

EPPO Datasheet: *Bretziella fagacearum*

Last updated: 2021-03-25

IDENTITY

Preferred name: *Bretziella fagacearum*

Authority: (Bretz) Z.W. de Beer, Marincowitz, T.A. Duong & M.J. Wingfield

Taxonomic position: Fungi: Ascomycota: Pezizomycotina:

Sordariomycetes: Hypocreomycetidae: Microascales:

Ceratocystidaceae

Other scientific names: *Ceratocystis fagacearum* (Bretz) J. Hunt,

Chalara quercina B.W. Henry, *Endoconidiophora fagacearum*

Bretz, *Thielaviopsis quercina* (B.W. Henry) A.E. Paulin, T.C.

Harrington & McNew

Common names: oak wilt, wilt of oak

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EPPO Categorization: A1 list

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EU Categorization: Quarantine pest ((EU) 2019/2072 Annex II A)

EPPO Code: CERAFA



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Notes on taxonomy and nomenclature

Phylogenetic analyses supported the establishment of the monophyletic genus *Bretziella* in the lineage of Ceratocystidaceae to accommodate the oak wilt fungus due to its distinct differences from other genera in the family (de Beer *et al.*, 2017). Formation of sporulating mats as mirror image structures on outer sapwood and inner phloem with pressure cushions required to open the bark is unique to this species among the species of Ceratocystidaceae; thus, further support these changes in taxonomy and nomenclature.

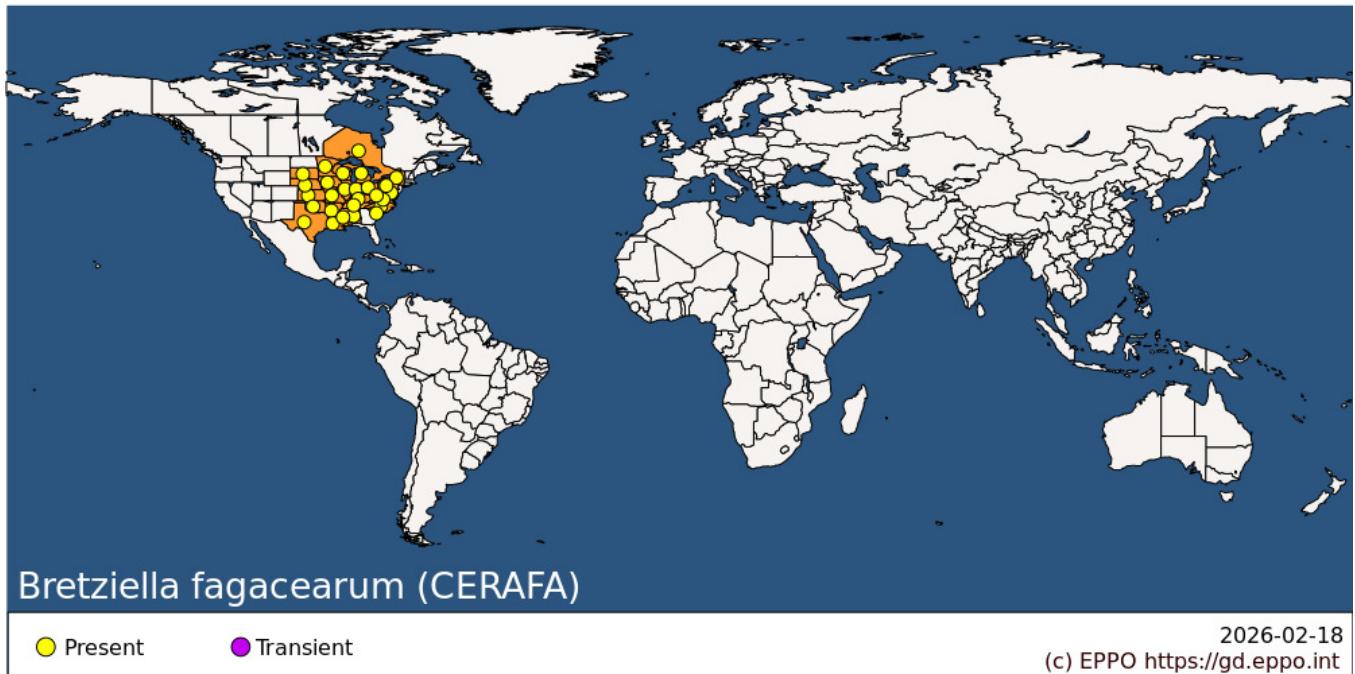
HOSTS

B. fagacearum infects *Quercus* spp. (more than 30 species) and several species in other genera of the Fagaceae (e.g. *Castanea mollissima*, *Lithocarpus densiflora*) based on documented natural infections and/or results of artificial inoculation studies. No North American oak is known to be immune. Red oaks (subgenus *Lobatae*) are highly susceptible and may die in as few as 4 to 6 weeks or early in the subsequent growing season. American white oaks (subgenus *Quercus*) are more resistant to disease development than red oaks. Moderately resistant oak species die over a period of 2 to 4 years several years while a highly resistant species (*Q. alba*) may die over many years or recover. To assess the susceptibility of European oaks, hundreds of European white oaks were artificially inoculated with *B. fagacearum* in West Virginia and South Carolina (MacDonald *et al.*, 2001). All individual oaks appeared susceptible regardless of the species (*Q. robur*, *Q. petraea*, *Q. pubescens*) and died within the year following inoculation. Foliar symptoms and subsequent tree death were equally observed after branch or main stem inoculations. Root graft transmission of the fungus to nearby trees also became obvious within a few weeks and led to the death of these trees during the subsequent year (MacDonald *et al.*, 2001; Pinon *et al.*, 2003). *B. fagacearum*-caused wilt and mortality has occurred in plantations of *C. mollissima* (Missouri and Ohio USA).

Host list: *Castanea hybrids*, *Castanea mollissima*, *Castanea pumila*, *Castanea sativa*, *Chrysolepis sempervirens*, *Notholithocarpus densiflorus*, *Quercus agrifolia*, *Quercus alba*, *Quercus chrysolepis*, *Quercus coccinea*, *Quercus dumosa*, *Quercus ellipsoidalis*, *Quercus engelmannii*, *Quercus falcata*, *Quercus fusiformis*, *Quercus garryana*, *Quercus ilex*, *Quercus imbricaria*, *Quercus kelloggii*, *Quercus laevis*, *Quercus laurifolia*, *Quercus lobata*, *Quercus macrocarpa*, *Quercus marilandica*, *Quercus muehlenbergii*, *Quercus nigra*, *Quercus palustris*, *Quercus petraea*, *Quercus phellos*, *Quercus prinus*, *Quercus pubescens*, *Quercus robur*, *Quercus rubra*, *Quercus shumardii*, *Quercus stellata*, *Quercus suber*, *Quercus texana*, *Quercus velutina*, *Quercus virginiana*, *Quercus wislizenii*

GEOGRAPHICAL DISTRIBUTION

The origin of *B. fagacearum* is unknown. Evidence for its status as either a native or an introduced pathogen have been considered (Juzwik *et al.*, 2008). For many years, the fungus was known only from a number of states in the eastern United States, but in June 2023 it was found for the first time in Ontario (Canada) where official control measures were taken (NAPPO, 2023). At present, the fungus has not spread to other continents.



North America: Canada (Ontario), United States of America (Alabama, Arkansas, Illinois, Indiana, Iowa, Kansas, Kentucky, Louisiana, Maryland, Michigan, Minnesota, Mississippi, Missouri, Nebraska, New York, North Carolina, Ohio, Oklahoma, Pennsylvania, South Carolina, South Dakota, Tennessee, Texas, Virginia, West Virginia, Wisconsin)

BIOLOGY

B. fagacearum is a classic vascular wilt pathogen. On red oak species (*Quercus* Section *Lobatae*), the fungus is confined to the vessels of the outermost xylem ring of the tree until it becomes moribund. Passive movement of spores within the transpiration stream usually results in the fungus being transported to all parts of a red oak. Upon complete crown wilt, the fungus hyphae grow radially outward to colonize the outer sapwood and inner bark and radially inward to colonize deeper sapwood. Sporulating mats commonly form in the cambium region of red oaks. Rectangular-shaped asexual spores are produced by endoconidiophores on the mycelial mat. Dense, sterile pads also form on the mats and the pressure exerted by opposing pads on well-developed mats force the bark to crack open. Volatiles produced by the fungus attract insects that access the mats through the ruptured bark. Sexual reproduction of the fungus is facilitated by insects that carry conidia of the opposite mating type. Sexual spores are exuded in a gelatinous matrix from the resulting perithecia. Mat production may be limited by high summer temperatures and colonization by competing fungi. Once formed, mat deterioration is hastened by higher temperatures, colonization by other microorganisms, and activity of larvae from insects utilizing mats for brood development. The pathogen usually dies in the above-ground parts of a colonized tree within one year of wilting and is quickly replaced by saprobes. Survival in the root system may be more prolonged, especially if the roots are grafted to those of neighboring trees.

Internal spread of the pathogen in water-conducting xylem of American white oak species is more restricted than in red oaks. Xylem vessel anatomy (e.g. smaller diameters, thicker walls) and natural propensity of some white oak species to produce tyloses (balloon-like structures that plug vessels) contribute to limited pathogen spread. Scattered

branch wilt in white oaks (e.g. *Q. macrocarpa*, *Q. fusiformis*, *Q. virginiana*) that exhibit moderate resistance leads to tree death over several years while 'highly' resistant species (e.g. *Q. alba*) exhibit symptoms typical of slow crown decline that occurs over many years. In the latter case, infected annual rings can be buried under new xylem and are considered an unlikely source of inoculum. Furthermore, sporulating mats rarely form on such species. Interestingly, European white oaks (*Q. robur*, *Q. petraea* and *Q. pubescens*) were found to be as susceptible to oak wilt development as North American red oak species. Reasons for this difference have not been investigated. For a review of many aspects of disease biology, see Juzwik *et al.* (2011) and Appel (1995).

The most common means of pathogen spread among trees occurs below-ground. In many parts of its range, the fungus spreads via functional unions of grafted roots. Root grafting is known to occur in numerous oak species and is also common in oak forests in Europe. In the USA, frequency of inter-tree root grafting varies by oak species, site factors (e.g. soil texture, topography) and geographic region (Juzwik, 2009). In Wisconsin, the highest frequencies (over 70%) of intra-specific grafting occurs in *Q. ellipsoidalis* and the lowest (5%) in *Q. macrocarpa* (Parmeter *et al.*, 1956). Spores move passively in the transpiration stream of the xylem from wilted oaks to actively transpiring, healthy oaks leading to enlargement of oak wilt centres. Highest rates of annual radial expansion range from 7.6 m/year on deep sandy soils in Minnesota to 12 m/year on sandy soils in Michigan (Bruhn *et al.*, 1991). The clonal production of *Q. fusiformis* in Texas results in shared root systems that can extend over multiple hectares. Following pathogen introduction, its rapid spread through roots leads to disease centre expansions of 11-16 m/year (Appel *et al.*, 1989).

Above-ground spread of the pathogen from diseased to healthy trees over short to long distances is mediated by insect vectors. Insect transmission is the way new oak wilt foci are established in the USA. Nitidulid beetles and oak bark beetles (*Pseudopityophthorus* species – Curculionidae: Scolytinae) are considered the most important vectors; however, their relative importance varies by geographic region (Juzwik, 2009). Between May and July in Minnesota, *B. fagacearum*-contaminated *Colopterus truncatus* and *Carpophilus sayi* (Nitidulidae) were found on 24 to 33% and 4 to 79% of the species, respectively, visiting fresh wounds on healthy oaks (Juzwik *et al.*, 2004). These vectors were found to acquire viable pathogen spores from fungal mats in an earlier study (Juzwik and French, 1983). The frequency of *B. fagacearum* spore dissemination by oak bark beetles (*P. minutissimus* and *P. pruinosus*) is quite variable. Although up to 30% of beetles emerging from wilted red oaks may carry viable spores, a more typical beetle contamination rate is between 0.4 and 2.5%. The frequencies of fungus-contaminated, free-flying (= dispersing) *P. minutissimus* in oak wilt centres in Minnesota during May ranged from 0.4 to 1.3% (Ambourn *et al.*, 2006). The oak bark beetle *Scolytus intricatus* is widespread in Europe and is considered a potential vector of *B. fagacearum* (Donganlar and Schopf, 1984; Yates, 1984). This species is known to feed on many *Quercus* species and is known to transmit two fungal pathogens (*Ophiostoma roboris* and *Ceratocystis piceae*) associated with oak decline in Europe (Eisenhauer, 1989; Srutka, 1996).

DETECTION AND IDENTIFICATION

Symptoms

In red oaks, the first appearance of foliar wilt may occur anytime from mid-spring to late summer. Early season wilt is indicative of previous season infection of the tree via root-graft spread. Early to late-summer wilting results from current season infections associated with insect transmission to wounds or via root grafts. Individual leaves exhibit bronzing or yellowing beginning at leaf margins and apex and progressing to mid-rib and leaf base. Such leaves are often cast and found on the ground near a wilting tree, although some turn brown and remain on a completely wilted tree.

In white oak species, leaf symptoms are more variable. Reddish-brown to brown discoloration start at the leaf margin and apex but are often limited to one side of the mid-rib. Veinal chlorosis and necrosis of the evergreen live oak *Q. fusiformis* are unique and diagnostic (Appel, 1986). Diseased live oaks (*Q. fusiformis* and *Q. virginiana*) in Texas usually die within 3 to 8 months after infection.

Vascular staining in the xylem of branches and main stem of moribund oaks is a less reliable symptom compared to the foliage. Diffuse, bluish-grey to dark-brown discoloration in the outer sapwood occurs in red oaks. More pronounced dark brown to blackish streaks in the outer xylem are found when bark is removed from branches and stems of at least some white oak species. Discoloration is also often evident in red and white oak species in cross-

sections and may appear as discrete brown spots or as coalescing brown spots in a ring in the outermost annual growth increment of recently infected trees. In white oak species, a new uninfected growing increment may be produced leaving discrete stained spots “buried” in older growth increments.

Sporulating mats are the definitive sign of the oak wilt pathogen in recently wilted trees. These fruity-smelling mats occur most commonly on red oak species and may be visually detected on vertical bark cracks several months after complete tree wilt. Mats are seldom to never formed on species in the white oak group.

Morphology

In culture, *B. fagacearum* produces rectangular-shaped (4-9 x 2-3 μm) endoconidia on endoconidiophores when grown on 2% yeast malt, lactic acid-amended potato dextrose agar, or oak wilt identification agar (per Barnett, 1953). Mycelia are fluffy, pale to dark grey on 2% yeast malt agar and emit a characteristic fruity odour. With mated (A and B types) cultures, perithecia appear in 7-10 days and are black flask-shaped structures with a spheroidal base and an erect beak (250-450 μm long). Ascospores exuded in a sticky creamy-white mass from the tip are hyaline, one celled, and elliptical in shape (5-10 x 2-3 μm in size). For further information, see DeBeer *et al.* (2017).

Detection and inspection methods

B. fagacearum can be isolated from xylem chips taken from outermost sapwood of branches showing active disease symptoms and plated on agar media (e.g. malt, acidified potato dextrose agar) (Pokorny, 1999). Isolation is also possible from the outermost xylem annual ring of wedge samples cut from main stem of recently wilted red oak species in late autumn through the subsequent spring. Isolation is most successful from chips obtained from sapwood with characteristic vascular staining. Isolation on oak wilt identification agar (Barnett, 1953) is useful for tests of logs sampled in winter. Viable propagules of the pathogen on insect vectors can be detected by plating sonicated and diluted suspensions on agar media. The latter protocol was used to determine peak dispersal periods of two nitidulid beetle vector species in Minnesota (Ambourn *et al.*, 2005) and during the development of a degree day model for emergence of the same insect species carrying *B. fagacearum* in Wisconsin (Jagemann *et al.*, 2018).

DNA of *B. fagacearum* can be detected in wood and on insect vectors using molecular methods. A nested PCR technique (Wu *et al.*, 2011) developed for phytosanitary inspection purposes (Wu *et al.*, 2011) was modified for use by diagnostic plant disease laboratories (Yang and Juzwik, 2017). The method was also used to evaluate presence of viable *B. fagacearum* in harvested oak logs before and after heat or chemical fumigation treatment (Juzwik *et al.*, 2019; Yang *et al.*, 2019). Real-time PCR protocols utilizing rDNA ITS region have also been used to detect the fungus in wood (Wu *et al.*, 2011; Yang and Juzwik, 2017). Specific TaqMan real-time PCR detection assays developed for multiple target pathogens demonstrated rapid and reliable detection of *B. fagacearum* in sapwood samples and on nitidulid beetles obtained from oak wilt mats (Lamarche *et al.*, 2015).

B. fagacearum isolated from infected wood tissue can be identified in culture based on specific morphological characteristics following the EPPO diagnostic protocols for regulated pests PM 7/1 (EPPO, 2001) which is currently under revision, and the revised version will include molecular tests.

PATHWAYS FOR MOVEMENT

As noted under Biology, *B. fagacearum* normally spreads slowly (over one to several years) between a diseased and a nearby healthy tree whose root systems are connected by grafts. However, in central and northern US stands with a high red oak component or in Texas with clonally propagated live oak, expansion of disease centres often occur outwardly in a radial fashion leading to large pockets of dead trees over time. Insect transmission over short distances can lead to multiple centers in a forested land parcel that may coalesce. Wind-directed spread of dispersing, pathogen-laden insect vectors may lead to new disease centers in disjunct land parcels up to 600 m (Shelstad *et al.*, 1991) or potentially greater distances (Bowen and Merrill, 1982) from the nearest existing centre. In the USA, human-aided spread of diseased oak material with sporulating mats infested with insect vectors is suspected to be the pathway implicated for several major extensions of the oak wilt range, e.g. in north central states and in New York (Jensen-Tracy *et al.*, 2009).

Over long distances, the main pathways for introducing *B. fagacearum* into new areas are international movements

of wood (with and without bark), as well as of isolated bark of *Quercus* species and other potential hosts. Plants for planting and cut branches are considered to be potential pathways, although it is noted that there are no reports of infections on oak seedlings or sapling in US nurseries (EFSA, 2018). There is no evidence that *B. fagacearum* can be transmitted via acorns.

PEST SIGNIFICANCE

Economic impact

Although *B. fagacearum* is currently considered to exist in at least 830 counties of 24 US states (Juzwik *et al.*, 2011) significant losses are only occurring in limited portions of the disease range. For example, even though the pathogen is reported in all but two counties in West Virginia, less than one tree per km² of oak forest is killed by the pathogen each year. In contrast, tens of thousands of oak trees (primarily red oak species) die in portions of Michigan, Minnesota, Texas, and Wisconsin each year where epidemics are on-going. Economic losses such as reduced production of timber and decreased property values, are associated with the large numbers of trees that die each year in impacted areas. However, salvage of timber from forest stands affected by the disease occurs for domestic production of wood products. Proceeds from sale of salvaged timber in oak wilt control zones usually covers the cost of treatments. Arguably, the greatest negative economic impact has been in urban and community forests due to tree removal and replacement costs of oaks of high amenity value. For additional information, see Haight *et al.* (2011) and Davies (1992).

Control

Control methods are available and carried out in both rural and urban forests in the USA, particularly in areas where *B. fagacearum* is causing significant losses or has the potential to do so. Several mechanical methods that sever roots are available for disrupting pathogen spread through connected root systems, e.g. vibratory plow, excavation, rock saw. Herbicides applied to encircling chainsaw cuts on lower stems of trees is used to kill all oaks within root-grafting distance of diseased oaks in some rural forests where heavy, large equipment cannot be used or is not desired. Removal of red oaks prior to sporulating mat formation (sanitation), avoidance of harvesting or pruning oaks during high-risk season and preventing movement of oak firewood are primary methods used to prevent insect transmission. High-risk season is the time period during which the following co-occur: a) large diameter vessel formation (= earlywood), b) a peak population period for free-flying, primary vector species, and c) a peak period of mat production. The time of this high-risk period varies by latitude. For trees of high amenity value, e.g. urban and community trees, systemic fungicides can suppress disease development but cannot eradicate the pathogen (Blaedow and Juzwik., 2010). The greatest success is obtained with an integrated management approach (i.e. use of two or more tools). Oak wilt has been managed successfully in localities of the USA where consistent and sustained disease control programs exist. Recent reviews include Juzwik *et al.* (2011) and Koch *et al.* (2010).

Phytosanitary risk

Oak trees are widely distributed across the EPPO region, although excluding northern parts of Scandinavia and Russia. They are of significant economic and ecological importance, as well of high symbolic value in many countries of the EPPO region. *B. fagacearum* constitutes a threat to the EPPO region because of the susceptibility of the major European oak species and their potential for root graft transmission of the disease. The white oaks, *Q. robur*, *Q. petraea*, *Q. suber* and *Q. ilex* are very important forest and amenity trees in the EPPO region. North American red oaks such as *Q. rubra* have been extensively planted in some countries (e.g. in France). The current distribution of *B. fagacearum* in North America correspond to climatic areas which largely overlap with the distribution of native *Quercus* species in Europe (EFSA, 2018). Therefore, climatic conditions prevailing in a large part of the EPPO region will not be a limiting factor for the establishment of the pathogen if it was to be introduced. The occurrence in the EPPO region of insects which appear to have the potential to be highly effective vectors, e.g. the European bark beetle (*Scolytus intricatus*) (Doganlar *et al.*, 1984; Gibbs *et al.*, 1984), also adds to the risk.

PHYTOSANITARY MEASURES

EPPO's recommendations to prevent the introduction of *B. fagacearum* are detailed in PM 8/5 *Quercus* (EPPO,

2017). For *Quercus* wood (including wood chips, and other wood residues) from the USA, several options are proposed (alone or in combination): pest-free area; debarking, treatments (heat, fumigation), and restrictions on transportation of wood to avoid possible infestations by insect vectors. A pest-free area is also recommended for imports of plants for planting and cut branches. Seeds should come from a pest-free area or should have undergone a hot water treatment. Guidance on how to inspect imported consignments of wood chips, hogwood and bark is provided in the EPPO Standard PM 3/87 (EPPO, 2019). Concerning fumigation treatments, recent studies have identified sulphuryl fluoride as a possible replacement for methyl bromide (Yang *et al.*, 2019) or use of vacuum and steam process (Juzwik *et al.*, 2019; EFSA, 2020). The possibility of fumigation or non-degrading heat treatment is principally relevant for red oak logs with bark attached, intended for the veneering industry.

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