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Canker staindisease: a major threat to natural stands of oriental plane in Greece

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Summary
Ceratocystis platani is an invasive fungal pathogen in Greece that was initially found in Messenia prefecture in southwestern Peloponnese in 2003. Gradually the pathogen was spread into many areas of Peloponnese and in 2010 it was reported in Epirus (northwestern Greece), while in 2011 it was detected in Thessaly (central Greece). This fungus infects only *Platanus* spp. and causes a destructive disease, known as canker stain disease of plane trees. The disease is fatal, since there is no cure for infected trees. The pathogen is spreading mainly due to human activities. The use of terracing machinery and cutting tools are two major pathways for *C. platani* tospread over short and long distances. The disease is killing trees of all sizes in town squares and parks, but the largest problem is in natural stands of oriental plane along rivers and streams. The pathogen has a devastating effect in the riparian vegetation in Peloponnese and Epirus, with thousands of dead trees. It is very important to apply phytosanitary measures for disease containment in order toavoid further spread of the pathogen into new areas of the country.

Key words*:Ceratocystisplatani*, *Platanus orientalis*,** disease spread

Introduction
Canker stain, caused by *Ceratocystisplatani* (Walter) Engelbrecht et Harrington (syn.:*Ceratocystisfimbriata* Elis & Halstead f.sp.*platani* Walter), is a very destructive disease that attacks *Platanus* spp. in North America and Europe. The fungus is of American origin and was introduced to Europe (Italy and France) during World War II, possibly with infected plane wood used to package ammunitions or other military equipment.The pathogen has spread throughout Italy and southern France and was also reported in Switzerland, where it has been eradicated. There are also unconfirmed reports on the presence of *C. platani* in Belgium, Spain and Armenia (Panconesi 1999).
Canker stain was first observed in Greece in 2003 in Messenia prefecture(southwestern Peloponnese) in natural stands of oriental plane (*Platanus orientalis* L.) as well as in ornamental plantings (Tsopelas & Angelopoulos 2004). It has been suggested that *C. platani* was introduced into Greece with infected planting stock of London plane from Italy (Ocasio-Morales *et al*. 2007). Since then, the pathogen has spread in many areas of Peloponnese, Epirus and Thessaly, mainly in natural stands of oriental plane.
Oriental plane is indigenous in south-eastern Europe, extending eastward to Asia Minor, Iran and central Asia. In Greece, this tree species occurs in many regions, mainly on moist sites along streams and rivers, being one of the dominant species of the riparian vegetation. It is also commonly planted as an ornamental tree, being a characteristic feature of town squares and recreation areas, especially in areas with

natural springs. Oriental plane is afast-growing and long living tree which may attain large dimensions. It is among the largest tree species in the country.In many areas of Greece there are centuries-old plane trees which are of historic value, and some of them have been declared as "Protected Monuments of Nature". In this report the spread of the fungus *C. platani* is examined in natural stands and ornamental plants of oriental plane along with the threat that it poses to this valuable and ecologically important tree species in Greece.

Biology of tree infection

*Ceratocystisplatani*isanascomycete (family: Ophiostomataceae). It sporulates on infected woodand bark, forming both sexual and asexual spores on surfaces of pruning cuts or broken branches, as well as in cracks under the bark in the region of the canker. Spores are also formed on the sawdust produced from infected trees during felling operations. Perithecia are globoseand ascospores accumulate at the tip of the neck as a white creamy mucilaginous mass, having a characteristic bowler's hat shape. The fungusalso forms three different types of asexual spores: cylindrical endoconidia, doliformendoconidia and thick-walled aleurioconidia (chlamydospores). Aleurioconidia are also formed inside the xylem vessels and can remain viable in the dead wood for long periods of time. These thick-walled spores can also survive in river water and soil and play a significant role in pathogen dispersal (Grosclaude*et al.* 1991, Engelbrecht*et al.* 2004).

Ceratocystisplatani is primarily a wound parasite, infecting plane treesmainly through wounds in the branches, the trunk or the roots. Mycelia of the fungus colonize the phloem and the sapwood and are spread into the vessels. The fungus advances longitudinally upwards and downwards and also grows along the periphery of the tree as well as radially in the medullary rays. In this way the mycelia are able to spread into adjacent branches and/or roots of the tree.Panconesi (1999) reported that the fungus can spread longitudinally up to 2 m per year on London plane (*Platanus* X *acerifolia*) trees. On oriental plane, we have measured longitudinal growth of the fungus exceeding 7 m, only 6 months after infection (Tsopelas unpublished data).Subsequent spread of the pathogen, once introduced into a new site, occurs through root grafts from infected trees into healthy adjacent ones.

The fungus causes necrosis of the cambium and inner bark forming cankers. In oriental planes cankers are not externally visible due to the thick bark, but cankers can be visible after bark removal. A characteristic staining of the sapwood, dark brown to bluish black, is very obvious on infected trunks and branches after bark removal. Very often this staining of the sapwood forms flame-shape patterns. These symptoms are obvious on infected trees that are still alive, but cannot be observed on trees that have been dead for a long time (Ocasio-Morales*et al.* 2007).

Infected trees may show symptoms of chlorosis and microphyllia over the entire crown, but very often one or more branches on one side of a tree die, while the rest of the crown is still green with or without symptoms. Finally, the whole tree dies as a result of the infection, since the disease is fatal. Small trees may die within one season, while larger trees usually die one to two years after infection.

Pathways of pathogen spread

Human activities play a significant role in disease spread into new areas.Cutting tools and terracing machinery seem to be two major pathways of pathogen dispersal over short and long distances. Fungal spores can survive for many days on cutting tools and machinery, especially when sawdust or pieces of infected wood are present. Cutting tools (chainsaws, axes etc.) are often used on diseased trees and then on healthy trees spreading the diseasein this way. Also, terracing machinery used in

infected areas may carry soil and pieces of infected wood into new areas; such machinery may cause damage to the roots of plane trees, starting new infections (EPPO 1997, Ocasio-Morales *et al.* 2007).
In the riparian vegetation *C. platani* may spread readily through running water. Dead logs and pieces of branches from diseased trees are carried downstream by the water, creating new infection foci. The thick walled aleurioconidia of the fungus can survive in stream water and cause new infections (Grosclaude *et al.* 1991). Once an infection focus is established, the fungus may spread to neighbouring trees through root grafting since oriental plane trees grow close together along rivers and streams.
Wood infected by *C. platani* has a fruity odor that attracts various kinds of insects. Beetles of the family Nitidulidae have been reported to transmit *C. platani* in the USA (Crone 1962). In recent studies in Greece, the ambrosia beetle *Platypus cylindrus* F. (Platypodidae) has been found to vector *C. platani* (Soulioti, unpublished data) and the pathogen was also isolated from frass of this insect (Ocasio-Morales *et al.* 2007). Certain other insects have also been found associated with diseased trees in Greece, but their role remains uncertain in disease spread. It is not known if ambrosia beetles play an important role as vectors of this disease, but certainly they should be taken into consideration, especially during management operations, since these insects can attack freshly wounded, healthy trees and possibly transmit the disease. Also, the frass produced by the ambrosia beetles can be easily dispersed by wind or river water, and it would be infective if it landed on the wound of a healthy tree.

Disease dispersal in Greece
When *C. platani* was detected for first time in 2003, the disease seemed to be limited to certain areas of the Messenia prefecture. In the next years the pathogen was found into the neighbouring prefectures of Ileia and Arcadia and gradually was dispersed all over Peloponnese. By 2012 the disease was spread to almost every prefecture of Peloponnese except Argolis (Argolida). In 2010 *C. platani* was reported in the prefectures of Thesprotia and Ioannina of the Epirus region (northwestern Greece). Most likely, inoculum of *C. platani* was transferred to Epirus from Peloponnese with terracing machinery that was used in the construction of the new highway "Egnatia" or in some other construction site (Tsopelas and Soulioti 2010). In 2011 the disease was detected in Thessaly, in central Greece, where it was introduced via contaminated cutting tools.
The situation is alarming in Greece; *C. platani* is spreading in natural stands of *P. orientalis*, which appears to be a very susceptible host. This is the first time that the disease has caused extensive damage to natural ecosystems of oriental plane worldwide. Thousands of trees have been killed by the disease in many areas of Peloponnese and Epirus. In Peloponnese, the pathogen has been spread in the rivers Alpheios, Ladon and Neda and has affected many smaller rivers and streams. In Epirus, *C. platani* has been found in some of the major rivers, such as Kalamas, Acheron and Louros. Up to now the pathogen has caused devastating effects on the Kalamas River, where the disease has reached unmanageable levels.
The fungus appears to spread rapidly along river courses causing extensive mortality to riparian oriental plane trees. Along some of the rivers and streams of these regions, the disease has been spread for many kilometres. Plane trees are found dying in groups of 20-50 trees, and in certain localities, where disease foci have been fused, hundreds of dead trees can be observed on a single site.
Infections by *C. platani* have also been reported in many residential areas, parks and recreation sites. The pathogen has killed trees of varying ages and sizes; some of these trees were centuries old and had large dimensions. The aesthetic value of some of

these trees was great, since they were predominant in town squares and parks and loss of such trees is hugely important.

Control measures

In some of the areas of Peloponnese and Epirus the disease appears to have reached unmanageable levels, however, it is important to apply phytosanitary measures of disease containment in order to avoid further spread of the pathogen into new areas of the country. A specific disease management plan has to be in place providing full details of all the steps that should be taken and the administrative procedures that should be followed in dealing with the disease.

Demarcated zones should be determined and specific phytosanitary measures should be enforced in order to avoid spread of the disease by human activities. The use of terracing machinery in these areas should be avoided. Vehicles and other machinery that have been used in infected sites must be thoroughly cleaned with the use of pressure washers; the ones using hot water or steam are more appropriate and they should then be sprayed with a disinfectant. Water run-off must not enter waterways. There is also a need for disinfection of machinery before use in areas with plane trees. All hand tools used on infected sites must be disinfected before leaving an infected site. Also, tool disinfection must always be practiced before pruning or felling plane trees (EPPO 1997, Ocasio-Morales *et al.* 2007).

A key factor in applying effective control measures is the early detection of new disease foci. Active surveillance all over the country is needed. In areas of limited infestation, the application of disease suppression treatments will be more effective and the cost relatively low. Eradication measures can be applied when the disease is detected at the initial stages. These control measures are very difficult to apply when the disease has taken epidemic proportions in forest ecosystems. Felling of diseased trees along rivers and streams which have many infected trees can create greater problems by further spreading the disease with the debris and the sawdust created.

In applying erradication measures, all living infected trees and their neighbouring healthy trees in a radius of 20-30m must be injected and killed with herbicides in order to avoid spread through root grafting (Grosclaude *et al.* 1992, Tsopelas unpublished data). All the trees treated with herbicide must be felled and the wood, as well as all the debris and sawdust, should be destroyed by fire or properly buried in sanitary landfills. The felling site, including the debris and sawdust left, should be sprayed with a fungicide (Panconesi 1999). Movement of infected material, such as firewood, into disease-free areas greatly increases the chance of introducing the pathogen into those areas and should be avoided.

A resistant clone of a hybrid plane (*P. orientalis* X *P. occidentalis*) was selected in a study in France that is resistant to *C. platani* and is commercially available (Vigouroux and Olivier 2004). Resistant plane trees can be used in infected areas; however, these plants cannot replace oriental plane trees in natural stands nor the magnificent trees that dominate in town squares or those that have been declared as Protected Monuments of Nature, living legends of the history of this country.

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