

## Revisão Bibliográfica

**Etiology, epidemiology, and management of white rot on onion and garlic: current knowledge and future directions for Brazil****Etiologia, epidemiologia e manejo da podridão-branca em alho e cebola: conhecimento atual e perspectivas futuras para o Brasil****Valdir LOURENÇO JR<sup>1</sup>; Bruno Sérgio VIEIRA<sup>2</sup>; Everaldo Antônio LOPES<sup>3</sup>; Oscar N. VILLALTA<sup>4</sup>**

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**Abstract**

*Stromatinia cepivora* (= *Sclerotium cepivorum*), causal agent of the white rot, is a major soilborne pathogen that attacks garlic (*Allium sativum* L.), onion (*Allium cepa* L.), and other plants of the *Alliaceae* family. The pathogen is difficult to control because it survives as sclerotia for decades in soil. White rot can cause total crop losses when sclerotia levels are high in soil and environmental conditions favorable for disease development. Aspects of the biology and epidemiology of *S. cepivora* have been investigated extensively worldwide. These studies have provided essential information to develop different control strategies. Currently, white rot management is based primarily on the application of fungicides and biocontrol agents to protect the crop against infection, and the use of natural and synthetic germination stimulants of sclerotia, soil fumigation and solarization to reduce sclerotia density in soils. In Brazil, few studies have been conducted to understand white rot epidemiology and effectiveness of control measures currently available, despite the disease being an economically important in garlic and onion production regions for many years. This review provides updated information on the biology of *S. cepivora*, and epidemiology and control of white rot to identify important knowledge gaps and future research directions for white rot in Brazil.

**Additional keywords:** integrated control; mycology; population biology; *Stromatinia cepivora*.

**Resumo**

*Stromatinia cepivora* (= *Sclerotium cepivorum*), agente causal da podridão-branca, é um dos principais fitopatógenos que infectam alho (*Allium sativum* L.), cebola (*Allium cepa* L.) e outras espécies de aliáceas. O patógeno é de difícil controle por sobreviver por décadas no solo, na forma de escleródios. A podridão-branca pode causar perdas totais de produção quando há alta quantidade de escleródios no solo e condições ambientais favoráveis para o desenvolvimento da doença. Aspectos da biologia e da epidemiologia de *S. cepivora* têm sido investigados extensivamente em todo o mundo. Estes estudos têm fornecido informações essenciais para o desenvolvimento de diferentes estratégias de controle. Atualmente, o manejo da podridão-branca é baseado no uso de fungicidas e agentes de controle biológico para proteger as culturas contra infecção, além do uso de estimulantes naturais e sintéticos de germinação de escleródios, fumigação e solarização do solo para reduzir a densidade de escleródios no solo. No Brasil, poucos estudos foram conduzidos para entender a epidemiologia da podridão-branca e a eficiência das medidas de controle disponíveis, apesar de a doença ser economicamente importante em regiões de produção de alho e de cebola. Esta revisão fornece informações atualizadas sobre a biologia de *S. cepivora*, além da epidemiologia e do controle da podridão-branca para identificar lacunas de conhecimentos relevantes e direções futuras de pesquisa para a podridão-branca no Brasil.

**Palavras-chave adicionais:** biologia de populações; manejo integrado; micologia; *Stromatinia cepivora*.

## Introduction

White rot, caused by *Stromatinia cepivora* (Berk.) Whetzel (1945) (Synonymy: *Sclerotium cepivorum* Berk.), is a devastating soilborne disease of onion (*Allium cepa* L.) and garlic (*Allium sativum* L.) crops in several parts of the world. The disease has been reported in many countries including: Argentina (Camiletti et al., 2016), Australia (Porter et al., 1991; Duff et al., 2001; Villalta et al., 2012), Brazil (Pinto et al., 1998), Canada (Couch & Kohn, 2000), Iran (Saremi et al., 2010), Mexico (Ponce-Herrera et al., 2008), New Zealand (Tyson et al., 2002), Netherlands, Spain, Switzerland, United Kingdom (Earnshaw & Boland, 1997), and USA (Davis et al., 2007).

The fungus, which survives as sclerotia in soil, is restricted to *Allium* species, however, it causes the most losses on onion and garlic plants (Crowe, 2008). Yield losses commonly range from 5-50%, but total losses have been reported under high amount of sclerotia and favorable environmental conditions for disease development (Utkhede, 1982). The disease can have a negative effect on growers' income due to the high cost associated with bringing a crop of onion or garlic.

The management of white rot is difficult because *S. cepivora* survives as dormant sclerotia in soil for decades, and little or no sclerotia germination occurs in the absence of host plants (Utkhede, 1982; Coley-Smith & Parfitt, 1986). Sclerotia germination is mostly triggered by specific sulfides compounds exudated from roots of *Allium* crops (Coley-Smith & Parfitt, 1986). Although crop rotation can reduce the population of soilborne plant pathogens in soil, however, this strategy is inefficient to white rot due to the long persistence of the fungus (Utkhede, 1982). Once established, *S. cepivora* has been difficult to eradicate with field treatments developed so far (Fuga et al., 2012).

In Brazil, white rot is one of the most important, widespread, and destructive plant diseases on onion and garlic crops (Fuga et al., 2012). It is prevalent in garlic and onion production areas in the states of Rio Grande do Sul, Santa Catarina, Paraná, São Paulo and Minas Gerais (Pinto et al., 1998; Fuga et al., 2012; Reis & Oliveira, 2013). Some growers have been forced to abandon garlic and onion production in traditional growing regions of Minas Gerais and São Paulo states due to high levels of sclerotia of *S. cepivora* in soils (Pinto et al., 1998; Fuga et al., 2012; Reis & Oliveira, 2013).

Currently, the main strategy used in Brazil to prevent the introduction of the fungus into farms, and disease spread within farms, includes restricting the movement of equipment, infected plant materials, and grazing animals (Reis & Oliveira, 2013). However, many growers do not adopt these measures and many often acquire contaminated garlic bulb seeds (Pinto et al., 1998; Dusi et al., 2009; Fuga et al., 2012; Reis & Oliveira, 2013). Although fungicides are often used to

control white rot in many fields, the knowledge on disease epidemiology and management required to optimize fungicide application is limited, with only a few studies on epidemiology and management carried out in Brazil (Pinto et al., 1998; Fuga et al., 2012; Fuga et al., 2016).

In contrast, the biology of *S. cepivora* and disease epidemiology and management have been investigated extensively in many countries affected by white rot. This research has provided the information needed to develop a range of chemical and non-chemical control methods to manage the disease that include fungicides, biocontrol agents, natural and synthetic germination stimulants of sclerotia, and soil fumigation and solarization (Ulacio-Osorio et al., 2006; Crowe, 2008). None of these methods has been properly evaluated in Brazil.

The objective of this review is to synthesize information available from *S. cepivora* biology, and disease epidemiology and management to highlight important gaps in knowledge of white rot of garlic and onion in Brazil. The review examines recent developments in disease control and novel technologies and discusses their potential integration into a future white rot management strategy for Brazil.

## The pathogen

### Etiology

White rot is caused by the fungus *Stromatinia cepivora* (Berk.) Whetzel (1945) (Synonymy: *Sclerotium cepivorum* Berk.), an ascomycete that belongs to the subphylum Pezizomycotina, class Leotiomycetes, order Helotiales, and family Sclerotiniaceae (Kirk et al., 2010; Xu et al., 2010; Fuga et al., 2012). Although Xu et al. (2010) determined that the fungus belongs to the family Sclerotiniaceae, the generic placement of this species is still uncertain (Farr & Rossman, 2017).

*Stromatinia cepivora* infects only species of the genus *Allium* such as onion, garlic, leeks, and chives (Entwistle, 1990). The fungus produces sterile mycelium and sclerotia, which are small, brown to black, and uniformly round (0.35 – 0.50 mm in diameter). They have a stromatic mantle composed of layers of pigmented and thickened cells with an inner medulla composed by polysaccharide hyphae and protein bodies (Backhouse & Stewart, 1987). Melanin present in the stromatic mantle is responsible for the dark color acting as a defense to desiccation (Willetts & Bullock, 1992). Sclerotia are easily produced around 10 days in potato dextrose agar (PDA) at 18 °C and are approximately equal in size measuring between 200-600 µm in diameter (Backhouse & Stewart, 1987). Primers developed by Haq et al. (2003) are useful in diagnostics and epidemiological studies of *S. cepivora*.

As sclerotia are constitutively dormant for several weeks or months, these propagules can survive for up to 20 years in soil without the presence of a host (Coley-Smith et al., 1990; Valle & Aguilar 2004;

Maude, 2006). The germination of sclerotia is triggered by exudation of alkyl and alkenyl-sulphoxides from *Allium* roots which are metabolized by soil microorganisms to produce volatile thiols and sulphide (Coley-Smith, 1960; Coley-Smith & King, 1969). The role of sulphoxides or their breakdown of products in promoting germination of dormant sclerotia in soil provided the idea for the use of synthetic *Allium* oil as a tool for disease management (Coley-Smith & Parfitt, 1986).

### Reproduction and variability

There is no report of sexual reproduction in the population of *S. cepivora* (Xu et al., 2010). Thus, it is expected that the population is clonal with low genetic variability. Since the teleomorphic phase (sexual) has not yet been described (Xu et al., 2010), it is theorized that the population of *S. cepivora* is clonal with low genetic variability.

Genetic variability has been investigated by determining mycelial compatibility groups (MGCs) in a population of 146 isolates from Canada (Ontario) and 23 isolates from other regions or countries (Australia, England, Netherlands, New Zealand, Spain, and Switzerland) (Earnshaw & Boland, 1997). The genetic variability was low among isolates from Canada with only two MGCs detected, but higher (six MGCs) among isolates from other regions.

In another population biology study, around 200 isolates from Canada, New Zealand, Italy, Netherlands, Chile, Mexico, and Brazil were characterized using MGCs, DNA fingerprints, and DNA sequence polymorphisms from EF-1a gene and five anonymous genomic regions (Couch & Kohn, 2000). Several isolates from different regions clustered in the same MGC and multilocus haplotypes supporting the theory of clonal dispersion. Although phylogenetic analysis provided evidence of recombination in the evolutionary history of *S. cepivora*, the fungus has most likely disseminated clonally in Canada and New Zealand. Interestingly, the only Brazilian isolate used in the study is similar with some isolates from Chile, Italy, Mexico, and New Zealand (Couch & Kohn, 2000).

The genetic variability of *S. cepivora* population was also analyzed in New Zealand where white rot causes significant crop damage and economic loss (Tyson et al., 2002). Two hundred and thirty-one isolates of *S. cepivora* from onion and garlic production areas were studied using mycelial compatibility, UP-PCR, and RAPD analysis. A representative sample of 30 isolates were selected and then paired in all combinations on PDA to identify compatible strains. Representative isolates identified in the initial pairings were then paired with the remaining isolates from New Zealand, and with 25 isolates collected from Australia, Brazil, Canada, Germany, Netherlands, Spain, Switzerland, and United Kingdom. Most of isolates from different regions and host (onion and garlic) were clustered in the MGC 1, 5, and 8. Six groups were found based on the combination of UP-PCR primer L15 and RAPD primer OPAX15. Thus, the genetic

diversity is presumed to be low in the population of *S. cepivora* in New Zealand (Tyson et al., 2002).

The results from studies so far support the theory that the population of *S. cepivora* is clonal with unknown sexual state. As there is little information about the population biology of *S. cepivora* in Brazil, it is important to study its genetic diversity to determine if evolutionary processes have affected the fungus genetics and white rot epidemiology in tropical and sub-tropical climates of Brazil.

### Infection and symptoms

*Allium* plants may be infected at any stage of growth when sclerotial germination is triggered by volatile sulphur compounds associated with host plant root exudates and environmental conditions are favorable for infection: damp soil and temperature at the range of 13-18 °C (Crowe & Hall, 1980a; Fuga et al., 2012). In southern latitudes, the optimum soil temperature for sclerotial infection occurs normally during the winter months of June, July, and August in sub-tropical regions like S.E. Queensland (Australia) and Rio Paranaíba (Minas Gerais, Brazil), and during autumn and spring in temperate regions. Soil temperatures favorable for mycelial infection (i.e. plant to plant infection) range from 9 to 21°C (Utkhede, 1982). In vitro, incubation at 20°C in darkness is optimal for sclerotia germination (Marcuzzo & Luiz, 2017).

Infection of seedlings occasionally occurs. However, the first infection is normally detected in plants with three to five leaves. Initial stages of infection are limited to the host root system and base plate (Crowe & Hall, 1980a; Massola Junior et al., 2016). The most easily recognizable symptom is the yellowing and dieback of the leaves beginning at the tips and progressing downward followed by death of the affected leaves (Massola Junior et al., 2016). A gradual decline in the plant continues for some days or weeks and in the case of young plants may constitute a rapid wilt and collapse of aerial parts that leads to death of the host (Entwistle, 1990; Massola Junior et al., 2016). Due to the death of the roots the diseased plants are easily detached from the soil. On underground parts, the fungus itself is visible as superficial and fluffy white mycelium (Entwistle, 1990).

The bulb presents soft rot and white mycelial growth is observed at the base of the plant associated with small black sclerotia giving a blackened appearance to the bulbs (Valle & Aguilar, 2004; Massola Junior et al., 2016). Above-ground symptoms are not normally evident until the pathogen has colonized and partially rotted the stem and leaf sheaths. Roots often extend horizontally, providing a direct path for mycelial growth to nearby plants. Infected plants therefore tend to occur in clusters from a few up to 40 or more adjacent plants (Crowe & Hall, 1980a). The pathogen may continue to decay bulbs in storage if adequate temperature and humidity is maintained (Schwartz & Mohan, 1995).

Levels of sclerotia in soil and secondary infections are factors that contribute to increased disease incidence and severity (Delgadillo et al., 2002). Soil tests have been used to determine the risk of white rot based on the levels and distribution of sclerotia in soils. However, sclerotia levels determined with soil tests can be unreliable indicators of disease risk because levels can be highly variable in a field, and sclerotia buried deeper in the soil profile are not detected by topsoil sampling (Crowe & Hall, 1980b; Ryley, 1995).

## Control

Three key strategies are used to control onion and garlic white rot. The first is minimize the introduction and spread of the pathogen within and between farms, which is the responsibility of growers who must implement and maintain effective on-farm hygiene practices. *S. cepivora* once established has been difficult to eradicate with available cost-effective field treatments. The use of non-infested planting materials, accurate mapping of hot spots, roguing, and cleaning equipment are important approaches to limit the spread of the fungus from infested areas to clean areas. Efforts should therefore be dedicated to education, not only to raise awareness of the problem, but to increase collaboration between pathologists and industry. The second is to reduce the population of sclerotia in the soil; and the third is to protect the growing crop against infection by reducing disease progress. Currently, the main tactics to manage white rot include cultural, physical, chemical, and biological techniques.

## Cultural and physical control

Cultural practices (crop rotation, planting date, biofumigation, deep cultivation) and physical methods of control (solarization and flooding) have been effective in reducing the population of sclerotia in soils and disease incidence. Since *S. cepivora* can survive for decades in the field, it is important to demarcate areas infested by the fungus to minimize transit of machines and workers on hot spots and to clean carefully the equipment used in infested fields. Diseased plants must be removed from the field and destroyed, especially when the hot spots are still small.

The planting date is a cultural practice that can be used to aid the host evade the pathogen. The effect of different planting times on the incidence of white rot on garlic cultivars was evaluated in the region of Amarantina and Ouro Preto in Brazil (Pinto et al., 1998). Early planting of garlic in February reduced the incidence of white rot and yield losses on the cultivars Amarante, Cateto Roxo, and Centenário. The warm temperatures above 20°C prevailing in the February planting hampered the germination of *S. cepivora* and disease development during bulbification (Pinto et al., 1998).

The narrow host range of the fungus, limited to *Allium* species, would facilitate the adoption of crop

rotation for the management of the white rot. However, such a strategy is limited by the ability of the fungus to survive in the field as sclerotia. In infested areas, the population of *S. cepivora* can be reduced by solarization, biofumigation, flooding, the application of germination stimulants, and the incorporation of organic amendments into the soil.

Solarization is a soil disinfestation method developed in Israel (Katan, 1981) and it has been used in many countries for the management of pathogens, pests, and weeds. In this method, the soil remains covered by transparent plastic during the periods of the year with high incidence of solar radiation (Katan & Gamliel, 2011). Due to the greenhouse effect under the plastic cover, the temperature of superficial layers (0 to 20 cm depth) in solarized soil usually range from 35 to 60 °C during the warmest periods of the year (DeVay, 1991). However, the temperature and the efficiency of the control decreases with depth in soil profile (Katan & Gamliel, 2011), which means that soil must be kept covered for longer periods. The soil usually remains covered for four to eight weeks (Katan & Gamliel, 2011).

An interesting approach to maximize reduction of viable sclerotia in soil profile is to plow the soil after solarization and to solarize the field again. The most used plastic films range from 40 to 150 µm. Thin films (25 to 30 µm) tend to tear easily. The thicker ones are more expensive; however, they can be reused (150 to 200 µm). Harrowing, plowing, removal of sharp objects, and irrigation to field capacity are required steps before covering the soil with plastic. The water in the soil activates pathogen propagules and enhances heat conduction. The borders of the plastic should be buried to avoid heat loss (Katan & Gamliel, 2011).

Solarization reduces *S. cepivora* population in the soil due to the deleterious effect of the heat and the action of the antagonistic microorganisms (Porter & Merriman, 1983; Satour et al., 1989; Cunha et al., 1993; Melero-Vara et al., 2000; McLean et al., 2001; Prados-Ligero et al., 2002). In New Zealand and Spain, solarization for eight weeks using transparent film of 40 or 50 µm reduced the number of viable sclerotia in the soil by more than 90% (McLean et al., 2001; Prados-Ligero et al., 2002). Sclerotia recovered from solarized soils in New Zealand were colonized by the fungi *Trichoderma*, *Verticillium*, *Fusarium*, *Mucor*, and *Aspergillus*, and four non-identified bacteria species (McLean et al., 2001). Soil amendment with vermicompost (50 Mg ha<sup>-1</sup>), followed by solarization, is an integrated strategy for reducing inoculum density of *S. cepivora* (Pereira et al., 1996). Temperatures in vermicompost-amended soils are increased by 4 to 5 °C in comparison to non-amended soils (Pereira et al., 1996).

The microbial decomposition of certain organic amendments incorporated into the soil, especially residues from some species of Brassicacea and other green manures, releases volatile toxic gases during the degradation process of the organic matter. This pro-

cess can suppress pests and pathogens and it is known as “biofumigation” (Kirkegaard et al., 1998). For enhancing the efficacy of biofumigation, the soil must have sufficient moisture for intense microbial activity and the escape of volatile toxic compounds from the soil must be avoided. In this case, the soil may be covered with plastic immediately after crushing and incorporating the organic residues, or the topsoil may be compacted with rollers. Transparent plastic cover increases soil temperature and accelerates the degradation of the residues (Kirkegaard et al., 1998; Gamliel et al., 2000). The integration of biofumigation with solarization may have a synergistic effect on the control of *S. cepivora*, and the time that the soil remains covered may be reduced.

The residue of Brassicaceae (*Brassica* spp.) has been the most studied organic material for biofumigation, due to a range of toxic substances released during its decomposition. Brassica plants are rich in glucosinolates, which are hydrolyzed by myrosinase into degradation products, such as isothiocyanates and nitriles (Brown & Morra, 1997). Glucosinolates are atoxic, but isothiocyanates (ITCs) suppress *S. cepivora* and other soilborne pathogens (Stapleton & Duncan, 1998; Smolinska, 2000; Coventry et al., 2002; Coventry et al., 2006; Ulacio-Osorio et al., 2006; Villalta et al., 2016). Biofumigation using broccoli, mustard, and rapeseed reduces the viability of *S. cepivora* sclerotia (Smolinska, 2000; Ulacio-Osorio et al., 2006). The concentration of 2-propenyl glucosinolate or sinigrin, a precursor of ITC and other sulfur compounds, in Brassica biofumigants is correlated with their efficiency on the suppression of soilborne pathogens (Villalta et al., 2016). Mustard (*Brassica juncea*) cultivars Nemfix® and Caliente 199®, which have high content of sinigrin (50 to 60  $\mu\text{mol g}^{-1}$  of shoot residues), showed greater suppression of *Sclerotinia minor*, *Rhizoctonia solani*, *Fusarium oxysporum*, *Pythium dissotocum* (Villalta et al., 2016), and *S. cepivora* (Villalta et al., unpublished data).

Flooding infested fields is also a management strategy of *S. cepivora* (Leggett & Rahe, 1985; Alexander & Stewart, 1994; Clarkson et al., 2004). Sclerotia germination depends on moderate soil moisture, but the sclerotia deteriorate under longer periods of soil moisture above field capacity (Clarkson et al., 2004). Degradation of sclerotia ranges from 87 to 97% at soil water potential of  $-9.40 \times 10^{-9}$  MPa (24% soil water content) after eight weeks at 20 °C (Clarkson et al., 2004). Water availability and the time required to maintain the soil flooded may limit the use of this technique for white rot management (Fuga et al., 2012).

Anaerobic soil disinfestation (ASD) is an ecological alternative to soil fumigation (Blok et al., 2000; Shinmura, 2000; Shinmura, 2004). It has been studied for the control of several soilborne pathogens, such as *Fusarium*, *Verticillium*, *Rhizoctonia*, *Sclerotinia*, *Pythium*, *Phytophthora*, *Macrophomina*, *Ralstonia*, and nematodes (Roskopf et al., 2014; Shennan et al., 2014; Shrestha et al., 2016). This technique consists of incorporating organic material that is easily decompos-

able (C:N ratio from 8-20:1) into the soil, irrigating to saturation and covering the soil with oxygen-impermeable plastic (Roskopf et al., 2014; Shennan et al., 2014). Carbon source stimulates rapid growth and respiration of soil microbiota, reducing available oxygen. Soil pore spaces filled with water and the plastic cover also enhance the creation of anaerobic conditions in soil (Roskopf et al., 2014; Shennan et al., 2014; Shrestha et al., 2016). Accumulation of toxic products from anaerobic decomposition (acetic, butyric and propionic acids, CO<sub>2</sub>, NH<sub>3</sub>, H<sub>2</sub>S, CH<sub>4</sub> and N<sub>2</sub>O), antagonism by anaerobic organisms, lack of oxygen and the combination of all of them are the main drivers that explain the efficacy of ASD (Runia et al., 2014; Shennan et al., 2014). Rice or wheat bran, soybean flour, ethanol, molasses, manure, and fresh crop residues have been assessed as carbon sources at rates ranging from 0.3 to 9 kg m<sup>-2</sup> (Shrestha et al., 2016). The incubation period has varied from 3 to 10 weeks (Shrestha et al., 2016). However, little is known about the effect of ASD on the management of the white rot. Further studies are required to assess the potential of this technique on the reduction of the density of viable sclerotia of *S. cepivora*.

Another potential control method is the application of compost extract and compost tea that act as a means of natural plant disease control by increasing microbial activity and diversity with populations of plant-beneficial and disease-antagonistic organisms in the soil. To evaluate the effect of these organic compounds on white rot management, a field experiment was conducted in an experimental area from Coopadap, naturally infested with the pathogen, at Rio Paranaíba, MG, Brazil. The aqueous and solid organic mixture were composed by fish food, rice bran, bone meal, wheat flour, corn meal, molasses, rock dust, and poultry manure. Both composts were applied and incorporated in soil before the onion planting. In addition, the compost tea was sprayed every 15 days. However, both composts did not reduce the white rot incidence on onion plants (unpublished data). As the soil organic matter was low in the experimental area, the microbial population with potential antagonistic activity to *S. cepivora* could be low in the experimental area. Therefore, the experiment should be repeated with the use of higher doses of compost tea and extracts and changes in microbial population diversity in soil and their effect on sclerotia investigated.

## Chemical control

### Fungicides

Growers have relied on fungicides for the control of onion and garlic white rot throughout the world. On onions, fungicides have been traditionally applied to the soil at planting, as in-furrow or fertilizer treatments, post-planting as soil surface sprays or after plant emergence as foliar/stem-based applications and as a combination of both planting and foliar applications. On garlic, they have been applied with the seed at planting, either as in-furrow sprays or as seed (garlic

clove) treatments, post-planting as foliar sprays and as a combination of seed and foliar sprays. Control with pre-plant treatments is difficult because fungicides must remain effective throughout the season. Below the zone of protection, the fungus may spread through the soil among roots and when the fungicide begins to degrade, the fungus may infect and/or kill plants if fungicide protection is not present (Crowe et al., 1980b). Fungicide not only protected plants against infection in early and late stages of growth, but also reduce formation of sclerotia at harvest period.

From the early to mid-1980s, the dicarboximides iprodione, and vinclozolin were commonly used for onion white rot control in New Zealand, United Kingdom, and Australia. By 1983, however, levels of disease in South Auckland, New Zealand, indicated that these fungicides did not provide satisfactory disease control, and similar results were obtained in field trials in 1984-1985 on Patumahoe clay loam soil of South Auckland (Fullerton & Stewart, 1991; Stewart & Fullerton, 1991). In Australia, these fungicides only protected the surface roots and most plants still developed disease prior to harvest and yields were poor (Merriman & Porter, 1984). Later studies suggested accelerated degradation by soil microbes was the most likely cause of control failures with iprodione and vinclozolin in New Zealand (Slade et al., 1992) and United Kingdom (Walker et al., 1986).

Field trials in Australia and New Zealand from 1984 to 1989 showed that the dicarboximide procymidone and demethylation inhibiting (DMI) fungicides tebuconazole and triadimenol (triazole group) were effective treatments for controlling onion white rot and alternatives to iprodione and vinclozolin (Merriman & Porter 1984; Porter et al., 1991; Stewart & Fullerton 1991; Fullerton et al., 1995). Procymidone considerably reduced onion white rot incidence and severity in many trials but their effectiveness was dependent on the method of application, soil conditions and levels of sclerotia in soil, but onion seed treatments with procymidone and tebuconazole were phytotoxic (Porter et al., 1988; Stewart & Fullerton 1991; Porter et al., 1991; Fullerton et al., 1995). Although procymidone was more stable than iprodione and vinclozolin in New Zealand and Australian soils (Slade et al., 1992), the product eventually lost efficacy and/or was withdrawn from the market due to residue concerns. Loss of efficacy by tebuconazole for onion white rot control was first noted at Pukekohe, New Zealand, in 1996-97, where it was applied as multiple spray applications after crop emergence (Fullerton, 2005).

The efficacy of procymidone and tebuconazole as fungicide-fertilizer treatments for onion white rot control were investigated in trials in Australia. Procymidone applied with bands of fertilizer 2 or 5 cm below the seed did not reduce onion white rot incidence in soil with high levels of sclerotia (Porter et al., 1991), probably because it did not move into the surface soil to provide young onion plants with a continuous layer of treated soil. Onion growers in Tasmania

have relied since the 1980's on the use of tebuconazole-lime super applied at sowing for white rot control, but its effectiveness has been affected by many issues including inoculum levels in soils, carrier characteristics and residual activity (Pung et al., 2008).

From the fungicides tested for garlic white rot control before 2001, tebuconazole has been one of the most effective when applied to the garlic clove (Duff et al., 2001; Jackson et al., 1997) or as a combination of soil and stem-based sprays (Delgadillo et al., 2002; Melero-Vara et al., 2000). As garlic seed (cloves) treatment, four rates of tebuconazole (0.25, 0.5, 0.75, and 1.0 L ha<sup>-1</sup>) provided significant superior control than five rates of procymidone (0.5, 1.0, 1.5, 2.0, and 2.5 L ha<sup>-1</sup>) in volcanic soil under high disease in Southern East (SE) Queensland, with the 0.75 and 1.0 L ha<sup>-1</sup> of tebuconazole providing the highest control compared to the control, which had less than 35% of plant survival (Duff et al., 2001). Melero-Vara et al. (2000) showed that dipping garlic cloves in tebuconazole (0.25 mL of the active ingredient per L of water for 5 min) reduced the incidence of plant death from 17.2% (untreated) to 4.1% in a soil with 3-6 viable sclerotia per kg soil, with a corresponding significant yield increase, but the seed treatment was not effective in another trial with more sclerotia (>10 per kg of soil), while combining the seed treatment with tebuconazole spraying of garlic stem bases at 10-days interval resulted in the best control (1.5% incidence). The researchers suggested the seed treatment protected plants against early infections and stem-based sprays supplemented the protection.

Research after 2001 confirmed triadimenol and tebuconazole can be effective fungicides for control of white rot on onion and garlic in different soils (Mueller et al., 2005; Zewide et al., 2007; Pung et al., 2008; Villalta et al., 2008) and spring onions in sandy soil (Villalta et al., 2012). The research also evaluated the efficacy of newer fungicides that were available in the domestic markets. Tebuconazole (430 g of the active ingredient per hectare), mixed with fertilizer (e.g. lime super 100-150 kg ha<sup>-1</sup>) drilled below the seed, significantly reduced onion white rot incidence in two trials from 53-60% to 38-33%, with corresponding yield increases in Red Ferrosol soils with 0-10 sclerotia per kg soil (Pung et al., 2008) and from 61% to 9.5% in a volcanic soil with 78-80 sclerotia per kg of soil in Southern East (SE) Queensland, but failed at another trial with >300 sclerotia per kg of soil in the same region (Villalta et al., 2008). In the two previous trials, researchers observed plants with infected roots and bulbs but without above ground symptoms indicating tebuconazole residual activity was low at the end of the season.

The usefulness of the seed treatment with tebuconazole was demonstrated in soils infested with >20 sclerotia per 500 g<sup>-1</sup> of soil in four trials over two seasons (2003-2005) at two locations in Ethiopia (Zewide et al., 2007). From the other four seed treatments tested comprising four protectant fungicides

(captan, benomyl, mancozeb, and thiram), only captan provided disease reductions similar to that provided by tebuconazole. In a trial in California with 4-15 sclerotia per 500 g of soil, tebuconazole, applied as a seed treatment or in furrow, significantly reduced disease severity from 4.0 (ie 10 = all plants dead) in untreated plots to 1.7-2.0, with corresponding significant garlic yield increases, while cyprodinil+fludioxidil or dicloran, applied in furrow also gave control comparable to tebuconazole (Mueller et al., 2005). Foliar applications (5x) with either cyprodinil+fludioxidil, iprodione, pyraclostrobin+boscalid or dicloran made in spring using low water volumes (27-30 GPA) did not provide any substantial disease control or yield benefits probably due to poor penetration into the root zone.

Currently, there are only a few effective fungicides for white rot control around the world. In New Zealand, for example, fungicidal protection on onions is achieved by sequential applications of foliar sprays of triadimenol in alternation with newer fungicides including pyraclostrobin+boscalid which have replaced tebuconazole (Fullerton, 2005). In the USA, fungicides recommended for garlic white rot control include tebuconazole and pyraclostrobin+boscalid (www.ucdavis.com). Newer fungicide tends to be more expensive than old fungicides, so they must be very efficacious to be considered in control programs. Optimizing fungicide application is thus a continuous research priority to improve their effectiveness. Research is required in Brazil to develop fungicide treatments and optimize application supported by new epidemiological knowledge. Currently, thiophanate-methyl, procymidone, and iprodione are the only fungicides registered in Brazil for white rot control (Brasil, 2018).

Disease control measures must initiate before infection occurs to achieve best control because root infections can spread gradually from root branches onto the stem base of bulbs and once the fungus reaches the base of the bulbs, the infected plants become wilted due to extensive root death and bulb decay. Control, both pre and post-emergence, can be enhanced by applying measures before soils temperatures are conducive for *S. cepivora* germination and infection of roots. This has been achieved by monitoring soil temperature and crop development. Fungicide field trial data in the Pukekohe region of New Zealand from 1989 to 2006 revealed correlation between effectiveness of fungicide programmes and cumulative average daily soil temperatures above a base temperature or white rot degree days, with disease normally occurring when degree days reached 250 and later start times associated with a consistent drop in programme effectiveness (Tyson et al., 2008).

Fungicides do not provide a long-term solution to the white rot problem because they do not directly kill sclerotia and control is difficult in soils with high levels of sclerotia even with effective fungicides (Villalta et al., 2008). Chemical soil treatments are required to reduce sclerotia numbers to levels below which fungicidal protection is possible and to restore infested fields

for sustainable onion or garlic production.

### Chemical soil treatments

Reduction of sclerotia and disease control has been achieved with the soil fumigants metham-sodium (Adams & Johnston, 1983; Perez-Moreno et al., 1996) and methyl bromide (Davis et al., 2007), and the biostimulant of sclerotial germination diallyl disulphide (DADS) applied before planting onion and garlic crops (Merriman et al., 1980; Coley-Smith & Parfitt, 1986; Tyson et al., 2000; Dennis, 2001; Hovius & McDonald, 2002; Villalta et al., 2012). None has provided complete eradication of sclerotia in soil or control of white rot when applied on their own and their efficacy has been affected by many factors including application method and soil conditions.

Application of metham sodium has been the predominant means of soil disinfestation to manage white rot due to its low cost compared to other soil fumigants. Effective control of white rot using shunk-applied metham sodium has been demonstrated in some situations allowing the production of a single *Allium* crop after treatment; however, positive results have been erratic and retreatment sometimes necessary, with enhanced bio-degradation linked to the loss of effectiveness (Warton et al., 2001) and the occurrence of resistant strains to fungicides (Gullino et al., 1983). In two garlic trials in California, sclerotia mortality with methyl bromide was over 90% in soil with 50-80 sclerotia/500 g of soil, and this significantly improved garlic root health and increased yields in crops planted about a year after soil treatment (Davis et al., 2007). Dimethyl disulphide (DMDS) also has been shown to destroy 90% of dormant resting structures of various pathogens including *S. cepivora*, *Sclerotinia*, *Rhizoctonia*, and *Phytophthora* species (Charles, 2006). Soil fumigants are expensive and more cost-effective for treating small areas infested.

The specific response of sclerotia to sulphoxides or their breakdown products (thiols and sulphides) suggested a potential use for these compounds as artificial germination stimulants for white rot control. They have been applied pre-planting into fallow soil to promote germination of sclerotia when soil temperatures and moisture conditions are optimal for sclerotial germination. Mycelium from germinated sclerotia eventually dies out after failing to infect *Allium* hosts and is also susceptible to hyperparasitism and lysis (Stewart & McLean, 2007; Fuga et al., 2012). Early attempts to use artificial onion oils, plant extracts, and distilled *Allium* oils as pre-planting soil treatments to reduce the number of viable sclerotia in soil probably failed due to inadequate application methods, low concentrations of key germination stimulants in products tested or products were too expensive for growers to use (Merriman et al., 1980).

Diallyl disulphide (DADS), the key sulphide biostimulant produced by *Allium* species, can be produced synthetically, in higher concentration and more cheaply, from petroleum distillation (Coley-Smith &

Parfitt, 1986). The application of new formulations of DADS to fallow soil, under favorable soil temperature and moisture levels for sclerotia germination, effectively reduced sclerotia populations in trials in New Zealand (Tyson et al., 2000), Canada (Hovius & McDonald, 2002), Australia (Villalta et al., 2008; Villalta et al., 2012), and in the United States (Davis et al., 2007). In trials under low-to-moderate disease pressure, DADS treatments provided significant disease control in subsequent onion and garlic crops, sometimes exceeding 90% (Tyson et al., 2000; Davis et al., 2007; Villalta et al., 2008) and similar to methyl bromide fumigation (Davis et al., 2007). In some cases, however, levels of sclerotia remaining after soil treatment caused unacceptable yield losses in garlic (Davis et al., 2007) and onion crops (Villalta et al., 2008).

Repeated application of DADS over two seasons or years has provided more effective control than with single applications, and DADS integrated with fungicides has improved disease control. In two garlic trials in California (1998-99 and 1999-2001), DADS applied at 0.5 mL m<sup>-2</sup> was as effective as methyl bromide in reducing the population of sclerotia in soil by more than 90%, but despite the efficacy of DADS treatments, the levels of sclerotia remaining in treated soil (i.e. <10 sclerotia per 500 g soil) still caused substantial root rot and yield losses in subsequent garlic crops planted a year later (Davis et al., 2007). In an onion trial in New Zealand (1999-2000), two applications of DADS (Alli-up<sup>®</sup>), the first in spring and the second in autumn, at a rate of either 5 or 10 L ha<sup>-1</sup>, significantly reduced onion white rot incidence from 42.2% (untreated) to 12-15%, with corresponding yield increases (Tyson et al., 2000).

In southern Australia, two pre-plant applications of either 5 or 10 L per ha<sup>-1</sup> (Alli-up<sup>®</sup>, 78% diallyl disulphide), applied to sandy soil during autumn and spring (2002-2003), were more effective than a single application, significantly reducing inoculum from 51 to 0-1.4 sclerotia per kg soil and subsequently disease incidence from 34% to 2-6% in a spring onion crop, with the DADS dual treatment integrated with procymidone providing the best control (0.1-0.4% incidence) (Villalta et al., 2012). In northern Australia, DADS (10 L Alli-up<sup>®</sup> per ha<sup>-1</sup>), applied twice in winter 2003 and again in 2004 significantly reduced sclerotia from 78-80 to 3-8 sclerotia per kg dry soil and subsequently onion white rot incidence from 61 to 11% in a subsequent onion crop grown in a volcanic soil (2005), with DADS integrated with the tebuconazole-fertilizer treatment reducing disease to 1.3% (Villalta et al., 2008). However, DADS dual treatments alone and in combination with fungicides were ineffective in controlling onion white rot in volcanic soil with >300 sclerotia per kg of soil (Villalta et al., 2008).

Despite the cost-effectiveness of synthetic DADS, its adoption has been affected by the lack of consistent supply, high cost of registration, logistics of field application and cost of treatment. Evaluation of alternative sources of biostimulant compounds in garlic

products (extracts, powder, oil) and onion compost has therefore been a research priority. Soil amendment with garlic extract (juice), garlic powder, garlic oil, and onion compost, applied under the conditions described earlier, have stimulated the germination of *S. cepivorum* sclerotia and reduced inoculum density (Crowe et al., 2000; Coventry et al., 2002; Davis et al., 2007; Villalta et al., 2008, Fuga et al., 2012).

Garlic products have effectively reduced sclerotia populations in the soil, achieving reductions statistically similar to DADS (Davis et al., 2007; Villalta et al., 2008) and methyl bromide fumigation (Davis et al., 2007). The content of key sulphides in garlic sources varies depending on the quality of the product and this and rates tested greatly influences the biostimulant effect. For instance, garlic powder at 112 and 224 kg ha<sup>-1</sup> was as effective as synthetic diallyl disulphide at 0.5 mL m<sup>-2</sup> in reducing the population of sclerotia in soil and garlic white rot incidence in one of three trials in California (Davis et al., 2007). Earlier results from trials in Oregon (1998-99) indicated that 90-100 g/m<sup>3</sup> of food grade dehydrated garlic powder, may be the minimum amount to achieve >90% population reduction (Crowe et al., 2000). Composted onion waste also can be used for decreasing the inoculum density of soil infested with *S. cepivora* (Coventry et al., 2002).

No DADS-based product is available for use in Brazil. Research is needed in Brazil to evaluate sulphur-containing soil amendments to optimize their dosages and application methods under a range of inoculum density and local soil conditions. Repeated application of these products over two or more years may be necessary, depending on initial levels of sclerotia in soil and concentrations of biostimulant compounds, to reduce sclerotia to levels below which sustainable control and profitable crop production is possible (Villalta et al., 2008; 2012).

### Biological control

Researchers around the world have prospected potential microorganisms for the biological control of white rot to increase the number of tools for disease control and reduce the use of fungicides (Clarkson et al., 2002; Stewart & McLean, 2007). After sclerotia germinate, the *S. cepivora* fungus produces hyphae which penetrate the roots of the host nearby - up to 2 cm from sclerotia (Crowe & Hall, 1980b; Fuga et al., 2012). The inactivation or death of pathogen sclerotia and mycelium can reduce disease incidence in the field. Biological control can occur naturally (eg. suppressive soils) or it can result from the addition of biological control agents (BCAs). Biocontrols may exert antagonistic pressure on pathogens including hyperparasitism and synthesis of antimicrobial compounds (Stewart & McLean, 2007). Occasionally, BCA induce systemic resistance (ISR) and promote plant growth (Zhou & Paulitz, 1994; Vallad & Goodman, 2004).

Biocontrol of white rot has been predominantly

attempted using BCAs. *Chaetomium globosum*, *Coniothyrium minitans*, *Trichoderma hamatum*, *T. harzianum*, *T. koningii*, and *T. virens* are examples of fungi that have demonstrated potential to control *S. cepivora*, by inhibiting growth and causing deterioration of sclerotia (McLean & Stewart, 2000). These antagonists can be used in the seed or soil (McLean & Stewart, 2000).

In New Zealand and Australia, *T. atroviride* TC52, a proven antagonistic (mainly via nutrient competition in the root zone) of *S. cepivora*, formulated in pellets (Onionmate® and Tenet®), has been used alone or in conjunction with chemical input to control onion white rot (Villalta et al., 2008; McLean et al., 2012). The performance of *T. atroviride* is, however, influenced by the timing of disease onset, rate and disease pressure (Villalta et al., 2008; McLean et al., 2012a). For instance, application of Onionmate® alone or combined with pre-plant DADS at 10L/ha or the tebuconazole-fertilizer treatment showed potential for disease control at a site with low disease pressure (<39% incidence), but not at a site with higher disease (Villalta et al., 2008). Although integrated application of germination stimulants with fungicides or *Trichoderma* species enhances the management of the white rot (Coventry et al., 2006; Villalta et al., 2008), the effect of these and other farm amendments and inputs such as pesticides, on the activity of the biocontrol agent must be elucidated to ensure they do not affect the establishment and activity of biocontrols (McLean & Stewart, 2000; McLean et al., 2005). As reported by McLean et al. (2012b), DADS and urea reduce the ability of *T. viride* TC52 spores to germinate and establish in soil. Therefore, the biocontrol agent must be applied after four weeks of soil treatment with DADS and do not applied together with nitrogenous fertilizers and incompatible fungicides (McLean et al., 2012b).

The action of biocontrol agents on the death or degradation of *S. cepivora* sclerotia occurs mainly by the production of toxic compounds and / or hyperparasitism. *Trichoderma* and *Bacillus* produce enzymes, antibiotics, volatile compounds, and acids that inhibit the development of plant pathogens (Cawoy et al., 2011; Silva et al., 2011; Olmedo-Monfil & Casas-Flores, 2014). Bacteria of the genera *Bacillus* are known to produce various microbial compounds (Cawoy et al., 2011); and approximately 4-5% of the *B. subtilis* genome, for example, is composed of genes associated with antibiotic synthesis (Stein, 2005). On the other hand, *Trichoderma* species are more commonly associated with colonization and degradation of resistance structures of pathogens (Dennis & Webster, 1971; Olmedo-Monfil & Casas-Flores, 2014). Exoglucanases, endoglucanases, cellobiases, and chitinases, secreted by *Trichoderma*, degrade and cause lysis of the cell wall from fungal plant pathogens (Papavizas, 1985).

In the United Kingdom, two isolates of *T. viride* and one isolate of *T. pseudokoningii* degraded up to 80% of sclerotia from four *S. cepivora* isolates in clay

soil (Clarkson et al., 2004). *T. harzianum* C4 strain applied as a colonized wheat bran in-furrow before planting (25 g m<sup>-1</sup>) delayed the incidence of the disease for 40 days in Mexico (Avila-Miranda et al., 2006). In Canada, the application of *Glomus intraradices* to onion decreased the incidence of white rot up to 50% with efficiency similar to tebuconazole (Jaime et al., 2008). In Mexico, the application of a commercial formulation of *C. minitans* at a dose of 10 kg ha<sup>-1</sup> in soil naturally infested with *S. cepivora* decreased the viability of sclerotia around 65% (Pérez-Moreno et al., 2004). In Canada, the onion seed treatment with *Bacillus subtilis* reduced the diseased plants above 80% with similar control levels of iprodione and vinclozolin fungicides applied at 30 kg a.i. ha<sup>-1</sup> (Utkhede & Rahe, 1983). Clarkson et al. (2006) also found that the integrated use of *T. viride* seeds treated with tebuconazole and onion residues were effective in the control of white rot. In Brazil, the effect of *Bacillus* spp. and *Trichoderma* spp. isolates on the mycelial growth inhibition of *S. cepivora* was evaluated in *in vitro* experiments. The isolates of *Bacillus* spp. (B03, B570, B571 and B854) and *Trichoderma* spp. (SF45, SF32 and SF311) isolates reduced mycelial growth of *S. cepivora* and are mutually compatible. Therefore, these isolates can be applied together for the white rot management (Fuga et al., 2016).

Despite the biocontrol potential shown by several BCAs in the laboratory, their performance in the field may be variable due to many factors including dosages and soil and environment conditions (Clarkson et al., 2002). For example, the BCAs tested in laboratories are often applied in high amounts which do not represent adequate commercial practice (Metcalf & Wilson, 2001). An additional concern is how different soil and environmental conditions affect BCAs and whether these microorganisms remain reliably antagonistic to *S. cepivora* in the field. Most isolates of *Trichoderma* and *Bacillus* antagonists to *S. cepivora* in Brazil grow at temperatures from 25 to 30 °C (Fuga et al., 2012). However, garlic and onion are cultivated during the cold seasons (autumn-winter) and sclerotia germination of *S. cepivora* occurs in much cooler conditions, between 13 to 18 °C (Coley-Smith et al., 1990), for effective BCAs growth. Bontempo (2016) selected biocontrol agents which grow and inhibit *S. cepivora* at temperatures below 18 °C. The isolates GF420, GF424, and GF426 of *Trichoderma* spp. and GF33, GF63, GF203, GF266, and GF340 of *Bacillus* spp. colonize and reduce the germination of sclerotia of *S. cepivora* at 16 °C. In Brazil, more research is required to evaluate in laboratory studies microorganisms, suited to local conditions, as BCAs for reduction of sclerotia and protection of host roots against infection. The best performers should then be evaluated in greenhouse and field studies to ensure they will be effective under field conditions.

### Host resistance

Host resistance should be an important component of a sustainable white rot management strat-

egy. However, there are no onion or garlic cultivars with resistance or tolerance to the disease. Only partial resistance has been detected in some onion, garlic, and leek genotypes.

Utkhede & Rahe (1978) screened 294 accessions from the USDA world germplasm collection of onion for resistance to *S. cepivora* in a field trial in Burnaby, Canada. Seed were inoculated at planting with sclerotia produced in sand-maize meal medium. Although none of the genotypes was immune to *S. cepivora*, there were differences in susceptibility among the accessions, with 8% to 100% of genotypes producing 12 or more surviving/emerged plants per plot and two plant accessions from Brazil, named 247068 and 256323, had 58 and 63% percent of infection, respectively (Utkhede & Rahe, 1978).

In another field experiment that screened several commercial onion cultivars, the lowest percent infection was recorded in the cultivars Ailsa Craig (22.13%), Dako (23.90%), and Wolska (27.39%) (Utkhede & Rahe, 1980). Based on this result, different seed lots of Ailsa Craig and Wolska were compared for resistance to white rot (Utkhede & Rahe, 1984). The lowest disease incidence was detected in the Ailsa Craig selfed progeny selection from the 1979 seed lot (11.5%) and for the Wolska seed lots, the lowest disease incidence was recorded in the Wolska Lot 79271 (37.7%). The authors concluded that the resistance to white rot was associated with particular seed lots or breeding lines (Utkhede & Rahe, 1984).

Another screening of onion accessions for resistance was carried out in the Netherlands under greenhouse conditions (Vandermeer et al., 1983). Only the cultivars Beth Alpha and Pukekohe Longkeeper showed the highest percent of surviving seedlings. In the evaluation of six leek cultivars, the highest percent of surviving seedlings was measured in Batina (22%), Carentan (18%), and Elephant (24%) (Vandermeer et al., 1983). In this study, the authors did not provide any explanation on the resistance mechanism of the cultivars.

In New Zealand, seedling mortality ranged from 76 to 100% in 61 of 69 *Allium* accessions evaluated for resistance to *S. cepivora* in a greenhouse (Bansal & Broadhurst, 1992). *Allium tel-avivense* showed no evidence of infection to *S. cepivora*. The lowest seedling mortality between 56 and 75% was estimated in the accessions of *A. albidum* (PI 286165), *A. cernuum* (PI 372503), *A. galanthum* (PI 280666), *A. jajlæ* (PI 369180), *A. ledebourianum* (PI 369183), *A. lineare* (PI 369184), and *A. oschaninii* (PI 281722) (Bansal & Broadhurst, 1992). The authors suggested that the lack of infection in *A. tel-avivense* is a result of the inability of the species to stimulates sclerotial germination.

For garlic, mutation breeding has been used to improve resistance to *S. cepivora* (Al-Safadi et al., 2000). In a field experiment in Syria, reduction of infected plants above 50% were found in the cultivars Kisswany and Yabroudly treated with gamma radiation.

However, the weight of the mutated bulbs was lower than the control without gamma radiation (Al-Safadi et al., 2000).

Although potential sources of partial resistance to white rot have been identified in onion and garlic, the inheritance of resistance and putative defense mechanisms were not elucidated. The low stimulatory capacity of root exudates, and the small root mass of some genotypes may be related with plant resistance against white rot (Brix & Zinkernagel, 1992). Currently, we are assessing Brazilian garlic and onion genotypes, looking for resistance to *S. cepivora*.

### Concluding remarks

White rot is difficult to control because *S. cepivora* can survive as dormant sclerotia for many years in soil without a host, and their ability to spread easily by different means and ample distribution in the soil profile. The disease is also difficult to control due to the influence of many biotic and abiotic factors on disease development and the long period onion and garlic crops require protection. The understanding of the biology of *S. cepivora* and disease epidemiology at the local/regional level is therefore essential to develop effective strategies to control this difficult soilborne disease. Since very little is known about white rot in Brazil, an obvious recommendation from this review is to increase expertise in these key areas to support development of local control strategies.

In countries where *S. cepivora* is endemic, the reliability of control has been improved by focusing on development of an integrated management strategy which incorporates measures to prevent the introduction and spread of the pathogen, cost-effective soil treatments that reduce sclerotia densities in soils and properly applied efficacious fungicides and biocontrol agents, with proven activity against *S. cepivora*, to protect plants against infection.

Research has demonstrated that early fungicide application, applied at sowing alone or supplemented with post-planting spraying before sclerotial germination and at later crop stages targeting the stem-base of plants, can provide good control of onion and garlic white rot under low to moderate disease pressure situations. Many efficacious fungicides, including tebuconazole and newer chemistry, and biological control agents (BCAs) have been shown to be effective and are important tools against *S. cepivora* infection.

Management of white rot with crop protection treatments alone is not possible because the long lived sclerotia in soil and the low performance of fungicides and BCAs in soils with high levels of sclerotia. In infested fields with less than 10-20 sclerotia per kg of soil, combining a soil treatment with fungicide and/or biocontrol may be necessary to maintain the disease at manageable levels. The germination stimulant DADS has been shown to be one of the most cost-effective pre-plant soil treatments available to reduce sclerotia

levels in soils over a two years period. In heavily infested fields, however, the use of different soil treatments for periods longer than two years may be necessary to bring the density of sclerotia to levels below which crop protection treatments are more effective at controlling disease. Cultural practices such as crop rotation and biofumigation, and physical methods such as solarization in regions with warm weather, have been shown to be effective in reducing inoculum and disease incidence and therefore must be considered as part of a future integrated strategy to restore heavily infested fields for *Allium* production in Brazil.

Extensive research efforts are required in Brazil to develop and optimize application of plant protection (fungicides) and soil treatments (synthetic DADS), as well as beneficial cultural practices and physical control methods, to improve management of white rot. Research efforts are also required to develop non-chemical control options including biological control, natural biostimulants of sclerotial germination, and anaerobic soil disinfestation, and novel technologies. Resistant cultivars to white rot are currently not available to growers. Advances in biotechnology may contribute to the development of resistant crops, especially by developing molecular markers for identifying resistance in conventional breeding and by developing transgenic crops. Future studies working towards this aim will benefit from the development of pathogen host interactions modeling to predict the performance of new cultivars in soils with different populations of *S. cepivora*. Both approaches, conventional and molecular and their integrated use would be required to develop the tools and strategies needed to sustainably manage white rot in infested fields.

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