows (*Salix* spp.), oleander (*N. oleander*) and oriental plane (*P. orientalis* L.) [31,92]. In the "Complesso Speleologico Villasmundo—S. Alfio" RNR, it was the species with the widest distribution and was isolated from the rhizosphere soil of mastic (*P. lentiscus* L.), southern live oak (*Q. virgilianae* (Ten.) Ten.), holm oak (*Q. ilex* L.) and cork oak (*Q. suber* L.), as well as from river water of *Platano-Salicetum pedicellatae*. Its polyphagia and the ability to produce resting spores, such as thick-walled oospores, might explain the cosmopolitan attitude of this species and its widespread occurrence in Mediterranean natural ecosystems. Interestingly, both *P. plurivora* and the other three species found exclusively in terrestrial habitats, i.e., *P. asaparagi*, *P. oleae* and *P. syringae*, are homothallic and produce oospores with thick walls. This could be a common adaptive strategy of these species to cope with adverse environmental conditions typical of many ecosystems in Mediterranean climate, such as long periods of drought alternating with intense rainfall, mild wet winters, high temperatures in summer, wide excursion of daily temperature and rapid fluctuation of air and soil humidity. In general, the ability to produce resting structures increases the competitiveness of *Phytophthora* spp. in terrestrial environments [93]. All the species isolated from the rhizosphere soil samples collected from diverse plant communities and streams in the "Complesso Speleologico Villasmundo—S. Alfio" RNR, with the only exception of *P. lacustris*, produce at least one kind of resting structure. However, only *P. asparagi, P. bilorbang*, *P. multivora*, *P. plurivora* and *P. syringae* are homothallic. In addition, *P. nicotianae, P. oleae, P. parvispora* and possibly *P. syringae* are able to produce chlamydospores.

Phytophthora lacustris is a ubiquitous species in riparian ecosystems, such as reed belts and riparian alder stands, throughout Europe and North America [20,94]. Like other species in clade 6, this species tolerates high temperatures, is sexually sterile and produces a great amount of zoospores [20]. For the clade as a whole, this combination of characters has been interpreted as an adaptation to riparian environments [17,95]. However, although *P. lacustris* is significantly less aggressive than other *Phytophthora* spp. with a terrestrial habitat, it can seriously infect fine roots of trees stressed by episodes of flooding or drought [20]. Accordingly, in the present study, *P. lacustris* was isolated prevalently from the stream crossing the gallery forest plant community, which is dominated by oleander (*N. oleander*), and from the rhizosphere soil of willow (*S. pedicellata*) trees in the *Platano-Salicetum pedicellate* riparian plant community, but *P. lacustris* was recovered even from the rhizosphere soil of southern live oak (*Q. virgilianae*) in the xerophytic *Pistacio-Quercetum ilicis* plant community.

Phytophthora asparagi, another species included in clade 6 but in a separate subclade [96], was recovered exclusively from terrestrial environments and was common in different plant communities, including *Myrto communis-Pistacietum lentisci*, *Pistacio-Quercetum ilicis* and *Oleo-Quercetum virgilianae*. In a previous survey of the National Park of La Maddalena archipelago (Sardinia), this species was isolated frequently from the rhizosphere soil of typical plants of the Mediterranean maquis vegetation, showing symptoms of decline, including white asparagus (*Asparagus albus* L.), *Phoenician juniper* (*Juniperus turbinata* Guss.) and mastic (*P. lentiscus* L.) [65]. In the "Complesso Speleologico Villasmundo—S. Alfio" RNR, *P. asparagi* was recovered from the rhizosphere soil of mastic as well as from the rhizosphere soil of evergreen oaks, including southern

live oak (*Q. virginiana* (Ten.) Ten.), holm oak (*Q. ilex* L.) and cork oak (*Q. suber* L.). As a result, the list of known host plants with which this species is associated has expanded.

In the present survey, two other species from clade 6, *P. gonapodiydes* and *P. bilorbang*, were detected. They were found exclusively in the riparian ecosystem and rivers, confirming a prevalently aquatic lifestyle [20,65,95,97]. Very probably, the high number of isolates of clade 6 species obtained in this study and their widespread occurrence is related to the high adaptability of these species to different environmental conditions and their ability to produce numerous zoospores, which in natural ecosystems are easily transported and spread by water courses.

The diversity and species richness of *Phytophthora* populations associated with different plant communities—as measured using three distinct indexes, Shannon diversity, Pielou evenness and Simpson dominance—were significantly higher in *Platano-Salicetum pedicellatae* and *Oleo-Quercetum virgilianae*, very probably reflecting the richness of host plant species and the complexity of these termo-mesophilous communities. Moreover, the higher richness and diversity of species in the *Platano-Salicetum pedicellatae* might be explained by the presence of both aquatic and terrestrial environments within this plant community. The Pielou evenness index value also suggests that the *Phytophthora* community in the *Platano-Salicetum pedicellatae* was more balanced than in the *Oleo-Quercetum virgilianae*, due to the dominance of *P. nicotianae*, an aggressive and polyphagous plant pathogen, in the latter community. Considering the proportions of isolates recovered from each of the abovementioned plant communities regarded together as an ecological succession, it can be supposed that the progressive decrease in complexity causes both the reduction in the diversity of *Phytophthora* communities and the progressive unbalance in their composition, resulting in the dominance of the most aggressive *Phytophthora* species. In accordance with this hypothesis, the very low complexity of the garrigue of *Sarcopoterium spinosum* (L.) Spach is the extreme ecological limit for the establishment of *Phytophthora* communities.

Nevertheless, the sets of *Phytophthora* species in each plant community, as represented by the Venn diagram, showed a tendency to cluster together, probably due to both the dominant presence of two invasive species (i.e., *P. plurivora* and *P. asparagi*) and the proximity of different ecosystems. The strong overlapping of *Phytophthora* species sets also indicates that environmental conditions are conducive to these oomycetes in all types of plant communities examined in this study. In particular, no obvious correlation was observed between the diversity of distribution of *Phytophthora* populations and some physicochemical soil properties that may influence the ecology of *Phytophthora* species, their aggressiveness and ability to survive, such as pH, salinity, textural class, active limestone, nitrates and organic matter content [98–104]. In all plant community types, the content of organic matter was relatively high, but this was particularly true for the *Pistacio-Quercetum ilicis*. This might explain the occurrence of *P. lacustris* in the rhizosphere soil of trees in this vegetation community. *Phytophthora* species, in fact, including several aggressive plant pathogens, have the ability to either survive or complete their lifecycle as saprobes, despite their poor ability to compete with other saprophytic organisms. This saprophytic attitude is more pronounced in clade 6 species, like *P. lacustris* [20]. Therefore, higher levels of organic matter favor the establishment and survival of *Phytophthora* spp., in general, and of clade 6 species, in particular.

Moreover, other soil parameters, with the only exception of the high level of active limestone in the *Oleo-Quercetum virgilianae*, which could be a limiting factor, can be considered within the optimum range in all plant community types, including the *Sarcopoterium spinosum* comm., where no *Phytophthora* species was found. This indicates that the aforementioned physicochemical soil properties were not a limiting factor for the settlement and survival of *Phytophthora* spp. in this xerophytic plant community type. Other major environmental factors conditioning the ecology of soilborne *Phytophthora* species are soil moisture and temperature [91]. The effects of soil water status, generally expressed in terms of matric potential, on the ability of *Phytophthora* species to sporulate and cause disease have been extensively investigated in agricultural systems [91,105]. The geographical range of many soilborne *Phytophthora* species or their ability to thrive and survive at high altitudes depend on their

extreme and optimum temperatures for growth [7,57,106] and many aquatic *Phytophthora* species are thermophilic [19,20,107], suggesting that this is an adaptive functional trait of this group of species. However, the values of these two soil parameters, matric potential and temperature, are not constant and, in the Mediterranean climate, they vary considerably and suddenly across the seasons or even the same day, so they were not considered in this study.

Overall, comparing *Myrto communis- Pistacietum lentisci*, *Pistacio-Quercetum ilicis* and *Sarcopoterium spinosum* comm., with *Platano-Salicetum pedicellatae* and *Oleo-Quercetum virgilianae*, the *Phytophthora* diversity shows a trend that could be related to the degree of maturity of plant communities. Results of this study are in agreement with those of a similar study carried out in forests of South Africa [108]. Conversely, the geographic distribution of *P. cinnamomi* in the Iberian Peninsula turned out to be influenced primarily by abiotic factors, including soil texture and climate, followed by land use and lastly by the presence of main host plant species [109]. However, comparisons with the results of this last study are impaired by the scale of the survey, the heterogeneity of environments investigated and the fact that only one Phytophthora species was involved.

When the *Phytophthora* species found in the "Complesso Speleologico Villasmundo—S. Alfio" RNR were grouped on the basis of the type of environment from which they were recovered, the Venn diagram clearly separated them into two distinct sets, including species with a prevalent or exclusive terrestrial habitat, such as *P. asparagi*, *P. oleae* and *P. syringae,* and species with a prevalent or exclusive aquatic or semi-aquatic habitat, such as *P. bilorbang* and *P. gonapodyides*, respectively. The first group was characterized by homothallic species, able to produce thick-walled oospores allowing them to survive adverse soil conditions; the second group comprised species that have adapted to thrive in aquatic and semi-aquatic environments. The presence of prevalently aquatic species in the first set and the presence of typically terrestrial species in the second set may be explained by the proximity of vegetation to water streams, flooding events and runoff of rain water. Consistently with our results, in a very recent study a correlation was found between the *Phytophthora* community and both the type of vegetation and environment in the French Guiana rainforest, which, like other neotropical forests, is considered a major plant diversity hotspot [110]. However, unexpectedly, the *Phytophthora* community in this forest showed a low diversity compared to the richness of species recovered from the "Complesso Speleologico Villasmundo—S. Alfio" RNR.

5. Conclusions

This study showed a correlation between the vegetation type and the diversity and distribution of *Phytophthora* species in a small natural reserve where different types of plant communities typical of the Mediterranean macroregion were represented. Moreover, it confirmed that *Phytophthora* species may be grouped according to an ecological criterion on the basis of their prevalently terrestrial or aquatic lifestyle. The ecology of these two distinct groups is conditioned by their aggressiveness as well as by some other biological characteristics, such as high temperature requirement, the ability to produce resting structures, the prevalence of asexual reproduction and the amount of zoospores that they produce. The presence of different ecosystems in a restricted area, i.e., in comparable environmental and climatic conditions, was a unique opportunity to highlight this correlation between the type of vegetation and the diversity of *Phytophthora* communities. However, only a more extensive survey of the same plant community's types in distant geographic areas would definitely demonstrate that the type of vegetation is a major driving factor shaping the *Phytophthora* communities in natural ecosystems. Other ecological and epidemiological aspects that deserve further attention are the seasonal pattern of *Phytophthora* populations in these ecosystems, the selective pressure exerted by invasive *Phytophthora* species on natural vegetation and their impact on spontaneous regeneration.

Supplementary Materials: The following are available online at http://www.mdpi.com/1999-4907/11/8/853/s1, Table S1: Vegetation, geographic coordinates, type of samples and soil texture of the 20 sites sampled in the "Complesso Speleologico Villasmundo—S. Alfio" regional nature reserve. Table S2: Isolates of the Phytophthora species characterized in this study, plant hosts of the "Complesso Speleologico Villasmundo—S. Alfio" regional Nature reserve and the GenBank accession numbers of their ITS sequences.

Author Contributions: Conceptualization, S.O.C., F.A., F.L.S., S.S. and M.R.; methodology, A.P., F.A., F.L.S., M.R., S.O.C. and S.S.; analysis of results, F.A., F.L.S., M.R. and S.O.C.; resources, A.P., S.O.C., E.S. and S.M.; original draft preparation, M.R., F.A., and S.O.C.; editing, M.R., F.L.S., F.A., S.M. and S.O.C.; supervision, S.O.C., S.M., E.S., A.P. and S.S. All authors have read and agreed to publish this version of the manuscript.

Funding: This research was funded by "MIUR-FFABR 2017" of S.O.C., grant number 5A725192051; University of Catania, "project DIFESA" of S.O.C. and A.P., grant number 5A722192134. This study is part of M.R. activity as PhD, Doctorate "Agricultural, Food and Forestry Science", University Mediterranea of Reggio Calabria, XXXV cycle.

Acknowledgments: The authors wish to thank the director of CUTGANA (Protected Area Management), University of Catania, Giovanni Signorello, and the manager of the Regional Nature Reserve, Elena Amore, for their support and helpful assistance.

Conflicts of Interest: The authors declare no conflict of interest.

References

1. Scott, P.; Bader, M.K.F.; Burgess, T.; Hardy, G.; Williams, N. Global biogeography and invasion risk of the plant pathogen genus *Phytophthora*. *Environ. Sci. Policy* **2019**, *101*, 175–182. [CrossRef]
2. Jung, T.; Scanu, B.; Brasier, C.M.; Webber, J.; Milenković, I.; Corcobado, T.; Tomšovský, M.; Pánek, M.; Bakonyi, J.; Maia, C.; et al. A survey in natural forest ecosystems of Vietnam reveals high diversity of both new and described phytophthora taxa including *P. ramorum*. *Forests* **2020**, *11*, 93. [CrossRef]
3. Rizzo, D.M.; Garbelotto, M.; Hansen, E.M. *Phytophthora ramorum*: Integrative research and management of an emerging pathogen in California and Oregon forests. *Annu. Rev. Phytopathol.* **2005**, *43*, 309–335. [CrossRef] [PubMed]
4. Haas, B.J.; Kamoun, S.; Zody, M.C.; Jiang, R.H.Y.; Handsaker, R.E.; Cano, L.M.; Grabherr, M.; Kodira, C.D.; Raffaele, S.; Torto-Alalibo, T.; et al. Genome sequence and analysis of the Irish potato famine pathogen *Phytophthora infestans*. *Nature* **2009**, *461*, 393–398. [CrossRef] [PubMed]
5. Grünwald, N.J.; Garbelotto, M.; Goss, E.M.; Heungens, K.; Prospero, S. Emergence of the sudden oak death pathogen *Phytophthora ramorum*. *Trends Microbiol.* **2012**, *20*, 131–138. [CrossRef]
6. Kamoun, S.; Furzer, O.; Jones, J.D.G.; Judelson, H.S.; Ali, G.S.; Dalio, R.J.D.; Roy, S.G.; Schena, L.; Zambounis, A.; Panabières, F.; et al. The Top 10 oomycete pathogens in molecular plant pathology. *Mol. Plant Pathol.* **2015**, *16*, 413–434. [CrossRef]
7. Burgess, T.I.; Scott, J.K.; Mcdougall, K.L.; Stukely, M.J.C.; Crane, C.; Dunstan, W.A.; Brigg, F.; Andjic, V.; White, D.; Rudman, T.; et al. Current and projected global distribution of *Phytophthora cinnamomi*, one of the world's worst plant pathogens. *Glob. Chang. Biol.* **2017**, *23*, 1661–1674. [CrossRef]
8. Moricca, S.; Linaldeddu, B.T.; Ginetti, B.; Scanu, B.; Franceschini, A.; Ragazzi, A. Endemic and emerging pathogens threatening cork oak trees: Management options for conserving a unique forest ecosystem. *Plant Dis.* **2016**, *100*, 2184–2193. [CrossRef]
9. Frisullo, S.; Lima, G.; Lio, G.M.D.S.; Camele, I.; Melissano, L.; Puglisi, I.; Pane, A.; Agosteo, G.E.; Prudente, L.; Cacciola, S.O. *Phytophthora cinnamomi* involved in the decline of holm oak (*Quercus ilex*) stands in southern Italy. *For. Sci.* **2018**, *64*, 290–298. [CrossRef]
10. Cacciola, S.O.; Gullino, M.L. Emerging and re-emerging fungus and oomycete soil-borne plant diseases in Italy. *Phytopathol. Mediterr.* **2019**, *58*, 451–472.
11. Hardham, A.R. Phytophthora cinnamomi. *Mol. Plant Pathol.* **2005**, *6*, 589–604. [CrossRef] [PubMed]
12. Grünwald, N.J.; Goss, E.M.; Press, C.M. *Phytophthora ramorum*: A pathogen with a remarkably wide host range causing sudden oak death on oaks and ramorum blight on woody ornamentals. *Mol. Plant Pathol.* **2008**, *9*, 729–740. [CrossRef] [PubMed]
13. Abad, Z.G.; Abad, J.A.; Cacciola, S.O.; Pane, A.; Faedda, R.; Moralejo, E.; Pérez-Sierra, A.; Abad-Campos, P.; Alvarez-Bernaola, L.A.; Bakonyi, J.; et al. *Phytophthora niederhauserii* sp. nov., a polyphagous species associated with ornamentals, fruit trees and native plants in 13 countries. *Mycologia* **2014**, *106*, 431–447. [CrossRef] [PubMed]

14. Panabières, F.; Ali, G.S.; Allagui, M.B.; Dalio, R.J.D.; Gudmestad, N.C.; Kuhn, M.L.; Guha Roy, S.; Schena, L.; Zampounis, A. *Phytophthora nicotianae* diseases worldwide: New knowledge of a long-recognised pathogen. *Phytopathol. Mediterr.* **2016**, *55*, 20–40.
15. Migliorini, D.; Khdiar, M.Y.; Padrón, C.R.; Vivas, M.; Barber, P.A.; Hardy, G.E.S.J.; Burgess, T.I. Extending the host range of *Phytophthora multivora*, a pathogen of woody plants in horticulture, nurseries, urban environments and natural ecosystems. *Urban For. Urban Green.* **2019**, *46*, 126460. [CrossRef]
16. Jung, T.; Pérez-Sierra, A.; Durán, A.; Jung, M.H.; Balci, Y.; Scanu, B. Canker and decline diseases caused by soil- and airborne *Phytophthora* species in forests and woodlands. *Persoonia-Mol. Phylogeny Evol. Fungi* **2018**, *40*, 182–220. [CrossRef]
17. Jung, T.; Stukely, M.J.C.; Hardy, G.E.S.J.; White, D.; Paap, T.; Dunstan, W.A.; Burgess, T.I. Multiple new *Phytophthora* species from ITS Clade 6 associated with natural ecosystems in Australia: Evolutionary and ecological implications. *Persoonia Mol. Phylogeny Evol. Fungi* **2011**, *26*, 13–39. [CrossRef]
18. Nagel, J.H.; Gryzenhout, M.; Slippers, B.; Wingfield, M.J.; Hardy, G.E.S.J.; Stukely, M.J.C.; Burgess, T.I. Characterization of *Phytophthora* hybrids from ITS clade 6 associated with riparian ecosystems in South Africa and Australia. *Fungal Biol.* **2013**, *117*, 329–347. [CrossRef]
19. Copes, W.E.; Yang, X.; Hong, C. *Phytophthora* species recovered from irrigation reservoirs in Mississippi and Alabama nurseries and pathogenicity of three new species. *Plant Dis.* **2015**, *99*, 1390–1395. [CrossRef]
20. Nechwatal, J.; Bakonyi, J.; Cacciola, S.O.; Cooke, D.E.L.; Jung, T.; Nagy, Z.Á.; Vannini, A.; Vettraino, A.M.; Brasier, C.M. The morphology, behaviour and molecular phylogeny of *Phytophthora* taxon Salixsoil and its redesignation as *Phytophthora lacustris* sp. nov. *Plant Pathol.* **2013**, *62*, 355–369. [CrossRef]
21. Redondo, M.A.; Boberg, J.; Stenlid, J.; Oliva, J. Contrasting distribution patterns between aquatic and terrestrial *Phytophthora* species along a climatic gradient are linked to functional traits. *ISME J.* **2018**, *12*, 2967–2980. [CrossRef]
22. Mammella, M.A.; Martin, F.N.; Cacciola, S.O.; Coffey, M.D.; Faedda, R.; Schena, L. Analyses of the population structure in a global collection of *Phytophthora nicotianae* isolates inferred from mitochondrial and nuclear DNA sequences. *Phytopathology* **2013**, *103*, 610–622. [CrossRef]
23. Migliorini, D.; Ghelardini, L.; Tondini, E.; Luchi, N.; Santini, A. The potential of symptomless potted plants for carrying invasive soilborne plant pathogens. *Divers. Distrib.* **2015**, *21*, 1218–1229. [CrossRef]
24. Biasi, A.; Martin, F.N.; Cacciola, S.O.; Lio, G.M.D.S.; Grünwald, N.J.; Schena, L. Genetic analysis of *Phytophthora nicotianae* populations from different hosts using microsatellite markers. *Phytopathology* **2016**, *106*, 1006–1014. [CrossRef] [PubMed]
25. Jung, T.; Orlikowski, L.; Henricot, B.; Abad-Campos, P.; Aday, A.G.; Casal, O.A.; Bakonyi, J.; Cacciola, S.O.; Cech, T.; Chavarriaga, D.; et al. Widespread *Phytophthora* infestations in European nurseries put forest, semi-natural and horticultural ecosystems at high risk of Phytophthora diseases. *For. Pathol.* **2016**, *46*, 134–163. [CrossRef]
26. Garbelotto, M.; Frankel, S.J.; Scanu, B. Soil- and waterborne *Phytophthora* species linked to recent outbreaks in Northern California restoration sites. *Calif. Agric.* **2018**, *72*, 208–216. [CrossRef]
27. Garbelotto, M.; Hayden, K.J. Sudden oak death: Interactions of the exotic oomycete *Phyotphthora ramorum* with native North American hosts. *Eukaryot. Cell* **2012**, *11*, 131–1323. [CrossRef] [PubMed]
28. Simamora, A.V.; Paap, T.; Howard, K.; Stukely, M.J.C.; Hardy, G.E.S.J.; Burgess, T.I. *Phytophthora* contamination in a nursery and its potential dispersal into the natural environment. *Plant Dis.* **2018**, *102*, 132–139. [CrossRef]
29. Sims, L.; Tjosvold, S.; Chambers, D.; Garbelotto, M. Control of Phytophthora species in plant stock for habitat restoration through best management practices. *Plant Pathol.* **2019**, *68*, 196–204. [CrossRef]
30. Sims, L.L.; Chee, C.; Bourret, T.; Hunter, S.; Garbelotto, M. Genetic and phenotypic variation of *Phytophthora crassamura* isolates from California nurseries and restoration sites. *Fungal Biol.* **2019**, *123*, 159–169. [CrossRef]
31. Jung, T.; La Spada, F.; Pane, A.; Aloi, F.; Evoli, M.; Jung, M.H.; Scanu, B.; Faedda, R.; Rizza, C.; Puglisi, I.; et al. Diversity and distribution of *Phytophthora* species in protected natural areas in Sicily. *Forests* **2019**, *10*, 259. [CrossRef]
32. Rizzo, D.M.; Garbelotto, M.; Davidson, J.M.; Slaughter, G.W.; Koike, S.T. *Phytophthora ramorum* as the cause of extensive mortality of *Quercus* spp. and *Lithocarpus densiflorus* in California. *Plant Dis.* **2002**, *86*, 205–214. [CrossRef] [PubMed]

33. Cahill, D.M.; Rookes, J.E.; Wilson, B.A.; Gibson, L.; McDougall, K.L. *Phytophthora cinnamomi* and Australia's biodiversity: Impacts, predictions and progress towards control. *Aust. J. Bot.* **2008**, *56*, 279–310. [CrossRef]
34. Jung, T.; Vettraino, A.M.; Cech, T.; Vannini, A. The impact of invasive *Phytophthora* species on European forests. In *Phytophthora: A Global Perspective*; CABI: Wallingford, UK, 2013.
35. Bonants, P.J.M.; Van Gent-Pelzer, M.P.E.; Hooftman, R.; Cooke, D.E.L.; Guy, D.C.; Duncan, J.M. A combination of baiting and different PCR formats, including measurement of real-time quantitative fluorescence, for the detection of *Phytophthora fragariae* in strawberry plants. *Eur. J. Plant Pathol.* **2004**, *110*, 689–702. [CrossRef]
36. Kong, P.; Hong, C.X.; Tooley, P.W.; Ivors, K.; Garbelotto, M.; Richardson, P.A. Rapid identification of *Phytophthora ramorum* using PCR-SSCP analysis of ribosomal DNA ITS-1. *Lett. Appl. Microbiol.* **2004**, *38*, 433–439. [CrossRef]
37. Khaliq, I.; St. Hardy, G.E.J.; White, D.; Burgess, T.I. eDNA from roots: A robust tool for determining *Phytophthora* communities in natural ecosystems. *FEMS Microbiol. Ecol.* **2018**, *94*, fiy048. [CrossRef]
38. Kunadiya, M.B.; Dunstan, W.D.; White, D.; Hardy, G.E.S.J.; Grigg, A.H.; Burgess, T.I. A qPCR Assay for the Detection of *Phytophthora cinnamomi* including an mRNA protocol designed to establish propagule viability in environmental samples. *Plant Dis.* **2019**, *103*, 2443–2450. [CrossRef]
39. Ioos, R.; Laugustin, L.; Schenck, N.; Rose, S.; Husson, C.; Frey, P. Usefulness of single copy genes containing introns in *Phytophthora* for the development of detection tools for the regulated species *P. ramorum* and *P. fragariae*. *Eur. J. Plant Pathol.* **2006**, *116*, 171–176. [CrossRef]
40. Bilodeau, G.J.; Lévesque, C.A.; De Cock, A.W.A.M.; Duchaine, C.; Brière, S.; Uribe, P.; Martin, F.N.; Hamelin, R.C. Molecular detection of *Phytophthora ramorum* by real-time polymerase chain reaction using TaqMan, SYBR Green, and molecular beacons. *Phytopathology* **2007**, *97*, 632–642. [CrossRef]
41. Cooke, D.E.L.; Schena, L.; Cacciola, S.O. Tools to detect, identify and monitor *Phytophthora* species in natural ecosystems. *J. Plant Pathol.* **2007**, *89*, 13–28.
42. Hwang, J.; Oak, S.W.; Jeffers, S.N. Detecting Phytophthora ramorum and other species of *Phytophthora* in streams in natural ecosystems using baiting and filtration methods. In Proceedings of the Sudden Oak Death Third Science Symposium, Albany, CA, USA, 5–9 March 2007.
43. Ghimire, S.R.; Richardson, P.A.; Moorman, G.W.; Lea-Cox, J.D.; Ross, D.S.; Hong, C.X. An in-situ baiting bioassay for detecting *Phytophthora* species in irrigation runoff containment basins. *Plant Pathol.* **2009**, *58*, 577–583. [CrossRef]
44. Martin, F.N.; Coffey, M.D.; Zeller, K.; Hamelin, R.C.; Tooley, P.; Garbelotto, M.; Hughes, K.J.D.; Kubisiak, T.; Bilodeau, G.J.; Levy, L.; et al. Evaluation of molecular markers for *Phytophthora ramorum* detection and identification: Testing for specificity using a standardized library of isolates. *Phytopathology* **2009**, *99*, 390–403. [CrossRef] [PubMed]
45. O'Brien, P.A.; Williams, N.; Hardy, G.E.S. Detecting *Phytophthora*. *Crit. Rev. Microbiol.* **2009**, *35*, 169–181. [CrossRef]
46. Sutton, W.; Hansen, E.M.; Reeser, P.W.; Kanaskie, A. Stream monitoring for detection of *Phytophthora ramorum* in Oregon tanoak forests. *Plant Dis.* **2009**, *93*, 1182–1186. [CrossRef] [PubMed]
47. Williams, N.; Hardy, G.E.S.J.; O'Brien, P.A. Analysis of the distribution of *Phytophthora cinnamomi* in soil at a disease site in Western Australia using nested PCR. *For. Pathol.* **2009**, *39*, 95–109. [CrossRef]
48. Tomlinson, J.A.; Dickinson, M.J.; Boonham, N. Rapid detection of *Phytophthora ramorum* and *P. kernoviae* by two-minute DNA extraction followed by isothermal amplification and amplicon detection by generic lateral flow device. *Phytopathology* **2010**, *100*, 143–149. [CrossRef] [PubMed]
49. Scibetta, S.; Schena, L.; Chimento, A.; Cacciola, S.O.; Cooke, D.E.L. A molecular method to assess *Phytophthora* diversity in environmental samples. *J. Microbiol. Methods* **2012**, *88*, 356–368. [CrossRef]
50. Schena, L.; Nicosia, M.G.L.D.; Sanzani, S.M.; Faedda, R.; Ippolito, A.; Cacciola, S.O. Development of quantitative PCR detection methods for phytopathogenic fungi and oomycetes. *J. Plant Pathol.* **2013**, *95*, 7–24.
51. Bilodeau, G.J.; Martin, F.N.; Coffey, M.D.; Blomquist, C.L. Development of a multiplex assay for genus- and species-specific detection of *Phytophthora* based on differences in mitochondrial gene order. *Phytopathology* **2014**, *104*, 733–748. [CrossRef]
52. Miles, T.D.; Martin, F.N.; Coffey, M.D. Development of rapid isothermal amplification assays for detection of *Phytophthora* spp. in plant tissue. *Phytopathology* **2015**, *105*, 265–278. [CrossRef]

53. Prigigallo, M.I.; Abdelfattah, A.; Cacciola, S.O.; Faedda, R.; Sanzani, S.M.; Cooke, D.E.L.; Schena, L. Metabarcoding analysis of *Phytophthora* diversity using genus-specific primers and 454 pyrosequencing. *Phytopathology* **2016**, *106*, 305–313. [CrossRef] [PubMed]
54. Junker, C.; Pfaff, A.; Werres, S. Validation of the bait test with rhododendron leaves for *Phytophthora ramorum*. *EPPO Bull.* **2018**, *48*, 595–608. [CrossRef]
55. Hüberli, D.; Hardy, G.E.S.J.; White, D.; Williams, N.; Burgess, T.I. Fishing for *Phytophthora* from Western Australia's waterways: A distribution and diversity survey. *Australas. Plant Pathol.* **2013**, *42*, 251–260. [CrossRef]
56. Oh, E.; Gryzenhout, M.; Wingfield, B.D.; Wingfield, M.J.; Burgess, T.I. Surveys of soil and water reveal a goldmine of *Phytophthora* diversity in South African natural ecosystems. *IMA Fungus* **2013**, *4*, 123–131. [CrossRef]
57. Jung, T.; Jung, M.H.; Cacciola, S.O.; Cech, T.; Bakonyi, J.; Seress, D.; Mosca, S.; Schena, L.; Seddaiu, S.; Pane, A.; et al. Multiple new cryptic pathogenic *Phytophthora* species from *Fagaceae* forests in Austria, Italy and Portugal. *IMA Fungus* **2017**, *8*, 219–244. [CrossRef]
58. Jung, T.; Jung, M.H.; Scanu, B.; Seress, D.; Kovács, G.M.; Maia, C.; Pérez-Sierra, A.; Chang, T.T.; Chandelier, A.; Heungens, K.; et al. Six new *Phytophthora* species from ITS Clade 7a including two sexually functional heterothallic hybrid species detected in natural ecosystems in Taiwan. *Persoonia Mol. Phylogeny Evol. Fungi* **2017**, *38*, 100–135. [CrossRef]
59. Jung, T.; Scanu, B.; Bakonyi, J.; Seress, D.; Kovács, G.M.; Durán, A.; Von Stowasser, E.S.; Schena, L.; Mosca, S.; Thu, P.Q.; et al. *Nothophytophthora* gen. nov., a new sister genus of *Phytophthora* from natural and semi-natural ecosystems. *Persoonia Mol. Phylogeny Evol. Fungi* **2017**, *39*, 143–174. [CrossRef]
60. Milenković, I.; Keča, N.; Karadžić, D.; Radulović, Z.; Nowakowska, J.A.; Oszako, T.; Sikora, K.; Corcobado, T.; Jung, T. Isolation and pathogenicity of *Phytophthora* species from poplar plantations in Serbia. *Forests* **2018**, *9*, 330. [CrossRef]
61. Mora-Sala, B.; Gramaje, D.; Abad-Campos, P.; Berbegal, M. Diversity of *Phytophthora* species associated with *Quercus ilex* L. in three Spanish regions evaluated by NGS. *Forests* **2019**, *10*, 979. [CrossRef]
62. Riddell, C.E.; Frederickson-Matika, D.; Armstrong, A.C.; Elliot, M.; Forster, J.; Hedley, P.E.; Morris, J.; Thorpe, P.; Cooke, D.E.L.; Pritchard, L.; et al. Metabarcoding reveals a high diversity of woody host-associated *Phytophthora* spp. in soils at public gardens and amenity woodlands in Britain. *PeerJ* **2019**, *7*, e6931. [CrossRef]
63. Vannini, A.; Bruni, N.; Tomassini, A.; Franceschini, S.; Vettraino, A.M. Pyrosequencing of environmental soil samples reveals biodiversity of the *Phytophthora* resident community in chestnut forests. *FEMS Microbiol. Ecol.* **2013**, *85*, 433–442. [CrossRef] [PubMed]
64. Català, S.; Pérez-Sierra, A.; Abad-Campos, P. The use of genus-specific amplicon pyrosequencing to assess *Phytophthora* species diversity using eDNA from soil and water in northern spain. *PLoS ONE* **2015**, *10*, e0119311. [CrossRef] [PubMed]
65. Scanu, B.; Linaldeddu, B.T.; Deidda, A.; Jung, T. Diversity of *Phytophthora* species from declining mediterranean maquis vegetation, including two new species, *Phytophthora crassamura* and *P. ornamentata* sp. nov. *PLoS ONE* **2015**, *10*, e0143234. [CrossRef]
66. Sims, L.L.; Sutton, W.; Reeser, P.; Hansen, E.M. The *Phytophthora* species assemblage and diversity in riparian alder ecosystems of Western Oregon, USA. *Mycologia* **2015**, *107*, 889–902. [CrossRef]
67. Dunstan, W.A.; Howard, K.; Hardy, G.E.S.J.; Burgess, T.I. An overview of Australia's *Phytophthora* species assemblage in natural ecosystems recovered from a survey in Victoria. *IMA Fungus* **2016**, *7*, 47–58. [CrossRef] [PubMed]
68. Pignatti, S.; Guarino, R.; La Rosa, M. (Eds.) *Flora d'Italia*, 2nd ed.; Edagricole: Milano, Italy, 2018; Volume 4.
69. Biondi, E.; Blasi, C.; Allegrezza, M.; Anzellotti, I.; Azzella, M.M.; Carli, E.; Casavecchia, S.; Copiz, R.; Del Vico, E.; Facioni, L.; et al. Plant communities of Italy: The vegetation prodrome. *Plant Biosyst.* **2014**, *148*, 728–814. [CrossRef]
70. Manuale Italiano di Interpretazione Degli Habitat Della Direttiva 92/43/CEE. Available online: http://vnr.unipg.it/habitat/ (accessed on 15 June 2017).
71. Jung, T.; Blaschke, H.; Neumann, P. Isolation, identification and pathogenicity of *Phytophthora* species from declining oak stands. *Eur. J. For. Pathol.* **1996**, *26*, 253–272. [CrossRef]

72. Jung, T.; Nechwatal, J.; Cooke, D.E.L.; Hartmann, G.; Blaschke, M.; Oßwald, W.F.; Duncan, J.M.; Delatour, C. *Phytophthora pseudosyringae* sp. nov., a new species causing root and collar rot of deciduous tree species in Europe. *Mycol. Res.* **2003**, *107*, 772–789. [CrossRef]
73. Cooke, D.E.; Drenth, A.; Duncan, J.M.; Wagels, G.; Brasier, C.M. A molecular phylogeny of *Phytophthora* and related oomycetes. *Fungal Genet. Biol.* **2000**, *30*, 17–32. [CrossRef]
74. White, T.J.; Bruns, T.; Lee, S.; Taylor, J.W. Amplification and direct sequencing of fungal ribosomal RNA genes for phylogenetics. In *PCR Protocols: A Guide to Methods and Applications*; Innis, M.A., Gelfand, D.H., Sninsky, J.J., White, T.J., Eds.; Academic Press Inc.: San Diego, CA, USA, 1990; Volume 18, pp. 315–322.
75. FinchTV v.1.4.0. Available online: https://digitalworldbiology.com/FinchTV (accessed on 18 May 2020).
76. BLAST Searches. Available online: https://blast.ncbi.nlm.nih.gov/Blast.cgi (accessed on 18 May 2020).
77. Phytophthora Database. Available online: http://www.phytophthoradb.org/ (accessed on 18 May 2020).
78. GenBank. Available online: https://www.ncbi.nlm.nih.gov/genbank/ (accessed on 18 May 2020).
79. R: The R Project for Statistical Computing. Available online: https://www.r-project.org/ (accessed on 18 May 2020).
80. Soil Science Division Staff. *Soil Survey Manual*; United States Department of Agriculture: Washington, DC, USA, 2017.
81. Meng, Y.; Zhang, Q.; Ding, W.; Shan, W. *Phytophthora parasitica*: A model oomycete plant pathogen. *Mycology* **2014**, *5*, 43–51. [CrossRef]
82. Hansen, E.; Delatour, C. *Phytophthora* species in oak forests of north-east France. *Ann. For. Sci.* **1999**, *56*, 539–547. [CrossRef]
83. Ruano-Rosa, D.; Schena, L.; Agosteo, G.E.; Lio, G.W.D.S.; Cacciola, S.O. *Phytophthora oleae* sp. nov. causing fruit rot of olive in southern Italy. *Plant Pathol.* **2018**, *67*, 1362–1373. [CrossRef]
84. González, M.; Pérez-Sierra, A.; Sánchez, M.E. *Phytophthora oleae*, a new root pathogen of wild olives. *Plant Pathol.* **2019**, *68*, 901–907. [CrossRef]
85. Santilli, E.; Riolo, M.; La Spada, F.; Pane, A.; Cacciola, S.O. First report of root rot of *Olea europaea* caused by *Phytophthora bilorbang* in Italy. *Plants* **2020**, *9*, 826. [CrossRef]
86. Scanu, B.; Hunter, G.C.; Linaldeddu, B.T.; Franceschini, A.; Maddau, L.; Jung, T.; Denman, S. A taxonomic re-evaluation reveals that *Phytophthora cinnamomi* and *P. cinnamomi* var. *parvispora* are separate species. *For. Pathol.* **2014**, *44*, 1–20.
87. Pane, A.; Faedda, R.; Cacciola, S.O.; Rizza, C.; Scibetta, S.; Lio, G.M.D.S. Root and basal stem rot of mandevillas caused by *Phytophthora* spp. in eastern Sicily. *Plant Dis.* **2010**, *94*, 1374. [CrossRef]
88. Cleary, M.; Ghasemkhani, M.; Blomquist, M.; Witzell, J. First report of *Phytophthora gonapodyides* causing stem canker on European beech (Fagus sylvatica) in Southern Sweden. *Plant Dis.* **2016**, *100*, 2174. [CrossRef]
89. Balci, Y.; Halmschlager, E. Incidence of *Phytophthora* species in oak forests in Austria and their possible involvement in oak decline. *For. Pathol.* **2003**, *33*, 157–174. [CrossRef]
90. Corcobado, T.; Cubera, E.; Pérez-Sierra, A.; Jung, T.; Solla, A. First report of *Phytophthora gonapodyides* involved in the decline of *Quercus ilex* in xeric conditions in Spain. *New Dis. Rep.* **2010**, *22*, 33. [CrossRef]
91. Erwin, D.C.; Ribeiro, O.K. *Phytophthora Diseases Worldwide*; American Phytopathological Society (APS Press): St. Paul, MN, USA, 1996.
92. Jung, T.; Burgess, T.I. Re-evaluation of *Phytophthora citricola* isolates from multiple woody hosts in Europe and North America reveals a new species, *Phytophthora plurivora* sp. nov. *Persoonia* **2009**, *22*, 95–110. [CrossRef]
93. Redondo, M.A.; Boberg, J.; Stenlid, J.; Oliva, J. Functional traits associated with the establishment of introduced *Phytophthora* spp. in Swedish forests. *J. Appl. Ecol.* **2018**, *55*, 1538–1552. [CrossRef]
94. Reeser, P.W.; Sutton, W.; Hansen, E.M.; Remigi, P.; Adams, G.C. *Phytophthora* species in forest streams in Oregon and Alaska. *Mycologia* **2011**, *103*, 22–35. [CrossRef]
95. Brasier, C.M.; Cooke, D.E.L.; Duncan, J.M.; Hansen, E.M. Multiple new phenotypic taxa from trees and riparian ecosystems in *Phytophthora gonapodyides-P. megasperma* ITS Clade 6, which tend to be high-temperature tolerant and either inbreeding or sterile. *Mycol. Res.* **2003**, *107*, 277–290. [CrossRef] [PubMed]
96. Yang, X.; Tyler, B.M.; Hong, C. An expanded phylogeny for the genus *Phytophthora*. *IMA Fungus* **2017**, *8*, 355–384. [CrossRef] [PubMed]
97. Aghighi, S.; Hardy, G.E.S.J.; Scott, J.K.; Burgess, T.I. *Phytophthora bilorbang* sp. nov., a new species associated with the decline of *Rubus anglocandicans* (European blackberry) in Western Australia. *Eur. J. Plant Pathol.* **2012**, *133*, 841–855. [CrossRef]

98. Nesbitt, H.J.; Malajczu, N.; Glenn, A.R. Effect of organic matter on the survival of *Phytophthora cinnamomi* rands in soil. *Soil Biol. Biochem.* **1978**, *11*, 133–136. [CrossRef]
99. Allmaras, R.R.; Kraft, J.M.; Miller, D.E. Effects of soil compaction and incorporated crop residue on root health. *Annu. Rev. Phytopathol.* **1988**, *26*, 219–243. [CrossRef]
100. Whalley, W.R.; Dumitru, E.; Dexter, A.R. Biological effects of soil compaction. *Soil Tillage Res.* **1995**, *35*, 53–68. [CrossRef]
101. Jung, T.; Blaschke, H.; Osswald, W. Involvement of soilborne *Phytophthora* species in Central European oak decline and the effect of site factors on the disease. *Plant Pathol.* **2000**, *49*, 706–718. [CrossRef]
102. Kong, P.; Lea-Cox, J.D.; Moorman, G.W.; Hong, C. Survival of *Phytophthora alni*, *Phytophthora kernoviae*, and *Phytophthora ramorum* in a simulated aquatic environment at different levels of pH. *FEMS Microbiol. Lett.* **2012**, *332*, 54–60. [CrossRef]
103. Corcobado, T.; Solla, A.; Madeira, M.A.; Moreno, G. Combined effects of soil properties and *Phytophthora cinnamomi* infections on *Quercus ilex* decline. *Plant Soil* **2013**, *373*, 403–413. [CrossRef]
104. Martin, D.A.N.; Rahmat, A. Relationship of soil physicochemical properties and existence of *Phytophthora* sp. in pineapple plantations. *Indones. J. Sci. Technol.* **2017**, *2*, 81–86. [CrossRef]
105. Cacciola, S.O.; Magnano di San Lio, G. Management of citrus diseases caused by *Phytophthora* spp. In *Integrated Management of Plant Diseases Caused by Fungi, Phytoplasma and Bacteria*, 1st ed.; Ciancio, A., Mukerji, K.G., Eds.; Springer Science + Business Media B.V.: Berlin, Germany, 2008; Volume 3, pp. 61–84.
106. Vettraino, A.M.; Morel, O.; Perlerou, C.; Robin, C.; Diamandis, S.; Vannini, A. Occurrence and distribution of *Phytophthora* species in European chestnut stands and their association with Ink Disease and crown decline. *Eur. J. Plant Pathol.* **2005**, *111*, 169–180. [CrossRef]
107. Yang, X.; Copes, W.E.; Hong, C. *Phytophthora mississippiae* sp. nov., a new species recovered from irrigation reservoirs at a plant nursery in Mississippi. *J. Plant Pathol. Microbiol.* **2013**, *4*, 5. [CrossRef]
108. Bose, T.; Wingfield, M.J.; Roux, J.; Vivas, M.; Burgess, T.I. Community composition and distribution of *Phytophthora* species across adjacent native and non-native forests of South Africa. *Fungal Ecol.* **2018**, *36*, 17–25. [CrossRef]
109. Hernández-Lambraño, R.E.; González-Moreno, P.; Sánchez-Agudo, J.Á. Environmental factors associated with the spatial distribution of invasive plant pathogens in the Iberian peninsula: The case of *Phytophthora cinnamomi* Rands. *For. Ecol. Manag.* **2018**, *419*, 101–109. [CrossRef]
110. Legeay, J.; Husson, C.; Boudier, B.; Louisanna, E.; Baraloto, C.; Schimann, H.; Marcais, B.; Buée, M. Surprising low diversity of the plant pathogen *Phytophthora* in Amazonian forests. *Environ. Microbiol.* **2020**. [CrossRef]