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Symptoms, Biology and Management of Ascochyta Blight (*Phoma exigua*) of French Beans: A Review

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ABSTRACT

French bean (*Phaseolus vulgaris* L.) is a major export vegetable crop that is experiencing severe production problems because of the disease Ascochyta blight (*Phoma exigua*). The crop can be consumed as a green-pods, dry (mature) beans and shell beans. This is one of the major crops that can be grown all over the world. The most damaging disease of this pulse crop occurring worldwide is Ascochyta blight, attracts a lot of attention from researchers. The pathogen has the ability to significantly reduce yields. In certain parts of the world, it is thus known as a major economically significant disease of the common bean. Understanding the Ascochyta blight of French bean pathogen, disease growth, disease cycle and management strategies for efficient disease control will be the subject of this review. This pathogen first infects the foliage and then on peduncle, petiole, node, on pods and even cause stem girdling also. And there will be formation of clearly visible concentric rings of the fruiting bodies in symptoms. The disease is seed-borne and short distance spread will be with the help of wind and rain-splashes.

Key words: Ascochyta blight, Disease symptoms, French beans, Management, Pathogen.

The French bean (*Phaseolus vulgaris*) its domestication started around 6000 years ago in central and south America, which eventually lead to its spread throughout the world. it is an important crop of cooler tropics (Chatterjee and Bhattacharyya, 1986). The crop is cultivated in a variety of agroecosystems around the world, ranging from tropical, sub-tropical and temperate climates (Popelka *et al.* 2004). It is primarily grown and consumed in developing Latin American, African and Asian countries. It is the most essential legume for direct human consumption on the planet.

Beans' ability to fix nitrogen in the soil, as legumes, contributes significantly to the enhancement and long-term survival of soil fertility. As a result, they're used in crop rotation, as well as cover crops and green manures (Stoddard *et al.* 2010). The states of Maharashtra, Jammu and Kashmir, Himachal Pradesh, Uttar Pradesh, Tamil Nadu (Nilgiri Hills, Palani Hills), Kerala (Parts of the Western Ghats), Karnataka (Chickmagalur Hills) and West Bengal (Darjeeling Hills) are the major producers of common bean in India (Choudhary *et al.* 2018). The crop can withstand daytime temperatures of 35°C on occasion, but flower abortion is common. Frost destroys the plant if the temperature drops below 10°C (Messiaen *et al.* 2004).

Fungal diseases, in general, have a negative impact on crop development and productivity (Parveen *et al.* 2021). *Phoma exigua* var. *exigua* desmaz (syn. *Ascochyta phaseolorum* Sacc.) induces Ascochyta blight (Parveen *et al.* 2019). The pathogen can survive on the seed for up to two years and it can also spread from diseased plants and old plant tissues (Srivastava *et al.* 2020). The fungus can spread across the plant and it is also spread *via* seed (Kumar *et al.* 2020a). The pathogen thrives in conditions that are cool and humid (Hanson *et al.* 1993). When disease severity is high, the plant loses leaves and pods under favorable conditions (Bardas *et al.* 2008). *Phoma* is a large genus of fungi that can be found in a variety of ecological niches and is found all over the world. Several *Phoma* species are also quarantine-relevant, posing significant challenges to organizations responsible for plant health quarantine control (Aveskamp *et al.* 2008).

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Symptoms

The symptoms Ascochyta blight first appear on the leaves (Fig 1A). These are black, concentric, zonate lesions that are 1-3 cm in diameter and may produce small black pycnidia later on (Liu *et al.* 2016). These dark to black lesions can affect the peduncle, petiole, node and pod, causing stem girdling and plant death. The fungus has the ability to spread across the plant in a systemic manner. During severe epidemics, premature leaf fall can occur and the fungus is seed-borne (Urinzwenimana, 2017). The disease usually appears during the early stages of pod development and becomes more severe as the pods mature. Small, circular, dark-brown spots appear first on the leaves, then the leaves and pods. The lesions then turn a dark brown to black colour and have clearly visible concentric rings. The concentric ring lesions range in size from 10 to 30 mm in diameter and are primarily filled with small black pycnidia. On the leaves, lesions may coalesce and cover the majority of the leaf surface area. This causes dark grey to black concentric rings to form on the leaves, which then coalesce and the diseased tissues' centres fall out over time. Sunken lesions that girdle the stem cause breakage and eventual death of the sections above the diseased region, resulting in stem attack. If pods are present at the time of the attack or if the pathogen spreads to emerging or existing pods as the disease progresses, they become infected and lesions with concentric rings emerge as shown in Fig 1B (Luthra and Bedi, 1932). The canopy of severely infected plants has a tattered appearance, with a significant loss of leaves (senescence). Sunken lesions on the pods are divided into zones with a pale centre and dark edges and are normally covered by several pycnidia (Kumar *et al.* 2020b).

Pod drop caused by the disease has been confirmed in other parts of the world, with a negative yield potential for the crop. Lesions on pods may also be a cause of seed infection contamination. Infected seeds have a brown to black discoloration, making them unsuitable for seed and grain markets (Kumar *et al.* 2020b). Seedlings developed from infected seeds sometimes develop dark brown to black sunken lesions on the stems and cotyledons, which senesce prematurely, resulting in stunted plant growth. The severity of the disease is weather-dependent, so high yield loss is normal in cold, wet and high-relative-humidity environments (Tivoli and Banniza, 2007).

The pathogen

The pathogen is linked to the development of pseudothecia at the end of the cropping season, which helps the pathogen to survive in plant debris (Tivoli and Banniza, 2007). In culture, *Phoma exigua* isolates create a hyaline, septate, submerged mycelium. Spores are typically made up of two cells (Zaumeyer and Meiners, 1975). At 21°C, sporulation and germination are at their peak, while mycelial development is at its peak at 24°C. Temperatures over 30°C destroy the fungus (Schwartz, 1989; Schwartz and Steadman, 1989). Pycnidia, which are 60-150 mm in diameter, are produced by the fungus (Fig 2). The pycnidia of *Phoma exigua* var. *diversispora* measure 160 by 120 nm and the conidia are 6.8 by 2.7 nm (Urinzwenimana, 2017).

Most conidia are single-celled (Boerema *et al.* 1981). Conidia were cylindrical to oval in shape, allantoid, hyaline, pale yellow to brown in colour, typically one-celled and 2 to 3 × 5 to 10 μm in length (Marin-Felix *et al.* 2019). Some diagnostic characteristics of pathogens appeared on French beans are revealed in Table 1.

In all above-mentioned cases, pycnidia on the leaves, when not present at time of collection will develop within 48 hours after incubating detached leaves with leaf spots in Petri dish on moist filter paper. The type species of *Phoma* is only distantly related to the other members of this genus, but relatively close to *Ascochyta pisi*, the type species of the older name *Ascochyta* (Aveskamp *et al.* 2010). Since earlier, the genera *Phoma* and *Ascochyta*, both classified in the order Pleosporales of Ascomycota, have already been considered as closely related (de Gruyter *et al.* 2009) although also having some differences mentioned in Table 2.

There is a convention of using a single generic name, based on priority but regardless of whether the genus is "anamorphic" or "teleomorphic". This classification is used for all unambiguous monophyletic phylogenetic lineages (Aveskamp *et al.* 2010). The nine *Phoma* sections have teleomorph relations described in the genera *Didymella, Mycosphaerella, Leptosphaeria* and *Pleospora* (Boerema, 1997) indicating that *Phoma* anamorphs represent a polyphyletic group (Table 3 and 4).

Favouring epidemiological factors

The pathogen attacks the plant at various stages of development, resulting in yield losses of up to 100% when infected seeds are used for sowing and the climatic conditions are favourable for pathogen survival (Allen *et al.* 1996). High humidity, constant rains with winds and cool to moderate temperatures are conducive to infection by *Phoma exigua* var. *diversispora* (Boerema *et al.* 1981).

Survival and spread of the disease

When contaminated seeds are used for sowing, the pathogen is known as a wound-pathogen in various plant species and can cause extreme dumping. It has a polyphagous life cycle that allows it to live for many seasons in plant debris (MacLeod and Sweetingham, 1999). If air currents disperse ascospores produced in the perithecia on plant debris after short rains, they can fly a few kilometers from the infected to the new host plant, though most land only a few meters from the source (Tivoli and Banniza, 2007). After rainfall, the pseudothecia release ascospores, which are then spread over long distances by the wind. This explains why plant debris is such an important source of primary inoculum for Ascochyta blight production in subsequent seasons. The pathogen targets the bean plant's pods, cotyledons, growing hypocotyl, leaves and other aerial parts. Long-distance spread of the disease is primarily accomplished by infected seeds, while short-distance spread is accomplished by pycnidiospores carried by the wind and raindrops.

Management strategies of ascochyta blight

There have been many attempts to regulate Ascochyta, but none have been successful. As part of integrated pest management, it has been possible to handle Ascochyta to some degree by using a combination of control options

Fig 1: (A) Ascochyta spots on bean leaves shows concentric rings along with dark margin. (B) Ascochyta spot on bean pods (http://www.pestnet.org/fact_sheets/bean_ascochyta_leaf_spot_319.htm).

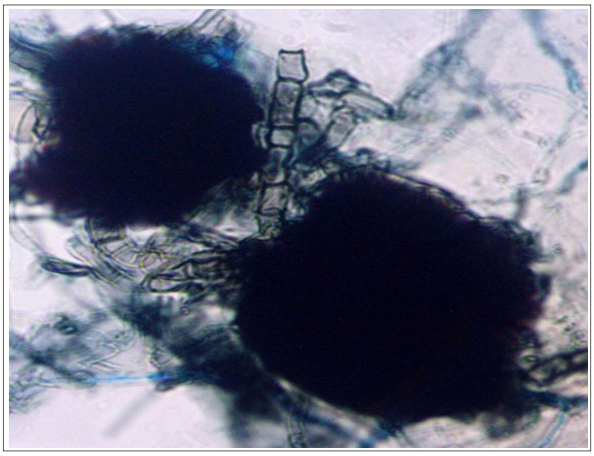


Fig 2: Microscopic image of Pycnidia of *Phoma* (pc by Sachinkumar Basavanagouda Koder).

(cultural, chemical and biological) (Buruchara, 2006). However, the use of resistant bean genotypes is the single most effective and realistic management strategy, particularly for resource-strapped farmers (Hall and Nasser, 1996). As only incomplete resistant varieties are available against this disease, some agronomic practices should be applied to control and reduce Ascochyta blight incidence wherever possible (Ouji *et al.* 2022). Crop rotation, wide plant spacing, clean seed planting, chemical seed treatment and foliar fungicide application are some of the control measures used (Schwartz, 1989; Schwartz and Steadman, 1989). Crop replacement with non-host crops for at least two years, use of uncontaminated approved plants, intercropping, field sanitation and destruction of infected crop residues may all help to contain the pathogen (Khan *et al.* 2013; Kumar *et al.* 2014). Since the fungus is dispersed in presence of water, fields should not be entered for cultivation or pesticide applications while the plants are wet, preventing unwanted movement in infested fields and reducing disease spread. This aids in the preservation of hygiene both on and off the farm (Moore *et al.* 2013).

Use of tolerant or resistant varieties

Host plant resistance is the most cost-effective, eco-friendly and recommended form of controlling bean diseases (Wahome *et al.* 2011). Bean breeding has aimed to create French bean cultivars that are more resistant to biotic stresses (Sharma *et al.* 2007). There are French bean varieties that are resistant or tolerant to *Ascochyta*. Current cultivars only have a low level of resistance to the pathogen, which is quickly broken down (Gan *et al.* 2006). This is because the pathogen's sexual stage (teleomorph) leads to the development of new races or pathotypes (Bretag *et al.* 2006; Gan *et al.* 2006). Because of the many physiological races of *Phoma exigua* and the pathogen's high variability, host resistance alone may not be enough. This is because varieties that are resistant to one race may be susceptible to another and tolerance sometimes breaks down when favourable conditions for disease development prevail (Duc *et al.* 2014). Many farmers have been forced to use foliar fungicides as a result of this. In order to control Ascochyta blight, a combination of genetic resistance and an effective foliar fungicide application program has been found to be effective (Gossen *et al.* 2011).

Biological control

The use of biocontrol agents has become a safer choice for the management of this disease. Several researchers are currently working in this field to discover environmentally friendly biocontrol agents having stronger capabilities against this phytopathogen (Kumar *et al.*

Table 1: Diagnostic characters of *Phoma exigua* var. *diversispora*, *Phoma exigua* var. *exigua* and *Stagonosporopsis bortensis* on *Phaseolus* beans.

Diagnostic characters	*P. exigua* var. *diversispora*	*P. exigua* var. *exigua*	*Stagonosporopsis hortensis*
Synonym	*Phoma diversispora*	*Ascochyta phaseolorum*	*Ascochyta boltshauseri*
Disease	Black node disease	Speckle disease	Blotch or leaf spot disease
Symptoms	Black nodes, dark, brown, black or greyish stems, crowded with pycnidia, large leaf spots with concentric rings, branches and whole plants killed, even when in full growth; pods may turn black at both ends and have concentric rings of pycnidia.	Small specks and spots, later coalesced and sometimes with concentric rings, on older leaves, stems and pods.	Reddish leaf spots, dark reddish-brown lesions on stems and pods. Plant in focus of attack may be severely stunted.
In vivo microscopic characters	Numerous black pycnidia on stems and petioles, sometimes also on leaves and pods.	Sometimes pycnidia on the leaves.	Usually, pycnidia on the leaves.
	Pycnidia about 150 mm, conidia continuous 6.8 (5.0 - 9.8) × 2.7 (2.3 - 3.2) µm, occasionally one or two septate along with two oil droplets inside.	Pycnidia about 150 mm, conidia continuous mostly 5 - 7 × 2.5 - 3.0 µm, ocassionally one or two septate along with two oil droplets inside.	Pycnidia about 100-200 mm conidia continuous, one, two, or three septate, 10 - 27 × 2.5 - 6.5 µm with two polar guttules. Stagonosporopsis was origi nally separated from *Ascochyta* due to occasional formation of multi-septate (Stagonospora-like) conidia (Diedicke, 1912).
In vitro microscopic structures	Conidia as above	Conidia as above	Conidia continuous 3.5 - 8.5 × 1.5 - 2.5 µm, one or rarely two septate 9 - 11 × 3 µm
Discoloration of culture medium with 1 N NaOH (E-test)	None (no E production)	Green (E production)	None (no E production)

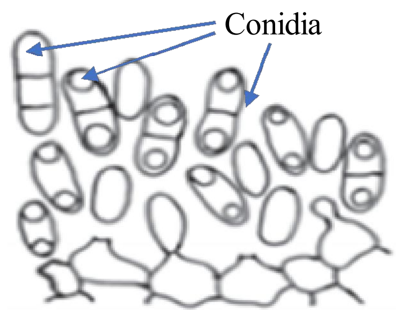
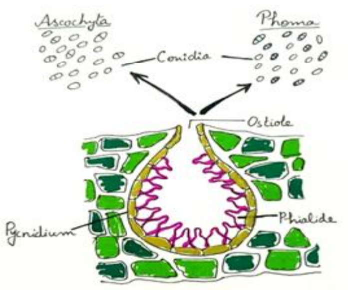
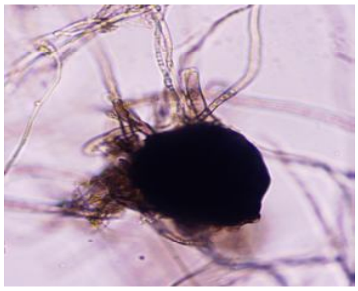


Table 2: Cultural and morphological characteristics of *Phoma* compared with published descriptions of *Ascochyta* blight causing fungi is mentioned below in the form of difference.

Ascochyta	*Phoma*
It is generally slow growing and may take 14-21 days to cover a standard 9 cm Petri plate (4-6 mm/day) (Harveson *et al.* 2011).	Colonies of *Phoma* grow very rapidly on culture media (Larone, 1995).
Colonies of the isolates on artificial media were flat, submerged with sparse mycelium. There were variations in colony colour in different isolates as the pathogen grew to advanced stages. The mycelium was pale cream at first but later turned greyish white or green to greenish dark and creamy white. However, most of the isolates were greyish white (Baite *et al.* 2016).	Colonies are flat, spreading, powdery to velvety and often largely submerged in the medium. From the front, the colour is initially white and later becomes olive grey with an occasional tint of pink. From thereverse, it is dark brown to black. Some species (particularly, *Phoma cruris-hominis* and *Phoma herbarum*) produce a reddish-purple to yellowish-brown diffusible pigment which is readily visible from the reverse (de Hoog *et al.* 2000).
Pycnidia often arranged in concentric rings within those lesions and abundant (Baite *et al.* 2016).	Distribution of pycnidia on infected portion is scattered as well as concentric (Davidson *et al.* 2009).
The pycnidium is spherical or pear-shaped with a single opening called an ostiole. The pycnidia contain numerous hyaline spores embedded in a mucilaginous matrix. In the presence of free moisture, the material within the pycnidia absorbs water, becomes wet and swollen, their causing conidia to ooze out the ostiole in a slimy mass (Davidson *et al.* 2009).	Pycnidia are the large, immersed, erumpent or with beak piercing the epidermis, round to pyriform, lenticular to globose; thin membranous; asexual fruiting bodies which are 70-100 µm in diameter (Li *et al.* 2020). They are dark in colour and bear peg-like phialides at their inner lining. Pycnidia have one to several openings (ostioles) on surface from which the masses of small conidia in slime are released outside (Larone, 1995; Deb *et al.* 2020).
Conidial ooze from pycnidia was cream or light brown in colour (sometimes carrot red) (Trapero-Casas *et al.* 2012).	Conidia ooze in a pale white to pinkish coloured matrix (Aveskamp *et al.* 2008).
The conidia were oval to oblong, straight to slightly bent at one or both ends, hyaline, occasionally two-celled, rounded at both ends under compound microscope (Baite *et al.* 2016).	Conidia are hyaline, unicellular (occasionally one or two septate) and oval-shaped. Each conidium typically has two oil droplets inside (Hou *et al.* 2020).
Conidia develop on short conidiophores (Baite *et al.* 2016).	Conidiophore short or almost obsolete (Grimes *et al.* 1932; Horst, 2013).





(pc by Seweta Srivastava)

Ascochyta	*Phoma*
Some *Ascochyta* spp. produce chlamydospores which are resistant structures important in survival (Wallen and Jeun, 1968).	Some *Phoma* species produce brown chlamydospores that are arranged singly or in chains. These chlamydospores may be unicellular or multicellular and "alternarioid" (resembling *Alternaria*) in appearance (Aveskamp *et al.* 2010).

Table 3: Overview of the characters of the various *Phoma* sections in the boeremaean classification system (Boerema *et al.* 2004).

Section	Teleomorph	Synanamorph	Sectional character
Heterospora	-	Stagonosporopsis	Production of distinctly large conidia in addition to the regular conidia.
Macrospora	*Mycosphaerella*	-	Conidia large, measuring 8 - 19 × 3 - 7 µm.
Paraphoma	-	-	Setose pycnidia
Peyronellaea	-	*Epicoccum**	*Multicellular chlamydospores*
Phoma	*Didymella*	*Phialophora**	-
Phyllostictoides	*Didymella*	-	Small septate conidia in addition to the regular conidia
Pilosa	*Pleospora*	-	Pycnidia covered by pilose outgrows
Plenodomus	*Leptosphaeria*	*Sclerotium** *Phialophora**	Pycnidia scleroplectenchymatous
Sclerophomella	*Didymella*	-	Pycnidia thick-walled

*Synanamorph only recorded in a single species.

Table 4: Microscopic characteristics of different *Teleomorphic* stages of *Phoma* spp.

Didymella	*Mycosphaerella*	*Pleospora*	*Leptosphaeria*
Pseudothecia immersed, rarely superficial, separate or gregarious, globose to flattened, ostiolate, 80-450 µm, with 2-5 (-8) layers of pseudoparenchymatal cells. Asci bitunicate, cylindrical to clavate or saccate, 8-spored; asci arising from a broad hymenium among pseudoparaphyses. Ascospores mostly hyaline, or brownish, 1-septate spores (didymospores) or multiseptate dictyospores (de Gruyter, 2012).	Species belonging to Mycosphaerella were characterized as having pseudothecial ascomata that can be immersed or superficial, embedded in host tissue or erumpent, having ostiolar periphyses, but lacking inter-ascal tissue at maturity. Ascospores are hyaline, but in some cases slightly pigmented and predominantly 1-septate, although taxa with 3-septate ascospores have been recorded (Quaedvlieg, 2014).	Ascomata perithecioid or rarely cleistothecioid, sometimes clypeate, mostly globose, thick-walled, immersed or erumpent, black, sometimes setose, peridium composed of pseudoparenchymatous cells, pseudoparaphyses trabeculate or cellular, asci cylindrical, fissitunicate, with a well-developed ocular chamber, rarely with a poorly defined ring (J), ascospores hyaline to brown, septate, thin or thick-walled, sometimes muriform, usually with sheath, anamorphs hyphomycetous or coelomycetous (Kirk *et al.* 2001, 2008).	Pseudothecia immersed, subglobose, solitary or aggregated, thick-walled, pseudoparenchymatous to scleroplectenchymatous, ostiolate, unilocular. Asci bitunicate, broadly ellipsoidal, 8-spored, interascal filaments pseudoparaphyses, Ascospores biseriate, broadly fusiform, transversally 3-septate, dark-brownish. Conidiomata pycnidial, globose to subglobose, scleroplectenchymatous, with papillate pore, unilocular. Conidiogenous cells phialidic, ampulliform to doliiform. Conidia hyaline, aseptate, oblong to ellipsoidal. Sclerotia sometimes produced (de Gruyter *et al.* 2013).

2020b). Seed treatment with *Trichoderma harzianum* and *Bacillus subtilis* could effectively be used for controlling the infection of *Phoma exigua* (Bhattacharjee *et al.* 2019). Few other fungal antagonists such as *Chaetomium globosum*, *Trichoderma viride* and *Acremonium implicatum* are also giving promising results (Kumar *et al.* 2020b). Some botanicals extract of *Allium cepa* and *Maesalan ceolata* var. *goulun gensis* showed effective results against test pathogen (Rai *et al.* 2000; Choudhury *et al.* 2018).

Chemical management

The key routes of introduction of Ascochyta blight to a healthy crop are seed-borne inocula as well as infected or contaminated crops (Gan *et al.* 2006). The use of wide spectrum fungicides in seed treatment can help to reduce the number of fungal pathogens on the seed or in the soil. Seed dressings, on the other hand, can only protect emerging seedlings from Ascochyta blight transmitted via the seed. This means that seed dressing alone is ineffective in protecting emerging seedlings from Ascochyta blight inoculum splattered by rain (Markell *et al.* 2008). Synthetic chemicals have been related to the production of pathogen resistance to the chemicals, pollution, human health hazards and becoming prohibitively costly for smallholders (Pinto *et al.* 2010).

CONCLUSION

Phaseolus vulgaris (French beans) is regarded as one of the most important pulse as well as vegetable crops, with widespread cultivation in many parts of the earth. A variety of biotic and abiotic stresses have an effect on it. Plant diseases are a significant cause of biotic stress. Among all diseases of French beans one of the most common disease is Ascohyta blight, which is caused by *Phoma exigua*. According to the studies and observations the numerous biological, chemical and agronomical activities that were mostly familiar had a positive effect on reducing bean Ascochyta blight. The pathogen's destructive effects must be successfully tackled. If this pathogen is not managed properly, severe yield losses up to 100% can occur. As a result, providing a comprehensive understanding of the pathogen and incorporating management practices assists in the development of good agricultural practices and effective disease-reduction management systems, as well as contributing to the global economy.

Conflict of interest: None.

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