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PLANT HEALTH CARE FEBRUARY 1, 2024 0

What Lurks Within? Endophytes' Potential Control of Tree Diseases

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TCI Magazine Audio Edition

What Lurks Wit

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The importance of mycorrhizae and the role they play in enhancing tree health is widely recognized by arborists. However, the role of endophytes, i.e., bacteria and fungi that live within a tree without causing any disease

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Photo 1: Shumard oak with oak-wilt disease. Photo courtesy of the Bartlett Tree Research Media Library.

Endophytes are bacteria and fungi that live within healthy tree tissue (sapwood, leaves, roots, stem, bark, buds) without causing any visible symptoms of disease. Of all the world's plants, only a few grass species have had their complete complement of endophytes studied. In contrast,

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tree species. Within oaks (*Quercus*), species such as *Penicillium*, *Trichoderma*, *Botryosphaeria*, *Cladosporium*, *Coryneum*, *Cytospora*, *Discula* and *Stereum* tend to dominate. Whereas within ash (*Fraxinus*), *Aureobasidium*, *Phoma*, *Dothideomycetes*, *Diaporthe* and *Lophiostoma* species are more prevalent.

Role of endophytes – growth

Endophytes play an important role in helping trees uptake essential nutrients from the soil. They help tree roots absorb phosphorous in phosphorous-deficient soils and can aid in the uptake of nitrogen in soils of low fertility, as well as allow trees to establish on sites contaminated with heavy metals. In addition, endophytes produce a range of hormones, such as auxins, cytokinins and gibberellic acids, that have essential roles in tree growth, seed and flower set, senescence (biological aging) and dormancy.



Burkholderia vietnamiensis, for example, is an endophytic bacterium isolated from poplar trees that produces the hormone auxin, responsible for root formation. This, in turn, has been shown to stimulate root growth following construction damage, helping a poplar tree survive. Endophytes also produce or stimulate the production of chemicals in leaves that protect trees from drought, heavy-metal toxicity, heat waves and high light intensity, problems trees routinely encounter in urban landscapes.

Disease resistance

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- As a direct biological-control agent against a specific disease or pest.
- By the production of chemical compounds that inhibit fungal and bacterial growth.

Endophytes as direct bio-control agents

It has been shown that an endophytic fungus living in the inner bark of elm trees reduced the movement of a bark beetle spreading Dutch elm disease. Another example is the endophytic fungi *Deadelea quercina*, which invades the galls of a cynipid insect and kills the wasp inside them. The endophytic fungus *Beauveria bassiana* has been found to control borer insects in coffee, and is now available as a commercial product (Naturalis), widely used for the management of thrips, mites, white fly and aphids (egg, larvae, adult).

In Western white pine (*Pinus monticola*), fungal endophytes reduced damage by white pine blister rust (*Cronartium ribicola*). Seedlings colonized with fungal endophytes had greater survival rates than control seedlings, with the levels of resistance observed the same as white pine trees selected in breeding programs for resistance against this disease. When a mixture of six different endophytes isolated from cacao (*Theobroma cacao* L.) trees was used to inoculate leaves of endophyte-free seedlings of the same plant species, the severity of *Phytophthora* infection on leaves was significantly reduced.

The endophyte *Trichoderma citrinoviride*, isolated from cork oak, was shown to possess a strong biological activity against the main pathogens causing oak decline within Mediterranean climates. Recent work at the

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Chemicals produced by endophytes

Many endophytes produce substances that can inhibit the growth of several disease-causing fungi. When produced by endophytes in or on the tree, these substances can constitute a powerful defense mechanism.

Compounds produced by endophytic microbes include Taxol (anticancer drug), Cryptocin (antifungal agent), Jesterone (antifungal agent), Oocydin (antifungal agent) and Ambuic acid (antifungal agent).

Of particular interest is the cancer-fighting drug Taxol, which has shown to be produced by 18 different fungi, primarily endophytes that live within *Taxus* (yew) and other trees. As Taxol is a fungicide, and the endophyte(s) producing it is (are) resistant to Taxol, this endophyte strategy has been used to prevent fungal attack by pathogens such as *Perenniporia subacida* (poroid fungus of Douglas fir), *Phaeolus schweinitzii* (velvet-top fungus of fir, spruce and hemlock, pine and larch) and *Heterobasidion annosum* (a decay fungi of conifer trees) at minimal “metabolic” cost to the tree.

Liquid cultures of *Trichoderma citrinoviride*, derived from cork oak, produced a mixture of antibiotics that demonstrated antifungal activity against seven forest-tree pathogens to include *Apiognomonia quercina* (oak anthracnose), *Botryosphaeria corticola*, *B. parva*, *B. obtuse* (canker diseases of oak), *Biscognauxia mediterranea* (charcoal canker of hardwood trees) and *Diplodia pinea* and *D. scrobiculata* (tip blights of pine).

Importance of endophytes in the U.S.

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Photo 2: Veteran English oak in the United Kingdom. Photo courtesy of the author.

grand fir. Spruce beetle, emerald ash borer and sudden oak death were also important sources of tree mortality throughout U.S. forests in 2018.

In addition to tree death, defoliating pests damage trees by eating leaves or needles, causing significant losses of foliage that in turn detrimentally impact forest health. While a “one-off” defoliation usually does not result in tree death, repeated attacks in combination with heat waves and drought, for example, can cause trees that would normally recover to succumb to these defoliating

insects.

Case scenerio 1

Oak wilt (*Ceratocystis fagacearum*) is a highly destructive fungus that causes wilt in oak trees in the Eastern and Midwestern U.S. (Photo 1) Extensive damage occurs particularly to red-oak species, while damage to white oaks tends to be less severe. Presently, there is no cure for oak-

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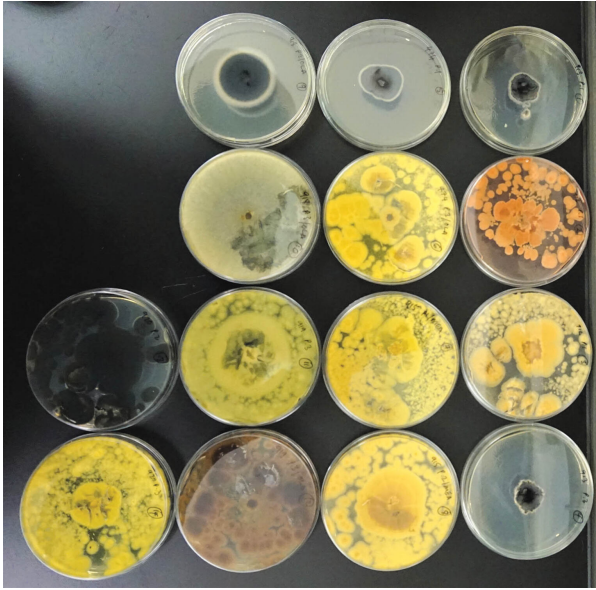


Photo 3: Endophytes growing in agar. Photo courtesy of the author.

Research from Texas A&M University isolated endophytic bacteria from several surviving live oaks (*Quercus fusiformis*) where oak wilt is epidemic, and evaluated their potential as biological control agents for oak wilt. Under laboratory conditions, 183 of the bacterial endophytes isolated showed inhibition of oak wilt. The six most

powerful endophytes were then further evaluated using containerized Spanish (*Quercus texana*) and live oaks.

Trees that were pre-inoculated with the bacterial endophyte *Pseudomonas denitrificans* (stem injection) were then inoculated with the oak-wilt pathogen. These tests showed a 50% reduction in the number of diseased trees and a 17% decrease in crown loss. In a second separate trial, pre-inoculation with either *P. denitrificans* or *P. putida* also significantly reduced crown loss caused by oak wilt.

These studies show that there is a potential for the use of endophytic bacteria to control oak wilt.

Case scenerio 2

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resistance to acute oak decline (AOD) was studied. (Photo 2) The trial site was a mature English-oak woodland in Writtle Forest, in Essex, where AOD is resulting in the death and decline of many oaks.

Endophytic fungi and bacteria from asymptomatic (showing no symptoms of AOD) and symptomatic (showing symptoms of AOD, e.g., bleeding trunk lesions) trees were isolated from trees located within the same area of woodland. This was achieved by removing cores of wood from both tree types, then growing fungi and bacteria from these cores on a number of different agar growth mediums. DNA was then extracted from any fungi that grew, followed by DNA sequencing.



Photo 4: Asymptomatic oaks side by side in Writtle Forest in Essex, England. Photo courtesy of the author.

Within asymptomatic oak trees, several species of *Penicillium* fungi were identified, including *Penicillium aeneum*, *P.citreosulfuratum*, *P.miczyskii*, *P.brevicompactum*, *P.aurantiacobrunneum* and *P.manginii*, as well as a number of other unidentified *Penicillium* species. (Photo 3) The benefits of penicillin as an antibiotic to treat bacterial diseases of humans, such as

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Brenneria goodwinii and *Gibbsiella quercinecans*, two of the bacteria responsible for AOD.

Further work

In further work, one *Bacillus* and three *Pseudomonas* bacterial strains were shown to inhibit growth of *Armillaria*, a root disease, by up to 40-50%. Combining the two bacteria resulted in 65% inhibition. Such a result proves that oak trees showing no symptoms of AOD contain within their tissue fungal and bacterial endophytes that confer resistance against both AOD and *Armillaria*.

Future research

While resistance to tree diseases is generally considered a genetic trait, there is now a growing body of evidence showing that the influence of endophytes within a tree may have been underestimated. (Photo 4) Ongoing research at the Bartlett Tree Research Laboratory aims to elucidate whether a specific endophytic profile exists between susceptible and resilient oaks to AOD. If such a profile does exist, then this can be used to identify resilient and susceptible oaks.

There also would be the potential to apply endophytes with known bio-control properties into oaks that are lacking these endophytes to enhance resistance. Developments in molecular diagnostic technology now make this type of research a very real possibility that could prove of importance in developing new strategies to protect trees. Importantly, no genetic manipulation or chemicals are being used in this management

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