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A brief review on stemphylium blight of onion caused by *Stemphylium vesicarium*

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Abstract
One of the most significant vegetable crops farmed worldwide is onion (*Allium cepa*). The yield and output of the crop are impacted by a variety of biotic and abiotic factors. The quantity and quality of both the onion bulb and the seed are significantly decreased by the most harmful fungal diseases, such as stemphylium blight. To breed varieties with long-lasting resistance to the onion stemphylium blight and identify the source of resistance against the range of virulence present in the pathogen population, it is imperative to record population and individual changes with variability in morphological, cultural, and pathogenic characteristics. Furthermore, it is possible to manage the stemphylium blight of onions by using fungicides, but repeated use of these chemicals has also resulted in the development of pathogen resistance and has dangerous environmental side effects. Integrated disease management (IDM), which combines other non-chemical eco-friendly management strategies with chemical approaches to treat diseases more efficiently while using less chemicals, is the most effective and cost-effective way to reduce the threat of this disease. Among non-chemical eco-friendly management techniques, the use of biological control in place of or in addition to chemical disease treatment is becoming more and more common. The current review article discusses the stemphylium blight of onions with distribution, symptoms, pathogenicity, variability, and integrated management.

Keywords: Stemphylium blight, integrated disease management, biological control, *stemphylium vesicarium*, variability

Introduction
The Alliaceae family of plants includes many important and well-known crops, including onion (*Allium cepa* L.). In many Asian nations, it is used as a staple spice, salad ingredient, and important vegetable. There are numerous ways to use onions as seasonings. Additionally, it is employed as a condiment to flavor a variety of foods and medications (Alamiri *et al*., 2016, Anonymous, 2012) [1, 4]. In addition to being a good source of various proteins and vitamin C, onion bulbs are rich in the supply of phosphorus and calcium. Onion has well-known fungicidal and insecticidal capabilities (Mishra *et al*., 2014) [39]. Onion rank second in terms of global production within the FAO's list of fifteen significant vegetables and spice crops (Anonymous, 2013, Anonymous, 2014) [2, 3].

Numerous diseases are brought on by fungi, bacteria, viruses, nematodes, and abiotic factors (Paibomesai *et al*., 2012, Bhat *et al*., 2008) [42, 10]. Stemphylium blight, is one of the most severe and damaging fungal diseases that affect onions, severely reducing the quality and quantity of both bulb and seed production (Nisha, 2008) [41].

Small yellow to orange streaks that quickly grow into elongated, spindle-shaped to ovate-shaped scattered spots with pinkish borders are the disease's hallmark. It can severely harm leaves and seed stalks, especially in the onion seed crop, and result in crop losses of roughly 80–85% (Byung *et al*., 2004, Vohra *et al*., 1974) [13, 52]. Using resistant cultivars is the most effective and cost-effective way to manage plant diseases. It is necessary to identify the source of resistance against the range of virulence existing in the pathogen population to breed the kinds with long-lasting resistance to the stemphylium blight of onions. Studies of variability are crucial to capture the population and individual changes that result from variations in morphological, cultural, and pathologic traits. As noted by several workers, *Stemphylium vesicarium* has been discovered to exhibit a wide range of heterogeneity in disease symptoms expression under natural epiphytotic depending on the onion cultivars, environmental conditions, etc. (Hosen *et al*., 2009, Arzanlou *et al*., 2012, Daljeet, 1992) [22, 5, 17].

It is possible to manage the stemphylium blight of onions by using fungicides, however repeated use of these chemicals has also resulted in the development of pathogen resistance and has dangerous environmental side effects. Additionally, controlling diseases through host resistance is a feasible and desirable method, but it has its own drawbacks. The main issue is how long genes last or how well they work under field conditions. However, if additional disease management strategies are used to support resistance and keep the pathogen under control, it may last longer. Integrated disease management (IDM), which combines chemical and other non-chemical eco-friendly management strategies to treat diseases more efficiently while using less chemicals, is the most effective and cost-effective way to reduce the threat of this disease. Growing in favour among non-chemical eco-friendly management techniques is the use of biological control in place of or in addition to chemical disease treatment. The distribution, symptoms, pathogenicity, variability, and integrated management of onion stemphylium blight are all included in the current review paper.

Distribution and status of the disease

Pathogen is hemibiotrophic that causes stemphylium leaf blight (SLB) in onions. It also affects a wide variety of crops, as well as other major foliar diseases in cultivated *Allium* spp. Australia, Brazil, Canada, Cuba, Egypt, Ethiopia, India, Japan, Korea, Mexico, Netherlands, New Zealand, South Africa, Spain, Taiwam, Tonga, Portugal, United States, and Venezuela have all documented cases of the disease in onions. SLB was reported in Welsh onion in China, Japan, Korea, and Taiwan. Australia, Brazil, Canada, China, Ethiopia, Germany, Korea, Myanmar, South Africa, Spain, and Turkey are among the countries where SLB in garlic (*Allium sativum*) has been documented. In China, there have been reports of garlic blight brought on by species *Sclerotium solani*. Another plant affected by *S. vesicarium*'s purple leaf blotch is the leek (*Allium ampeloprasum*).

The first report of the Stemphylium blight caused by *S. vesicarium* was reported from Varanasi in India. The pathogen manifests perfect condition as *Pleospora allii* (Pers. er. Fr.) Rabenh (Rao, 1975) [44]. Later, *S. vesicarium* was discovered to cause onion and garlic leaf blight in Spain and garlic leaf blight in Brazil (Bassalote *et al*., 1992, Boiteux *et al*., 1994) [8, 12].

The disease is now common over the globe (Bhatia *et al*., 2014) [11]. Following the initial report of this disease incidence from India in 1975, other reports of its occurrence from different parts of the nation, including Punjab, Maharashtra, and Bihar, have been made. After surveying various areas, revealed the widespread presence of the disease and noted a greater disease incidence on onion seed crops than on bulb crops (Jakhar *et al*., 1996) [27].

While researching vegetable seed-borne diseases, Wu *et al*., 1979 [54] found that the germination of onion seeds was severely reduced by *Alternaria porri* and *S. botryosum*. Miller *et al*., 1978 evaluated the severity of infected onion leaves from the beginning of the bulb through bulb maturity and discovered that younger onions had significantly less severe leaf damage than older ones, however documented significant damage from stemphylium blight in South Texas. Another species of Stemphylium, *S. botryosum*, was identified by Singh and Sharma as the cause of the Kullu valley in Himachal Pradesh causing garlic (*Allium sativum*) leaf blight. Later, Thind *et al.* (70) revealed that Punjabi onion leaf blight was caused by both Stemphylium species, *S. botryosum* and *Stemphylium vesicarium*. According to Gupta *et al*., 2013 [19], purple blotch (*A. porri*) and stemphylium blight (*S. vesicarium*) are significant diseases wreaking significant havoc on onion crops in India.

The first report of SLB on onions came from Texas in 1976, and the disease was subsequently discovered in New York in 1985. In 1990, there was a significant SLB epidemic in New York, with the disease being found in all growing fields inspected there and causing severe foliar dieback in certain areas. SLB, however, was uncommon in 1993 and only sometimes found in NY onion crops in 1991 and 1992.

SLB was initially discovered in Canadian onion fields in the Holland Marsh and it has now spread to all of the province's regions that produce onions. SLB is now the foliar disease that causes the most damage. Even when SLB prevalence and severity are low, leaf blight, which is caused by *Botrytis squamosa*, used to be the most devastating foliar fungal disease of onions in the area. However, indications of this disease are now extremely uncommon. SLB has consequently emerged as a significant onion foliar disease in eastern North America as well as other significant onion production regions. Additionally, there has been a recent SLB outbreak in New Zealand onion production.

While conducting an onion survey in Canada between the years of 2012 and 2013, it was discovered that the severity of stemphylium leaf blight ranged from 2 to 60%. (Thind *et al*., 2001) [50].

Symptomatology

Many workers from different parts of the world have described the disease's symptoms. Singh and Sharma, 1977, described the initial symptoms on the leaves of garlic as small, yellowish circular to oblong spots, 2-3 mm in diameter, that quickly developed into spindle-shaped lesions of dirty white or grey colour, growing to a size of 4-5 cm in length and 1-1.5 cm in width with profuse sporulation in the lesion's centre. Purple lesions on the leaves and inflorescence stalk are the hallmark of onion stemphylium leaf blight (Wang *et al*., 2010) [53]. Shishkoff and Lorbeer 1989 [47], on the other hand, saw mild, oval lesions that grew and ultimately aggregated to obliterate *Stemphylium vesication*-affected leaves.

Onion Stemphylium blight initially manifested as dark purple and white leaf spots and then progressed to severe necrosis, mostly on older leaves (8). Dark purple dots with a halo of straw colour, a purplish black centre, and an eye-like form (5–15 mm long). Onion leaves initially had small, light yellow to brown water-soaked streaks in the middle that later evolved into spindle-shaped to ovate-elongated spots on the leaves and inflorescence stalks as a result of *Stemphylium vesicarium* infection (Sharma *et al*., 1999) [46].

The disease began as tiny yellow to pale orange flecks that appeared in the center of the leaf (Fig.1) and quickly developed into elongated, spindle-shaped ovate patches around distinctive edges (Fig 4). Later, as conidiophores and conidia mature, these spores turn grey in the center, giving the leaves a diseased appearance (Tomaz *et al*., 1988) [51].

Fig 1: Typical Damaged symptoms of Stemphylium Blight of Onion

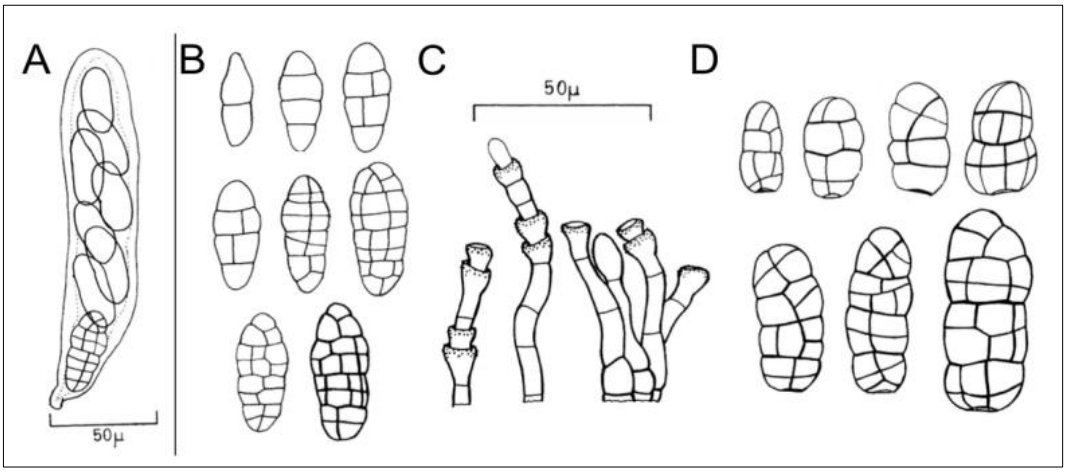

Fig 2: *Stemphylium vesicarium* A) Ascus, B) Ascospores, C) Conidiophores, and D) Conidia (57, 60).

Pathogenicity and host

As a pathogen and saprophyte, *Stemphylium vesicarium* has a diverse spectrum of hosts. Asparagus, leek, Welsh onion, garlic, and European pear (*Pyrus communis* L.) have also been reported to be susceptible to *Stemphylium vesicarium* infection. In addition to known hosts, the pathogen can also grow as endophytes in the living tissues of a variety of plants and produce asymptomatic infections. The pathogenicity of *Stemphylium vesicarium* has been demonstrated by a variety of researchers using a variety of bulbous vegetable varieties. Both wounded and unharmed infected leaves exhibited typical spindle-shaped lesions and copious sporulation within seven days (Barnwal *et al*., 2003) [7]. When incubated in a moist chamber following inoculation with the pathogen, the fungus causes lesion formation on leaves of all ages of onion plants, notably on older ones. Additionally, after 5 to 8 days, either dark purple or white leaf patches or both lesion kinds.

Small yellow to pale orange specks or streaks that appear in the middle of the leaf at the beginning of the disease quickly evolve into elongated, spindle-shaped to ovate-shaped spots with distinctive pinkish borders that turn dark brown to black when sporulation takes place (Camara *et al*., 2002) [14]. When

examining the pathogenicity and virulence of several *Stemphylium vesicarium* strains, significant variation in the progression of the development of necrotic spots as well as the final incidence of the disease. When the incubation period was over, these fungi strains only displayed little random necrosis (Cedeno *et al*., 2003, Chaisrisook *et al*., 1995) [15, 16].
After five days of incubation, white dots were seen on inoculated leaves. After 15 days of inoculating with a fungal isolate from garlic in a damp room, conidiophores and conidia were found on onion and garlic leaves (Ellis, 1971) [18]. The pathogenicity of *Stemphylium vesicarium* isolates from various hosts revealed that isolates from dead grass leaves and pear orchards were harmful in bioassays on pear leaves or fruits. Additionally, it was said that *Stemphylium vesicarium* from onions or asparagus did not cause pathology in pears (Hassan *et al*., 2007) [21].
The appearance of *Stemphylium vesicarium* lesions was seen on onion leaves 9 to 14 days after inoculating each isolates with a spore suspension (2×10^6 conidia ml^{-1}) on onion leaves in a controlled setting. Regarding the number of lesions per leaf, considerable variations between isolates were detected (Hassan *et al*., 2007) [21].

Crop Loss
SLB has the potential to reduce onion crop productivity and quality by up to 90%. Reduced photosynthetic area caused by SLB defoliation leads to the production of smaller bulbs. The disease is connected to up to 74% premature mortality during severe conditions. SLB, in the state of Georgia causes losses of up to 60% in the Vidalia onion crop. SLB infection happening later season, or a delayed disease development could explain why the disease does not affect yield. Early leaf senescence, which is frequently brought on by SLB, makes the crop more vulnerable to postharvest losses. Maleic hydrazide is typically treated before lodging to prevent sprouting during storage. This product needs to be sprayed while the plant has still five to eight green leaves on it to enable optimum maleic hydrazide uptake into the bulbs. Because the crop must be sold promptly after harvest and frequently at a time when prices are lower without the sprout inhibitor, the yield is reduced. Additionally, when onion leaves drop off early and don't lodge, plants are more vulnerable to bacterial bulb rots, which reduces the quality and value of the harvested products (Hosen, 2010) [23].

Life Cycle
Volunteer onions, transplants, and onion seed
Seeds, transplanted seedlings, and leftover onion plants may all contribute as the primary source of inoculum of *S. vesicarium.* The pathogen can be found in or carried by onion seeds, and infected seeds quickly infect seedlings. It was hypothesized that contaminated seeds played a role in the 1990 SLB epidemic in New York.
SLB is viewed as a colonizer of lesions caused by many other diseases, which is how the majority of commercial onion seed used in North America and Europe is produced, even though SLB is not a dominant pathogen under arid circumstances.
The pathogen *S. vesicarium* was not discovered during health testing of various commercial untreated onion seed batches. To reduce the risk of transmission through seeds and seedlings, the majority of commercially available onion seed used in conventional production is fungicide-treated. Due to this, it is now believed that traditional onion production in the north-eastern United States does not significantly rely on contaminated seed as a source of primary inoculum (Hunter *et al*., 2006) [24].
Transplants have been found a significant source of viruliferous thrips for direct-seeded crops in various regions of New York. Two ways that transplants might be a source of SLB are the introduction of infected transplants from the southwest United States, where infected transplants are produced and planted or the provision of early-planted crops in which can establish, grow, and subsequently provide inoculum for nearby onion fields (Fig 2).

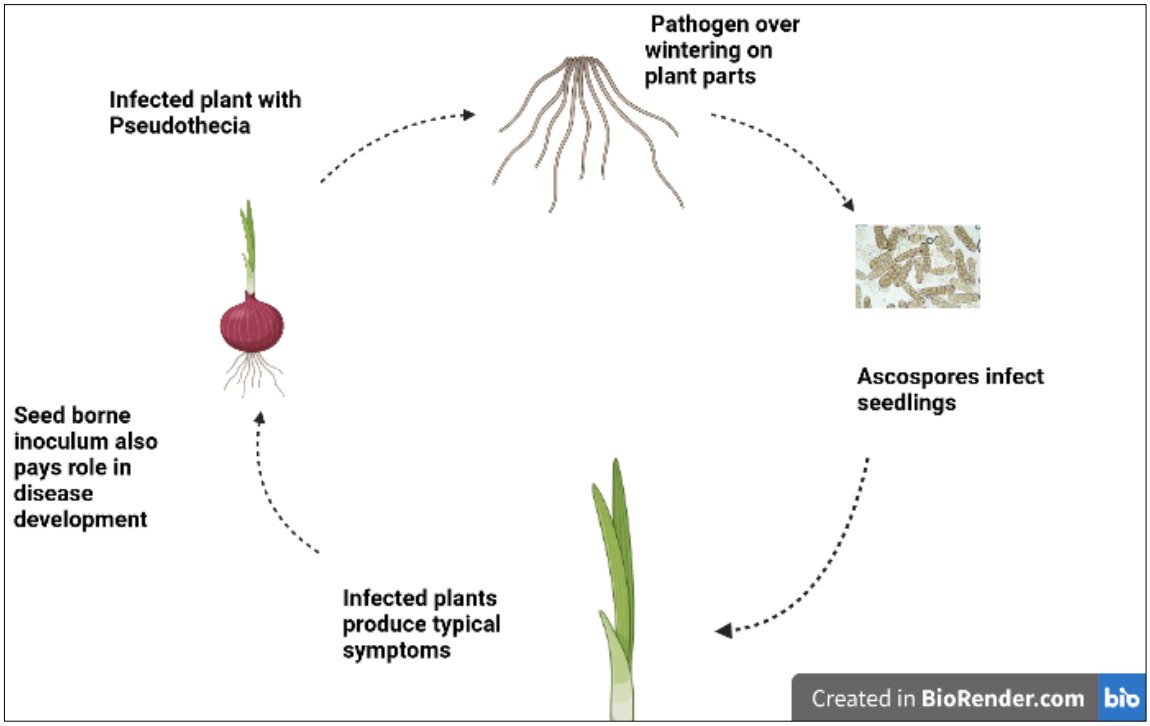

Fig 3: Life cycle of *Stemphylium vesicarium* on onion (60)

Crop residue

S. vesicarium overwinters on the leaf remnants of numerous crops as mycelia or pseudothecia, while it is unclear how ascospores contribute to the major inoculum on onions. For instance, pseudothecia on dry stalks from the earlier year were a significant source of inoculum for purple spot-on spears. Pseudothecia that overwintered on the orchard floor of pear orchards produced ascospores that colonized the dead grasses and detritus, creating conidial inoculum for fruit infection (Ihsanul *et al*., 2007) [26].

S. vesicarium pseudothecia on garlic residue developed best in the temperature range of 5 to 10 °C. When temperatures climbed by at least 14 °C, the spores that had formed over the winter were expelled from the pseudothecia. A minor vapor pressure deficit, frequent precipitation, and temperatures between 11 and 21 °C, all happened at the same time as the ascospore discharge on the garlic. Similar to this, pseudothecia on pear residue only developed gradually and only in conditions of high relative humidity of 98% and temperature of 10 and 15 °C. In Ontario and New York, ascospores may have different effects on the primary inoculum of onions. On leaves and stalks, pseudothecia often develop at the end of the season. Onion leaves in New York abound with immature pseudothecia late in the growing season, and ascospores have been found in pseudothecia on leaves stored for several weeks at 5 °C in plastic bags (Kohl *et al*., 2009) [28].

Ascospores were found in large quantities in onion fields in Ontario early in April, but ascospore capture decreased and was nearly non-existent when the onion crops were sown. This showed that *S. vesicarium* overwintered close to onion fields, although conidia were probably the predominant source of inoculum causing SLB outbreaks on onions in Ontario (Koike *et al*., 2005) [29].

Alternative hosts

Pathogen overwinter on weed remains before releasing ascospores in the early spring. In Ontario and New York, *S. vesicarium* asymptomatic hosts have recently been found in common weeds that border onion farms. These plants include jimson weed, bull thistle (*Cirsium vulgare*), purslane (*Portulaca oleracea*), and redroot pigweed (*Amaranthus retroflexus*). It looks likely that both regions early-season inoculum contains a portion of weeds that were previously infected and their winter remnants. Weeds may or may not be a substantial source of inoculum, and because airborne spores can spread far, eliminating such inoculum reservoirs may not be useful for suppressing disease in onion crops (Kumar *et al*., 2007) [30].

Secondary spread

Large amounts of conidia are present in onion crops during the growing season, and this increases the amount and increases disease incidence and severity. This shows that secondary disease transmission may include airborne conidia. Splash dispersal of the pathogen from various sporulating leaves appears to be an important source of inoculum in New York. In Ontario, sporulation is less common on young onion plants, and older lesions are the first to show signs of sporulation before necrotic leaf tips do. Airborne distribution may therefore be more significant. The transmission of *S. vesicarium* conidia has also been connected to *Thrips tabaci*, which feed on onion leaves and induce feeding damage. When there is a lot of pressure, both SLB and onion thrips might result in severe leaf dieback (Llorente *et al*., 2006) [32].

Host-specific toxins

These compounds are produced by plant diseases that encourage pathogen colonization and the onset of symptoms in susceptible host plants by damaging or disrupting host tissues. The secondary metabolites produced by the pathogen that are host-specific or selective poisons are stemphylin, stemphyperylenol, stemphyloxin, and stemphol. Isolates of pathogens emit one or more host-specific toxins during spore germination in the pear, which causes necrosis and the death of plant cells. Toxins are thought to play a part in the pathogenesis to onions, albeit this has not been proven. The role played by the pathogen in the emergence of toxic symptoms that result in SLB may help to explain the presence of foliar lesions and leaf dieback (McKenzie, 2013) [34].

First, the emergence of distinct lesions is typically the first sign of SLB-related leaf necrosis. Additionally, when large numbers of *S. vesicarium* conidia are inoculated into onion leaves, acute chlorosis, and leaf blight take place rather than lesions being formed. Research revealed that in Ontario that different onion cultivars were consistently but somewhat susceptible to SLB. There were occasionally different lesions that were established among cultivars when compared to the amount of dieback detected within the same cultivars. Although most fungicide treatments generated outcomes that were similar for both leaf dieback, some treatments induced severe leaf dieback with substantial numbers of SLB lesions (Mehta, 2001, Mirjalili, 2011, Misawa *et al*., 2012) [35, 36, 37].

Factors Affecting Disease Epidemics

Temperature

S. vesicarium ascospores in the air are dropped with cumulative temperatures in the spring, but this may be more a result of ascospore depletion than a causal link between temperature and spores. Conidial releases were positively correlated with the number of temperatures of at least 15 °C.

Garlic and pears are thought to be related in several ways. Four *S. vesicarium* isolates required a temperature of 25 °C to grow mycelial in a test tube. Conidia should be cultivated at 23 °C or 29 °C, respectively, while ascospores should be germinated at a temperature of 31 °C. Pear infection peaked at 23 °C for fruit and 21 °C for foliage. The severity of the *S. vesicarium* infection on asparagus in New Zealand peaked at 14 °C. *S. vesicarium* conidia can infect onion leaves between 10 and 26 °C, yet the cardinal temperatures for onion infection have not been determined (Nguyen and Seifert, 2008) [40].

Moisture

Ascospore concentration on leek, garlic, and onion increased 24 to 36 hours following brief rainfall but decreased with extended, significant rainfall events. The amount of precipitation within ten days before was strongly correlated with an increase in the number of conidia recognized on onion in Ontario, where the concentration of conidia increased significantly 2 to 72 hours after rainfall. On garlic and other crops, it has also been noted that conidia discharge increases after rain. Spores being washed out of the air and off plant lesions, there were few to no *A. porri* conidia caught during rain occurrences. According to a recent study, an increase in the airborne concentration of *S. vesicarium* spores is correlated with an average vapor pressure deficit of £0.5 kPa over a given number of days.

S. vesicarium infection of garlic leaves required at least eight hours of leaf wetness at 10 °C, and the severity increased with rising air temperature (10 to 25 °C) and leaf wetness duration

(8 to 24 h). A lengthy dry period can also permanently stop *S. vesicarium* infection of pear leaves (Pattori *et al*., 2006) [43].

Cultural variability

Stemphylium vesicarium colonies on potato carrot agar media (PCA) are effetely grey to brownish grey in colour, olivaceous brown to black, and slightly velvety. After 7 days, the colonies formed concentric rings, were flat, and reached a diameter of 50 mm with sparse aerial mycelium growth (Shahnaz *et al*., 2013) [45]. Twenty four *Stemphylium vesicarium* isolates were analyzed in terms of cultural, morphological, and molecular characteristics from various onion-growing regions. It was noted that colony colors ranged from filthy white to deep greenish white, light grey to whitish, and deep greenish brown to dirty white. Brown, deep brown, and light brown were the reverse colony colors. The umbonate, elevated, and flat colony elevations had circular and asymmetrical colony forms. Colony margins were complete, undulate, and filiform, with cottony, fluffy, and velvety textures (Slimestad *et al*., 2007) [48].

Morphological variability

Oblong to ovoid, thickly verrucose, with 1-5 transverse and many longitudinal septa, and measuring 25-40, 13-21 µm in size, *S. vesicarium* conidia were described (18). Contrarily, it was found that *S. botryosum* isolated from lucerne (*Medicago sativa*) had conidial dimensions that ranged from 33 to 35, 24-26 m, and length/width ratio (61). In *P. allii*, the pseudothecia are black and carry many cylindrical asci. Ascospores are released by pseudothecia during springtime precipitation (60). The upper half is of ellipsoidal, yellowish-brown with ascospores and slightly tapered. Ascospores that have reached maturity have 0 to numerous longitudinal septa and 5-7 complete transverse septa.

The *S. botryosum* conidiophores had many nodular swellings and were short, septate, and light brown. The spores from the lesions and culture were ovate, muriform, echinulate, and had 3–4 cross septa. They were dark brown to black. 20-35 × 16-24 µm at the septa, slightly constricted (62). *Stemphylium vesicarium* conidia are medium golden brown to olive brown, oblong to broadly oval, and occasionally inequilateral (41). They measure 25-42 × 12-22 µm in size and have 1-6 transverse and 1-3 longitudinal septa. Conidiophores were cylindrical and ranged in size from 33-47×5-8 µm. They were straight to irregularly curved, simple or occasionally one-branched, and enlarged epically at the location of conidium production. *Stemphylium vesicarium pseudothecia*, or perfect state fruiting bodies, mature in 3 to 6 months. Young ascospores are ellipsoidal with the upper half narrowly tapered and cylindrical to clavate in shape. With seven transverse septa, ascospores ranged in size from 18 to 38 µm (Srivastava *et al*., 2005) [49].

Conidia of *S. botryosum* are olive brown, oblong, or muriform in shape, and have three constricted transverse septa. In contrast, the size of conidia in other Stemphylium species ranges from 78 × 24 to 13 × 8 µm, and that of conidiophores from 25 × 2 to 285 × 6 µm. (Wu *et al*., 1979) [54].

The conidia of *Stemphylium vesicarium* were up to six transverse and several longitudinal septa, most of which were constricted at the primary transverse septa, and they were verrucose, pale to mid brown, or olivaceous brown in color (Yun Fei *et al*., 2010) [55].

On PDA, young *Stemphylium vesicarium* conidia were spheroid in shape and had rounded ends. Mature conidia measured 20-24, 12-15 µm, were acrogenous, single, oblong to broadly ellipsoid, sub-truncate basally, and rounded to sub-truncate apically. They also had 1-3 transverse and 1-4 longitudinal or oblique septa, which were frequently constricted at one or more of the septa (5). Mature perithecia were evident in the cultures after around two months. Perithecia were gregarious, black, spherical to subspherical, rostrate, up to 500×1,000µm, and the neck was typically 25µm long. They could also be partially or completely submerged in the agar. Asci were cylinder-shaped, short-stalked, thick-walled, 8-spored plants that could grow up to 180×45µm. The mature ascospores were uniseriate to somewhat overlapping, yellowish brown, oblong to ovoid, obtuse basally, domical apically, 34-36, 14-17 µ m, broader in the upper half with 7 transverse and several longitudinal septa, more or less constricted at the septa. The mature ascospores were uniseriate to somewhat overlapping, yellowish brown, oblong to ovoid, obtuse basally, domical apically, 34-36,14-17 µm, broader in the upper half with 7 transverse and several longitudinal septa, more or less constricted at the septa (Zheng *et al*., 2007) [56].

Stemphylium vesicarium ascomata are globose and up to 0.5 mm broad. Bitunicate, narrowly cylindrical to clavate, and measuring 110-150×24-35 µm are asci. Ascospores are uniseriate, ellipsoidal, with an upper half that is narrowly tapered to a tip and a base that is rounded, measuring 33-38×15-20µm, 6-14 longitudinal septa, and 3-7 transverse septa. Major transverse septa are where ascospores are most constricted. Conidiophores frequently have one or more nodose swellings and black bands from which conidia protrude as a defining feature of the grouping. Smooth conidiophores or tiny verrucae are both possible. Conidia are solitary, straight or slightly curved, typically ellipsoidal, 20 to 50 µm by 15 to 26 µm, pale to olivaceous brown, verrucose, with up to 6 transverse septa and numerous longitudinal or oblique septa, frequently constricted at the 3 major transverse septa, and with a very distinct basal scar. (Fig 3).

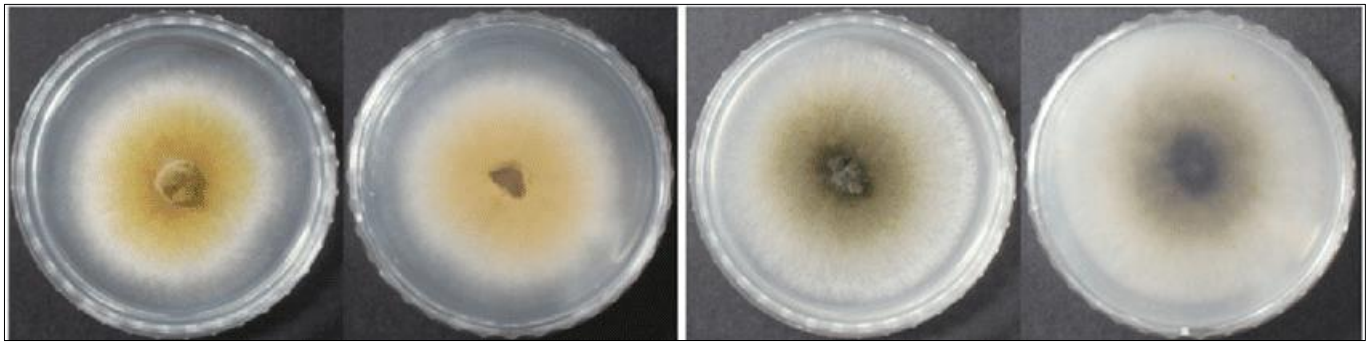

Fig 4: Pure Culture of *Stemphylium vesicarium*

On the basis of conidial morphology, five pathogens from onions were isolated and classified as *Stemphylium vesicarium*. The conidia ranged in size from 19-23×22-46µm and were oblong in form. Except for NO35, all of the isolates sporulated on vegetable 8 agar media (V-8), and the mean number of conidia recovered per colony for each isolate was 1×104 ml-1 (Bhatia and Chahal, 2014) [11].

Molecular variability

Stemphylium species are distinguished primarily by the morphological traits of the conidium and conidiophore, which frequently overlap between species and make species determination challenging. Even more so, these qualities change depending on the substrate and the temperature. It is not always simple and frequently inaccurate to identify a fungus species based solely on its shape. In order to evaluate morphological concepts and other taxonomic assumptions, DNA sequence data are currently being utilised (Hassan *et al*., 2007) [21]. For the identification of fungi, the internal transcribed spacer regions (ITS) sequence is an extensively used DNA marker (48). Genetic distance and genetic similarity, which both indicate that there are either differences or similarities at the genetic level, are often used to quantify genetic diversity. The exact classification of the fungal species is made possible by the accessibility of several polymorphism markers. RFLP, RAPD, SSR, ISSR, and SNP based markers are among the molecular markers currently accessible to evaluate the variability and diversity at the molecular level. *Pithomyces chartarum* and *P. atro-olivaceus* isolates, as well as twenty eight monoconidial isolates from five *Stemphylium* species classified according to morphology, were all found to exhibit DNA polymorphisms (16). Utilizing random amplified polymorphic DNA (RAPD) markers, the genomic similarities of geographically distinct isolates of the *Stemphylium* species isolated from alfalfa. From whole genomic DNA, 11 oligodeoxynucleotide 10-base primers produced 205 RAPD fragments. Two groups were formed using principal component analysis of RAPD fragment occurrence among the 28 Stemphylium isolates. *S. globuliferum* and *S. botryosum* were both part of one cluster. *S. alfalfae*, *S. herbarum*, and *Stemphylium vesicarium* made up the second cluster. The three species were distinguished in a separate examination of the second cluster. The distance between *P. chartarum* and *P. atro-olivaceous* and *Stemphylium* was considerable. All Stemphylium isolates shared a large RAPD fragment of roughly 2.5 Kb, while the *Pithomyces* species lacked it. Major RAPD segments showed significant cross-species cross-hybridization, suggesting that they shared the same nucleotide sequences, according to a Southern study. There was no evidence of cross-hybridization to the Pithomyces pieces. These findings confirmed recent taxonomy revisions based on morphology and showed that at least five genetically different Stemphylium species can produce leaf spot in alfalfa.

More significantly, Wang *et al*., 2010 [53] identified two new Stemphylium species based on morphological traits and molecular phylogenetic analysis. In Hebei Province of China and Angres, France, diseased onion (*Allium sativum* L.) leaves were the sources of the isolation of *S. phaseolina* and *S. variabilis*, respectively. The two species differ from related species based on the shape and development of conidia, however they both have typical Stemphylium morphology. Sequencing was done on the glyceraldehyde-3-phosphate dehydrogenase (gpd) and internal transcribed spacer (ITS) nuclear rDNA regions. The combined DNA sequences of these two gene areas underwent phylogenetic analysis, and the results established *S. phaseolina* and *S. variabilis* as two separate phylogenetic species.

It was discovered that testing of seven decamer primers did not reveal any band of DNA from *Stemphylium vesicarium* isolates during molecular variability research of 18 isolates out of 24 *Stemphylium vesicarium* isolates of onion. Using an ITS primer, the ITS sections from the DSTR 01 isolate were identified for DNA sequencing (ITS1F and ITS4R). Through the NCBI-BLAST algorithm, SV-DSTR 01 showed no discernible resemblance to any gene (49). A combined dataset of the internal transcribed spacer and glyceraldehyde 3 phosphate dehydrogenase regions were used for the sequencing analysis to identify 79 isolates as *S. lycopersici* while researching the tomato leaf spot disease. Amplification fragment length polymorphism (AFLP) analysis was performed on the 79 isolates using three different primer sets. The genetic diversity of the *S. lycopersici* population derived from the two cultivars was found to be extremely low (H = 0.0948). Cluster analysis revealed that isolates from the two cultivars were mingling. Additionally, study of molecular variance revealed that populations derived from the two cultivars had very little genetic difference, with Fst = 0.0206.

Management of the disease

The sowing date and spacing affect the disease incidence of *Stemphylium vesicarium*. It was highest (52.2%) when the crop was sowed on September 30 with a spacing of 30×45 cm, and lowest on October 30 with a spacing of 60×45 cm. The application of just K at 100 kg ha^{-1} resulted in the highest mean yield of onion cv. N-53 (136.25 q ha^{-1}) and had a substantial impact on the incidence of stemphylium blight (*S. botryosum*). At this level of K, average disease incidence and severity were 32.1 and 53.8%, respectively. When N and P were administered in the ratio of 100:100 kg ha^{-1}, a maximum mean yield of 155.4 q ha^{-1}, mean disease intensity of 33.9 percent, and mean disease incidence of 58.5 percent were recorded. The application of N, P, and K in the proportion of 100:50:100 kg ha^{-1} produced the highest mean yield of 161.3 q ha^{-1}. At this level, average disease incidence and severity were 31.9 and 49.2 percent, respectively. According to the findings of a three-year study on the impacts of various nitrogen rates and irrigation frequency for the management of the stemphylium blight disease, watering at 7-day intervals significantly reduced the prevalence of the disease and increased output. The 10-day irrigation interval and high nitrogen doses (125–150 kg ha^{-1}), however, were discovered to be more effective in minimizing stemphylium blight disease and increasing bulb production (Ihsanul and Nowsher, 2007) [26].

Triazoles, such as Contaf 5 EC (Hexaconazole), Folicur 25 EC (Tebuconazole) and Score 25 EC (Difenaconazole), and as well as contact fungicides like Antracol, Indofil M-45, and Kavach, have shown promising activity against the stemphylium blight (*S. botryosum*) of onions in both laboratory and field settings. When examining the effects of eight fungicides on the mycelial growth of *Stemphylium* spp. The systemic fungicides azoxystrobin and propiconazole and the contact fungicides Indofil M45 (mancozeb) and Antracol (propineb) showed the greatest inhibition.

Propiconazole at a concentration of 0.1 percent was applied

four times to the leaves, followed by Indofil M-45 (mancozeb at a concentration of 0.25 percent) and Blitox (copper oxychloride at a concentration of 0.3 percent).

In a field trial carried out during the rabi season of 1998–2000, the effectiveness of *Pseudomonas fluorescens* and hexaconazole in reducing *Stemphylium botryosum* that causes blight in onions (Ihsanul and Nowsher, 2007) [26]. The application techniques included seed treatment, root dipping, or foliar spraying, either separately or in combination. All of the treatments lessened the severity of the condition compared to the control, with hexaconazole treatment having a smaller impact than *P. fluorescens* treatment on the condition. Hexaconazole treatment increased crop yield compared to *P. fluorescens* treatment. The treatment that produced the lowest disease severity and maximum yield in onions was twice-foliar spraying.

Under *in vitro* circumstances, a number of bioagents, including *Bacillus subtilis*, *P. fluorescens*, *Trichoderma harzianum*, *Gliocladium spp.*, and *Saccharomyces cerevisiae*, were investigated against stemphylium blight. *P. fluorescens*, *B. subtilis*, and *T. harzianum* showed the greatest inhibition of *Stemphylium vesicarium* mycelial growth, and in an *in-vivo* research, these bioagents showed the most improvement in disease severity whereas *T. harzianum* showed the least improvement. After testing the effects of seven different fungicides, including Rovral 0.2%, Dithane M-45 0.2%, Tilt 0.05%, Cupravit 0.3%, Macuprax 0.25%, Ridomil MZ-72 0.2% and Bavistin 0.15% in the field to control *Stemphylium* blight lentil, fungicides Rovral 0.2% was found as most effective fungicide followed by Dithane M-45 0.2% and Tilt 0.05% (Koike and Matheron, 2005) [29].

Additionally, five bioagents (*T. harzianum*, *T. viride*, *Aspergillus niger*, *Penicillium citrinum*, and *G. virens*) and six plant extracts (*Azadirachta indica*, *Datura metel*, *Lantana camara*, *Parthenium hystorophorus*, and others) against *S. botryosum* were assessed. *A. indica* (66.5%) and *D. metel* (64.5%) were the plant extracts that were most effective at limiting pathogen development compared to controls, whereas *T. harizianum* (81.2%) and *T. viride* (74.5%) significantly slowed pathogen growth when used as bioagents. The most successful method for managing this disease in the field was pathogen suppression by treating garlic bulbs with *T. harzianum* at 0.2 percent coupled with two foliar sprays of *A. indica* at 0.2 percent and *T. harzianum* at 0.2 percent spaced 15 days apart.

Similar to this, eight plant extracts and bioagents for their ability to treat onion stemphylium blight brought on by *Stemphylium vesicarium* in an *in vitro* setting. Clove extracts of *Allium sativum* at 10% resulted in the greatest growth inhibition (57.31%) among the plant extracts, followed by Aloe vera at 10%. (47.15 percent). *T. viride*, one of the bioagents, was quite successful in preventing growth (56.15 percent). Tebuconazole, propiconazole, and the mixture of carbendazim percent + mancozeb (SAAF) appear to be promising substitutes for traditional fungicides like mancozeb and copper oxychloride for the effective management of *Stemphylium* blight disease of onion on seed crops (Kumar *et al*., 2012) [31].

Six pathogens were discovered to be linked to onion foliar blight disease while working on integrated disease management, including *A. alternata*, *A. porri*, *A. tenuissima*, *Stemphylium vesicarium*, *Colletotrichum circinans*, and *Cladosporium allii-cepae*. Mancozeb @ 0.25 percent and hexaconazole @ 0.06 percent were used as chemicals, along with *Trichoderma viride* (Tv1) and *T. harzianum* (Th-1), each at 1×109 spores ml^{-1} and phyto extracts (*Cannabis indica* @ 10% and *Curcuma longa* @ 10%). Mancozeb @ 0.25 percent proved to be the most successful in controlling onion foliar blight. Although both *T. viride* and *T. harzianum* were statistically equivalent, application of *T. harzianum* (Th^{-1}) led to less severe disease than *T. viride* (Tv^{-1}), and both were considerably better than the control. The use of the plant extracts *C. indica* and *C. longa* to treat disease was futile (Lu *et al*., 2008) [33].

Conclusion

Onion foliar diseases like Stemphylium leaf blight and others can reduce storage quality and yield. When using foliar fungicides, growers are concerned about the leaf dieback and the lack of disease suppression. It is crucial to comprehend this host-pathogen system and offer improved management measures if the region is to continue producing onions sustainably. The goal of the paper is to provide evidence for the following claims: confirmation of the identification of *S. vesicarium* isolates found in onion fields; confirmation of the insensitivity of *S. vesicarium* to commonly used fungicides; confirmation of seed-borne inoculum from naturally infected flowers; confirmation of the life cycle of *S. vesicarium* in the area; and proposal of a model to predict sporulation events.

Only little changes between cultivars and declining fungicide efficacy over the past ten years have been found in field research on cultivar resistance and onion. A mineral oil product that was evaluated over the course of a year and had potential to produce systemic resistance also failed to decrease SLB. It is obvious that additional study is required to give producers new strategies to combat this disease.

Because onion leaf debris can serve as a major inoculum the following spring, gardeners are advised to remove or bury it from the field based on the findings of this article. To lower the possibility of inoculum from alternate hosts, hand-pulled weeds should also be removed from the field and completely composted or buried far from onion fields. Azoxystrobin, pyrimethanil, fluopyram, and difenoconazole-containing fungicides are no longer helpful at reducing the symptoms of SLB, hence using these products is no longer advised. If use of pyrimethanil, fluopyram, and difenoconazole is stopped, it is possible that sensitivity will reappear. However, azoxystrobin insensitivity is likely to persist in the population for perpetuity. Additionally, it is advised to adhere to suggestions for managing resistance, such as switching between fungicides with various modes of action and avoiding the first application of a foliar FRAC group 7 fungicide if the seed has already had efficient treatment.

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