Tip over disease of eggplant - *Diaporthe vexans*

*Phomopsis vexans* is a pycnidial fungus with an apparent sexual form in the genus *Diaporthe*. Easily seedborne and producing large numbers of conidia, it causes disease in *Solanum melongena* (aubergine, brinjal, eggplant), its only significant host, ranging from poor seed germination and damping-off of seedlings to leaf and stem lesions and to fruit rot, both in the field and after harvest. The fungus has been reported from widely distributed areas of most continents, but only a few of those are in Europe and Africa, even though the climates are favorable. Seed transmission may explain its broad historical distribution, but limitation of its host range to a non-staple vegetable crop can allow for its avoidance and eradication by cultural methods. As a result, it does not appear often on lists of restricted pathogens, even though it may cause yield losses of more than 50%. *Phomopsis vexans* may be introduced readily in the seed, as well as in or on harvested fruit. Phytosanitary regulation of imported seed and fruit, as well as a grower's selection of clean seed, will readily prevent most introductions. If introduction occurs, destruction of crop debris and crop rotation for several years will reduce or eliminate the fungus from a specific area.

*Diaporthe vexans* (Sacc. & P. Syd.) Gratz 1942

Pycnidia subepidermal, erumpent, dark, thick-walled, flattened to globose, varying in size, often 100-300 µm diam, with or without a beak; beak to 76 µm. Phialides hyaline, simple or branched, sometimes septate, 10-16 µm long, arising from the innermost layer of cells lining the cavity. Alpha conidia hyaline, aseptate, sub-cylindrical, 5-8 x 2-3 µm. Beta conidia filiform, curved, hyaline, septate, 18-32 x 0.5-2.0 µm, non-germinating. Hyphae hyaline, septate, 2.5-4 µm diameter. (see Edgerton and Moreland, 1921; Sherf and MacNab, 1986; Singh, 1987).

Perithecia in culture usually in clusters, 130-350 µm diameter, beaked; beaks sinuous, carbonaceous, irregular, 80-500 µm long. Ascii clavate, sessile, 24-44 x 5-12 µm, eight-spored. Ascospores biseriate, hyaline, narrowly ellipsoid to bluntly fusoid, one-septate, constricted at the septum, 9-12 x 3-4.5 µm (see Gratz, 1942).

A number of workers have studied the factors affecting growth and sporulation of the fungus in culture (Gratz, 1942; Pawar and Patel, 1957; Lapis and Deangkinay, 1967; Panwar and Chand, 1968; Hasija and Chowdhury, 1980; Singh and Chand, 1986; Islam and Pan, 1990a).

**Host range:** Apparently only a serious pathogen on *Solanum melongena*, however reported on multiple genera of Solanaceae as well as reports on *Acacia* sp. (Fabaceae), *Prunus* sp. (Rosaceae), and *Sorghum bicolor* (Poaceae).

**Geographic distribution:** Cosmopolitan.

**Notes:** Although this is supposed to be a teleomorph name, it is only valid as a new anamorph combination as there is no Latin diagnosis (Art. 59, ICBN). *Phoma solani* Halst. may be a nomen nudum and thus invalid.

Species of *Phomopsis* often have similar or overlapping ranges of morphological measurements, and the actual host specificity of each reported species is usually unknown (Uecker, 1988). At least nine other species are reported on *Solanum* hosts, including those on *Lycopersicon esculentum* L. (tomato), now considered to belong in *Solanum* (USDA-ARS, 2009). Comparative studies of morphology and pathogenicity under identical conditions may be needed to provide a basis for the accurate separation of these *Phomopsis* species.

Similar post-emergence damping-off of seedlings may be caused by *Rhizoctonia solani*, which does not produce pycnidia; its distinctive broad hyphae may be observed with the microscope (Edgerton and Moreland, 1921).

Other fungi cause spots on leaves and fruits of eggplant (Schlub and Yudin, 2002), but the large dark *Phomopsis pycnidia* produced in the lesions are distinctive (Chupp and Sherf, 1960). *Phoma exigua*, which may colonize the lesions as well, produces only small ellipsoid conidia, some of which may be septate (Boerema et al., 2004).

Blossom end rot of eggplant, due to a physiological condition, occurs only on the bottom part of the fruit (Meurant et al., 1999); fruit rot due to *Phomopsis* is more likely to begin at the top from infection of the calyx (Edgerton and Moreland, 1921).

The existence of several pycnidial fungi causing leaf spots on *Solanum melongena* L. resulted in difficulties with the identification of each one. Spegazzini (1881) described a fungus occurring on leaves...
of Solanum melongena in Italy as Phyllosticta hortorum. Halsted (1892) reported the same fungus on leaves and fruits of eggplants [aubergines] in New Jersey, USA, as Phoma solani. However, the name P. solani was already applied by Cooke and Harkness to another fungus on another host, and therefore Saccardo and Sydow (1899) substituted Phoma vexans. In 1905, Smith, observing septate conidia in the USA, proposed the name Ascochyta hortorum instead of P. hortorum. In Italy, Voglino (1907) studied a fungus on aubergine and agreed with Smith, concluding that the fungus described by Spegazzini as P. hortorum was an Ascochyta.

Cross-inoculation tests and morphological studies indicated to Harter (1914) that Phoma solani and Phyllosticta hortorum were the same species. He also concluded that the genus to which the fungus belonged was not Phoma, Phyllosticta or Ascochyta but Phomopsis. Unlike the previous workers, Harter observed and described the beta conidia (stylospores) characteristic of the genus. He proposed the name Phomopsis vexans for the fungus, and Spegazzini agreed that the American isolates were different from Phyllosticta hortorum (Harter, 1914).

While the coelomycete on eggplant that produces both alpha and beta conidia is a true Phomopsis (Uecker, 1988), the species Phoma hortorum Speg. and Ascochyta hortorum (Speg.) C.O. Sm. recently have been synonymized with Phoma exigua Desm. var. exigua, a weak pathogen of many plants that may be found in older lesions caused by other fungi (Boerema et al., 2004). Smith and Voglino were apparently observing yet another species, Ascochyta lycopersici Brunaud (Harter, 1914).

The teleomorph of the fungus has not yet been encountered in nature. Gratz (1942) observed perithecia on 2% potato dextrose agar in culture, and assigned the name Diaporthe vexans. The current view is that D. vexans is the teleomorph of P. vexans (Rehner and Uecker, 1994). Nevertheless, although the known connections of some Phomopsis species are to sexual forms in the genus Diaporthe, the name D. vexans (Sacc. & P. Syd.) Gratz is illegitimate, because Gratz did not provide a description in Latin of the new species (Punithalingam and Holliday, 1972).

Species concepts in Phomopsis, furthermore, have often been based on host specificity, but the phylogeny based on molecular data obtained so far indicates that either species have broader host ranges or significant changes, 'jumps', between hosts have occurred in species evolution (Rehner and Uecker, 1994). Additional molecular evidence, then, might connect the eggplant pathogen to an older species in either the anamorph genus or the teleomorph genus.

**DISTRIBUTION**

Phomopsis vexans has been reported from many areas in the warmer parts of most continents, but is unknown in Europe except in Romania (Smith et al., 1988) and known in only a few African countries. It is probably native to southern Asia, the area of origin of the host Solanum melongena (Prance and Nesbitt, 2005) where it is also reported to infect some wild Solanum species (Datar and Ashtaputre, 1988). That it is readily transmitted in and on the seed (Porter, 1943; Vishunavat and Kumar, 1993) of a crop that is only grown in limited areas may explain its lack of a continuous distribution in the tropics and subtropics. The fungus could be introduced to a region within a seed lot, but then die out if its presence discouraged continuous local cultivation of eggplant [aubergine].

**DETECTION AND INSPECTION METHODS**

Infection is easily visible in the field on close examination of leaves, stems and fruits; characteristic conidiomata appear as black pinhead-sized structures, which are often concentrically arranged on fruits. Infected fruits are soft and mushy or mummified and black. Infection of seed may be confirmed using the methods described for Seed Health Tests in ‘Seedborne Aspects of Disease’.

**DIAGNOSTIC METHOD**

A pure culture can be isolated from pieces of infected tissues on agar plates (Islam and Pan, 1990a). Phomopsis vexans produces abundant conidiomata on 4-7% oat meal agar medium at 30°C under light (Divinagracia, 1969). Pawar and Patel (1957) reported good production of pycnidia on agar made with an extract of the host. The blotted method can be used to confirm infection on seeds, as described under Seed Health Tests.

Sequences of ITS and LSU regions of rDNA for two isolates identified as P. vexans are available in GenBank for comparison (NCBI, 2009).

**NOTES ON CROPS/OTHER PLANTS AFFECTED**

Phomopsis vexans has been considered to be restricted to Solanum melongena (Edgerton and Moreland, 1921; Pawar and Patel, 1957; Sherf and McNab, 1986), but there are reports of pathogenicity to Capsicum annuum (pepper) and Lycopersicon esculentum (tomato) (Sawada, 1959; Tai, 1979) as well as of isolation from Acacia arcaefolia (Mathur, 1979), Prunus armeniaca (Dal Bello and Sisterna, 2000; Cho and Shin, 2004), and seeds of Sorghum bicolor (Mathur, 1979) and interception on imported
Capsicum frutescens (BPI, 1945). In India, it has been reported to infect some wild Solanum species in inoculation trials (Datar and Ashtaputre, 1988), and S. incanum (Dubey et al., 1987). Edgerton and Moreland (1921), nevertheless, were unable to obtain infection of tomato, pepper, potato or wild Solanum species, and Pawar and Patel (1957) report identical results for tomato, pepper and potato, as well as finding no infection of Solanum nigrum. Those reports did not specify the plant parts inoculated, but uninjured tomato and pepper fruits were found to be unaffected by the fungus in parallel trials with brinjal [eggplant] in India (Chaudhary and Hasija, 1979). Both young and fruiting pepper and tomato plants sprayed with suspensions of conidia were not infected (Harter, 1914).

SYMPTOMS

The symptoms range from poor germination and seedling blight to fruit rot. Post-emergence damping-off of seedlings results from infection of the stem just above the soil surface. The symptoms on leaves are more prominent during the early stages of plant growth. The lesions first are small, more or less circular, and buff to olive, later becoming cinnamon buff, with an irregular blackish margin (Pawar and Patel, 1957). Irregular spots result from coalescence. After transplanting, leaves coming in contact with the soil may become infected directly or develop leaf spot due to infection by conidia. Lesions on the petiole or the lower part of the midrib can result in death of the entire leaf. Affected leaves may drop prematurely, and the blighted areas become covered with numerous black pycnidia.

On stems and branches elongated, blackish-brown lesions are formed, eventually containing pycnidia. The diseased plant bears smaller leaves and the axillary buds are often killed. When stem girdling occurs, the shoot above the infected area wilts and dries up and the plant may be toppled by the wind (Edgerton and Moreland, 1921; Pawar and Patel, 1957; Sherf and MacNab, 1986). Pycnidia develop readily in lesions on young stems, but rarely on older ones (Harter, 1914).

On the fruits the symptoms appear first as minute sunken greyish spots with a brownish halo, which later enlarge and coalesce, producing concentric rings of yellow and brown zones. These spots increase in size and form large rotten areas on which conidiomata often develop concentrically, covering most of the rotten fruit surface. Pycnidia on fruit are larger than those on stems and leaves (Harter, 1914). If the infection enters the fruits through the calyx, the whole fruit may become mummified due to dry rot (Pawar and Patel, 1957). Rot may appear in fruit in transit after harvest (Sherf and MacNab, 1986).

BIOLOGY AND ECOLOGY

Life cycle: Conidia germinate after 6 hours and penetration occurs after 12 hours. In tissue the spread of the fungus is both intercellular and intracellular. Seedlings and young stems are highly susceptible. Mature tissue exhibits hypertrophy and hyperplasia below the infected region, preventing further spread of the fungus (Divinagracia, 1968).

Epidemiology: Phomopsis vexans requires hot and humid conditions for infection and disease development. Spore germination is optimal at 27° C, and pycnidial formation is greatest between 30° and 35° C (Pawar and Patel, 1957). The optimum relative humidity for disease development is 55% RH and above (Chaudhary and Hasija, 1979) and the optimum temperature for fungal growth is 28°C (Pawar and Patel, 1957). Fruit rot was maximal at 30°C and 50% RH in the growth chamber (Islam and Pan, 1990b); temperatures of 5°, 10° and 40°C were unfavourable for disease development in inoculated detached fruit.

Physiology and phenology: Isolates from various locations and different parts of the plant varied in some characteristics in culture, but the differences in source could not be related to differences in virulence (Islam and Pan, 1990a). Differences in colony morphology and growth rate, in production of the two forms of conidia, and in virulence on different plant parts were also observed among isolates by Edgerton and Moreland (1921).

MOVEMENT AND DISPERSAL

Natural dispersal: Conidia are disseminated locally by wind and rain (Edgerton and Moreland, 1921). The fungus also survives in crop debris (Ogilvie, 1924; Panwar et al., 1970).

Vector transmission: Edgerton and Moreland (1921) stated that insects may carry the conidia, but no particular genera or species were reported.

Accidental introduction: The fungus can be transmitted in and on seed (Porter, 1943; Vishunavat and Kumar, 1993) and on tools (Edgerton and Moreland, 1921). Infected seedlings may be transplanted from the nursery (Nolla, 1929).

SEEDBORNE ASPECTS OF DISEASE

Incidence
The fungus is seedborne at significant levels (Edgerton and Moreland, 1921; Ogilvie, 1924; Martin, 1934; Toole et al., 1941; Porter, 1943; Singh and Chakrabarti, 1982; Pan and Acharya, 1995), although certain varieties are more likely to be infected (Porter, 1943). Infected seeds contain profuse branched septate mycelium aggregated in the seed coat, between the seed coat and endosperm and in the embryo region of the seeds. Pycnidia are produced in the seed coat, between the seed coat and endosperm, and in the endosperm tissue (Vishunavat and Kumar, 1994).

Effect on Seed Quality

Infection in seed adversely affects the seed quality, causing seed discoloration, reduced seed weight and density, poor germinability and reduced viability (Toole et al., 1941; Porter, 1943; Panwar et al., 1970; Vishunavat and Kumar, 1993).

Pathogen Transmission

The fungus is seed transmissible (Nolla, 1929; Martin, 1930; Vishunavat and Kumar, 1993; Ogilvie, 1994; Pan and Acharya, 1995). Seedborne infection leads to pre-emergence and post-emergence damping-off of seedlings (Kaushal and Sugha, 1995). Infected seedlings bear conidiomata on the first true leaves, which serve as sources of primary inoculum. Conidia are disseminated by rain splash to other plants. The fungus also survives on infected crop debris, but seedborne inoculum is of great concern when the seeds are exported or imported to areas where the fungus is not already present.

Seed Treatment

Hot-water seed treatment has been recommended to reduce the incidence of infection in seed without adversely affecting seed viability (Martin, 1930; Felix et al., 1965). Seed treatment with formaldehyde is also effective (Edgerton and Moreland, 1921). Chemical seed treatment with captan, carbenzadim, carboxin, metasulfuron, thiram and triadimenol was found to increase germination and to reduce the incidence of damping-off of seedlings in artificially infested soil (Kaushal and Sugha, 1995). In the Republic of Georgia, extracts of garlic and celery were found effective as seed treatments for the control of P. vexans (Kuprashvili, 1996). Treatment with captan, carbenzadim, carboxin, dithane and mancozeb reduced the incidence of seed-borne fungi, including P. vexans, in local farmer seed lots, but not without reducing seed germination in some cases (Thippeswamy et al., 2006).

Seed Health Tests

Dry seed examination: Examining dry seed with a magnifying lens or under a stereo-microscope reveals the presence of black pycnidia on the seed surface. However, this test may only give a partial measure of the presence of P. vexans; the absence of conidiomata on the seed surface does not indicate the absence of the fungus on or in seeds. Infected seeds are often discoloured, appearing rusty-brown to black (Vishunavat and Kumar, 1993).

Blotter test: A 9.5 cm diameter Petri dish, made of glass or clear plastic, should be used to allow light to penetrate. Three layers of blotting paper, moistened with sterile water, are placed in the dish. Seeds from working samples are placed at a rate of 25 seeds per plate, equidistantly. Petri dishes are incubated at 25 ± 1°C for 7 days under artificial daylight or NUV light with alternating periods of 12 hours light and 12 hours darkness (Vishunavat and Kumar, 1993). The seeds are examined under a microscope. Infection is measured by the appearance of black conidiomata on the seed surface.

Economic impact: Fruit rot is the most destructive stage of the disease, as it damages the fruits partially or completely in the fields or during transit. The disease on stems and leaves results in reduction of fruit size and weight as well as loss of plants.

In Louisiana, USA, in 1921, at least 50% yield reduction was observed in eggplant crops due to infection in the field (Edgerton and Moreland, 1921). Later, Martin (1930) in the USA and Nolla (1929) in Puerto Rico also reported losses of 50% or more due to Phomopsis blight in aubergines. In Brazil in 1944, P. vexans caused such devastating losses that all control measures were impractical (De Figueiredo and Pereira, 1944). In India, the yield losses due to fruit rot ranged from 10% to 20% in the Punjab and Delhi (Panwar et al., 1970). In an advanced stage of disease, seed quality is also adversely affected, and infected seed becomes discoloured, with poor germinability and reduced seed viability (Toole et al., 1941; Porter, 1943; Vishunavat and Kumar, 1993). Seed infection results in pre-emergence and post-emergence damping-off of seedlings; approximately one-third of the plants were lost at each stage (Kaushel and Sugha, 1995).

MANAGEMENT

Prevention

Early warning systems
A linear model, based on environmental factors, for predicting *Phomopsis* blight in aubergines has been developed in India (Islam and Pan, 1992) but is not yet in use. Leaf blight severity was correlated with maximum and minimum temperatures and the number of rainy days.

**Cultural control and sanitary measure**

Burning of crop debris and burying it by deep ploughing are some of the cultural practices which may help to reduce disease incidence (Singh, 1987). The fungus is also capable of growing well on sterile vegetative structures of a number of other field and garden crops, such as cauliflower petioles and carrot and beet roots, some of which could then serve to perpetuate the fungus indefinitely (Howard and Desrosiers, 1941). Therefore, the efficacy of crop rotation as a control measure may vary, although a three-year rotation can be useful in reducing initial inoculum (Sherf and MacNab, 1986).

Use of an appropriate nitrogen source at a reduced level with higher rates of phosphorus and potassium fertilizer may increase yield without increasing disease (Sugha and Kumar, 2003).

Because the pathogen also survives on and in seeds, seeds should be collected from healthy plants and only disease-free seeds should be used.

**Chemical control**

Chemical control, especially the use of fungicides, is largely practised for *Phomopsis* blight control in aubergine [eggplant] crops throughout the world where the disease is prevalent (De Figueiredo and Pereira, 1941; Felix et al., 1965; Teo, 1982, 1984; Singh and Chakrabarti, 1982; Grewal and Jhooty, 1987; Jacqua and Gerion, 1988; Islam and Pan, 1989, 1993; Mohanty et al., 1994; Manna et al., 2004). The more common fungicides applied as foliar sprays are Bordeaux mixture, captan, captafol, carbendazim, carboxin, chlorothalonil, copper oxychloride, dithiocarbamates, manebe, mancozeb, thiophanate-methyl, tolclofos-methyl, ferbam and zineb.

Confirming the results of other workers, Beura et al. (2008) found that carbendazim provided the best control of *Phomopsis* under their test conditions in Orissa state (India); its use also allowed for the maximum increase in yield. In the laboratory, carbendazim completely inhibits culture growth (Mohanty et al., 1994); sensitivity of spore germination to the fungicide is high, though not as high as sensitivity to prochloraz (Sugha and Kumar, 2004). The newer systemic fungicide tebuconazole also provides a high level of control at a low concentration (Manna et al., 2004).

Tests of some natural plant extracts and homeopathic drugs showed that thuja, teucrium and extracts from *Allamanda cathartica* and *Aegle marmelos* could prevent or reduce growth of the fungus in vitro as did an effective fungicide, though higher concentrations of active ingredient were required (Panda et al., 1996). Some unidentified compounds extracted from *Allamanda cathartica* using organic solvents prevented growth of *P. vexans* in culture at unspecified concentrations (Masuduzzaman et al., 2008).

**Host resistance**

The use of resistant varieties can be one of the most effective methods of control (De Figueiredo and Pereira, 1944). Extensive work in breeding for resistance to *Phomopsis* blight in aubergines has been carried out with some success in Florida, USA (Decker, 1946, 1947, 1948, 1949), India (Kalda et al., 1976; Datar and Ashtaputre, 1988; Pandey et al., 2002), China (Ren and Zhang, 1993; Liu, 1998) and Brazil (Reifsneider and Ashtaputre, 1993). In India, other *Solanum* species have been identified as sources of genes for resistance (Sherf and MacNab, 1986; Datar and Ashtaputre, 1988). Nevertheless, Pandey et al. (2002) found no variety tested to be immune from stem blight or fruit rot. Some were moderately resistant and one escaped severe disease due to early maturity.

Resistance to *P. vexans* is probably due to chemical and protoplasmic factors rather than structural and mechanical processes (Howard and Desrosiers, 1941).

**GAPS IN KNOWLEDGE/RESEARCH NEEDS**

The frequency of occurrence of the sexual (*Diaporthe*) form in nature and its possible role in the epidemiology and biology of the pathogen remain undetermined. Additional molecular examination of *Phomopsis* species on *Solanum* hosts could clarify their identities and host ranges. Continued breeding for resistance may yield better cultivars for areas where the pathogen is endemic.

**References**


Use this link to revisit SMML website
Pycnidia on *Solanum melongena* fruit. 7.5X. BPI 617500. BPI 617500

Pycnidia on *Solanum melongena* fruit. 10X. BPI 617500. BPI 617500

Cross section of pycnidium from lesion on *Solanum melongena* fruit. 200X. BPI 617500. BPI 617500

Betaconidia from pycnidium. 400X. BPI 617500. BPI 617500

Betaconidia from pycnidium. 1000X. BPI 617500. BPI 617500