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Biological control of Sclerotinia stem rot disease (*Sclerotinia sclerotiorum*) in soybean

Soya fasulyesinde Sclerotinia kök çürüklüğü hastalığının biyolojik kontrolü

Ayşegül ÇOLAK[a*]

https://orcid.org/0000-0003-3712-1132

[a]*Adana Biological Control Research Institute, Kışla Caddesi, 31321 Yüreğir, Adana, Türkiye*

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* Corresponding author: Ayşegül ÇOLAK
aysegulcolak@hotmail.com

ABSTRACT

In soybean (*Glycine max*) cultivation, the disease pathogen causing Sclerotinia stem rot (SR) disease is *Sclerotinia sclerotiorum* (Lib.) de Bary. The *Sclerotinia sclerotiorum* causes significant crop losses due to its seed and soil borne infecton. In this study, the effectiveness of biological control agents such as *Coniothyrium minitans* (CM), *Trichoderma harzianum* (TH), and *Bacillus subtilis* (BS), which can be used as alternatives to chemical control in managing SR, was investigated when applied together and at 10-day intervals. In the study, the lowest infected plant rates in biological control with SR were obtained from biological agent applications 10 days before disease inoculation. In this context, the lowest disease severity % and the highest % effect rate were obtained from the CM+10 days later SR application with 31.68% and 61.94%, followed by BS+10 days later SR and TH+10 days later SR, respectively. All biological agents have been found to significantly reduce disease occurrence compared to control. In the study, the most successful biological agent alternative to chemical control against *Sclerotinia sclerotiorum* was *Coniothyrium minitans*, followed by *Bacillus subtilis* and *Trichoderma harzianum*, respectively. The study results revealed that the applications of biological preparations before, during, or after fungal infection is an important factor in improving control of the disease pathogen.

INTRODUCTION

Soybean (*Glycine max* (L.) Merr.), a species of the legume family, is one of the most important industrial plants. Soybean seeds contain an average of 18-24% oil, 36-40% protein, 26% carbohydrates and 8% minerals (Arıoğlu 2007). Approximately 1/3 of the world's cooking oil needs and 2/3 of the protein source are obtained from soybeans (Golbitz 2004). Soybean provides raw materials for different industrial branches and plays an important role in human and animal nutrition in various ways (Canpolat and Maden 2017, Keleş 2019). There are many diseases and pests that affect plant development in soybean cultivation (Canpolat et al. 2022, Keleş 2023). Fungal diseases occurs in soybean, which is rich in nutritional value, reduce the yield and market value of the product. One of the important fungal disease agents that cause significant problems in soybean cultivation areas is *Sclerotinia sclerotiorum* (SR), which causes Sclerotinia stem rot disease. *Sclerotinia sclerotiorum*, a soil-borne pathogen, has a very wide host range and

causes root, stem and fruit rots in approximately 400 plant species, including tomato, cucumber, lettuce, bean, cabbage, sunflower and soybean (Boland and Hall 1994, Çolak 2019, Elshestawi et al. 2017, Haddad et al. 2017, Nelson et al. 1998, Smolinska and Kowalska 2018). The fungal pathogen overwinters as sclerotia in the soil or in plant debris. The first symptoms observed in the plant are usually wilting and drying of the leaves, followed by plant death. When looking closely under the vegetation, cottony, white mycelial growth is seen on the stems, leaves or pods. Lesions develop over time on the main stem and side branches. Sclerotia formed by *S. sclerotiorum* have been recorded to survive in the soil for more than 10 years (Adams and Ayers 1979, Agrios 1997, Cook et al. 1975, Smolinska and Kowalska 2018). *S. sclerotiorum* pathogen is a problem in many plants species grown in Türkiye (Aksay et al. 1991, Çetinkaya and Yıldız 1988, Çınar and Biçici 1982, Çolak 2019, Onaran and Yanar 2004, Soylu et al. 2005, Soylu et al. 2020, Tozlu and Demirci 2008). It has been reported that crop losses caused by white rot disease caused by *S. sclerotiorum* in soybean production reach up to 60% (Hoffman et al. 1998, Workneh and Yang 2000, Wrather et al. 1997, Yang et al. 1999). It has been reported that the disease causes significant yield loses, as well as decreased seed germination, seed number and weight, and loss of market value due to reduced oil and protein concentrations in heavily infected fields (Danielson et al. 2004, Hoffman et al. 1998, Yang et al. 1999).

The *S. sclerotiorum* is a very difficult fungal pathogen to control due to the resistant structures create in the soil. In the control of the disease; it has been stated by many researchers that there are no resistant soybean varieties. The pathogen has a wide host range, its chemicals control harms environment and the fungicides increases the resistance of the pathogen (Delen and Yıldız 2006, Yıldız and Çoşkun 2017). In the control of Sclerotina rot diseases, biological preparations suppress the pathogens by secreting antibiotics, competing with them for food and space or by having an antagonistic effect on them (Cook and Baker 1983). It has been reported in many studies that biocontrol agents *Coniothyrium minitans*, *Bacillius subtilis* and *Trichoderma* species are effective in controlling *S. sclerotiorum* (Jones et al. 2003, Krutova 1987, Zazzerini et al. 1987). *C. minitans,* a biocontrol agent, shows an ability to control sclerotia with growing mycelium which invades sclerotia intracellularly and intercellularly (Jones 1970). The components of the cell walls of *S. sclerotiorum* are β (1-3) glucan and chitin. Lysis is caused by endo-glucanases and exo-glucanases (chitinases and β (1-3) glucanase) produced by *C. minitans* (Melo et al. 2011). It has been reported that *C. minitans* attacks the sclerotia of *S. sclerotiorum*, forming numerous pycnidia on the surface of the sclerotium and that infected sclerotia soften and deteriorate (Bora and Özaktan 1998). The sclerotic mycoparasite *C. minitans* preparation is a successful biocontrol agent that is not licensed in Türkiye but is widely used in the USA and other G8 countries in the control of the soilborne plant pathogen *S. sclerotiorum*. The *C. minitans* has been reported by many researchers to infect and degrade the *S. sclerotiorum* in both field and greenhouse trials (Budge and Whipps 2001, Çolak 2019, Gerlagh et al. 1999, Jones et al. 2003, McQuilken and Whipps 1995). However, no study has been conducted in Türkiye to evaluate the success criteria of these biological preparations in the control of Sclerotinia stem rot disease caused by *S. sclerotiorum* in soybean fields.

Bacillus spp. is the most numerous rhizobacteria in the soil. *Bacillus subtilis* is antagonistic bacteria; usually act through antibiosis, parasitism and competition for space and nutrients. (Nagórska et al. 2007, Santos et al. 2006). *Bacillus* species are Gram-positive and are important in biological control studies due to their spore-forming abilities, which allow them to be resistant to adverse environmental conditions, and the many secondary metabolites they produce. In addition, it has been reported that these bacteria promote plant growth and increase host resistance by producing hormones such as auxin and cytokinin, and by converting phosphate and iron into forms that plant can absorb (Ahmed et al. 2008, Amer et al. 2000, Kim et al. 2003, Liu et al. 2011).The strong mycoparasitic effect of *Trichoderma* species and the effectiveness of the antibiotics they produce have led researchers to focus on *Trichoderma* (Aydın 2015). When *Trichoderma* species are applied to soil or seeds, they rapidly develop along the root system of the plant (Harman 2000). The antagonistic effect occurs through different mechanisms such as the production of antifungal metabolites by *Trichoderma*, competition for nutrients and space, and mycoparasitism (Kredics et al. 2003). *T. harzianum* first dissolves the cell wall of the pathogen mycelium and performs hyphal penetration. After penetration, it breaks down the susceptible host hyphae using enzymes such as lipase and protease (Elad et al. 1980). The ability to secrete active compounds varies greatly among *Trichoderma* species and isolates. Therefore, the mechanisms used by *Trichoderma* spp. in biological control vary according to species, pathogen and host plant. In case of antifungal activity against *S. sclerotiorum*, the mycoparasitic properties of *Trichoderma* play an important role. It has been reported that *Trichoderma harzianum* and some *Trichoderma* species significantly increase plant germination, root and shoot development, dry weight and leaf area, and also increase resistance to biotic and abiotic stresses (Harman et al. 2004, Windham et al. 1986, Yedidia et al. 2001).

Sclerotinia sclerotiorum is a fungal disease that is difficult to control due to the resistant structures it creates in the soil. In the control of the disease; inadequacy of environmentally friendly fungicides, residue risks, emergence of fungicide resistance have brought the research of biological control possibilities to the agenda in recent years. In this study; we aimed to demonstrate the effectiveness of *Coniothyrium minitans*, *Bacillus subtilis* and Trichoderma harzianum, which are alternative biological agents to chemical control, in the control of Sclerotinia stem rot disease, which causes economically significant product losses in soybean cultivation, when applied together and 10 days intervals.

MATERIALS AND METHODS

Fungal disease inoculum and preparation of biological preparations

The inoculum suspension of the fungal disease pathogen *Sclerotinia sclerotiorum* (SR) obtained from soybean plants was incubated in PDA medium for 10 days at 25 oC (Figure 1). The disease inoculum covering Petri dish surface and forming sclerotia was prepared by mixing it in 500 ml of sterile water. In the experiment, 20 ml of fungal inoculum suspension was applied to the pots where SR would be applied. Sterile water was applied to the pots as a negative control (Soylu 2011).



Figure 1. *Sclerotinia sclerotiorum* isolate and image from the experimental area

Coniothyrium minitans (CM), *Trichoderma harzianum* (TH) and *Bacillus subtilis* (BS) which are alternative biological agents to chemical control against Sclerotinia stem rot disease were applied to the pot containing soil with 10 days intervals according to manufacturer label information. For this purpose, *C. minitans* (CM, strain CON/M/91-08, $1x10^{12}$ spores/kg, Bayer) biological preparation was calculated as 4 kg/ha, *B. subtilis* (BS, *Bacillus subtilis* QST-713, $1x10^{9}$ cfu/ml, Serenade) biological preparation was calculated as 1000 ml/da, *T. harzianum* (TH, *Trichoderma harzianum* Rifai race KRL-AG2, T-22 Planter Box, $1x10^{7}$ cfu/g, Bioglobal) biological preparation was calculated as 50 g/100 l. The biological preparations used in the experiment were calculated based on licensed doses and applied as 20 ml to each pot.

Experimental design

The experiment was set up according to randomized plot design with 5 replicates for each application and 12 soybean seeds for each replicate (Karman 1971). Nazlıcan soybean variety was used in the trials. In the experiment, applications were made to the pots together (SR+CM, SR+BS, SR+TH), SR and biological preparations were applied at 10-day intervals (CM+10 days later SR, TH+10 days later SR, BS+10 days later BS and SR+10 days later CM, SR+10 days later TH, SR+10 days later BS). SC application in control pots was considered as positive control, and only water application was considered as negative control. The experiment was set up in 30x30 cm pots containing sterile peat:perlite (1:1) environment and at 25 ± 2 °C controlled greenhouse conditions in Adana Biological Control Research Institute (Figure1).

Statistical analysis

The pots with applied applications were kept in a controlled greenhouse for 45 days, and lesions on plant roots, wilting on leaves, and growth retardation were observed for evaluation disease severity (%) and disease index value according to a 0-3 scale where 0: no symptom, 1: symptom on lateral branches only, 2: symptom on the main stem but little or no damage on pods, 3: symptom on main stem resulting in plant death and poor or no pod fill (Kim et al. 2000). Disease severity (DS) was calculated according to a formula DS: (Σ (disease scale rating of each plant) / 3x number of plants reacted) x100. Variance analysis (ANOVA) and comparison of mean values were performed with the trial results and LSD test (P: 0.05) was determined in Jump 13.0 statistical package program (Townsend and Heuberger 1943).

RESULTS

Collected data on the studies reveals effectiveness of *C. minitans, T. harzianum* and *B. subtilis*, which are alternative biological agents to chemical control, in both applications with 10 days intervals are given in Table 1. When Table 1 is examined; the differences in disease severity rates between Sclerotinia stem rot disease and biological control applications were found to be statistically significant ($P<0.05$). In the control with SR, the lowest infected plant rates in the applications of disease and biological agents together and at an interval of 10 days were obtained from the applications of biological agent 10 days before disease inoculation; this was followed by the application of SR+biological agent together. In this context, the lowest disease severity % and the highest % effectiveness rate in the applications were obtained from CM+10 days later SR application with 31.68% (61.94%), this was followed by BS+10 days later SR with 42.78% (48.62%) and TH+10 days later SR with 45.00% (45.98%), respectively (Table 1).

Table 1. Effect of biological control agents applications on soybean Sclerotinia stem rot disease

Applications	Disease severity %	% Effect	Disease scale
SR+CM	55.54 b	33.25	1.67
SR+TH	64.46 bc	22.65	1.93
SR+BS	61.12 bc	26.65	1.83
SR+10 days later CM	68.34 cd	18.01	2.05
SR+10 days later TH	72.76 ef	12.60	2.18
SR+10 days later BS	71.66 de	13.93	2.15
CM+10 days later SR	31.68 a	61.94	0.95
TH+10 days later SR	45.00 b	45.98	1.33
BS+10 days later SR	42.78 ab	48.62	1.28
SR (positive control)	83.34 f	-	2.50
NC (negative control)	0.00	-	0.00
Lsd 0.05	11.123		

Among all applications in the experiments, the highest infectious disease severity and the lowest % effect were obtained from the application of biological agents to the soil 10 days after the disease inoculation of *S. sclerotiorum*. The highest disease severity in the study was obtained from SR+10 days later TH (12.6%) with 72.76% followed by SR+10 days later BS (13.93) with 71.66% and SR+10 days later CM (18.01%) with 68.34%, respectively. In the study the most successful biological agent used as an alternative to chemical control in the control of fungal disease pathogen *S. sclerotiorum* in soybean plants was *C. minitans* followed by *B. subtilis* and *T. harzianum* (Table 1).

DISCUSSION

Sclerotinia sclerotiorum is a soil-borne pathogen; it is known that fungicides with different active ingredients are successful in its chemical control. *S. sclerotiorum* always has the risk of developing resistance to fungicides and the fungicides used have a very fast disintegration time in the soil, so they are not very sufficient in eliminating sclerotia. Due to the spore structure of the disease, there is no high level of resistance to the pathogen in soybean host plants damaged by *S. sclerotiorum* (Bardin and Huang 2001, Demir and Delen 1991, Huang et al. 1993). Due to negative effects of fungicides applied to the soil on beneficial organisms in the soil, previous studies have intensified on biological agents that suppress soil-borne pathogens such as *Sclerotinia* and prevent germination by parasitizing sclerotia. In this context, our study results demonstrated; successful controlling against Sclerotinia stem rot disease, which causes significant yield losses in heavily infested areas and destroys market value of soybean plants; it is thought that the application of the biological agent *C. minitans* to the soil can be an effective alternative to chemicals. The success of biological preparations may vary depending on the soil type, type of biological agent, pathogen concentration, plant interactions, and disease pathogen (Smolinska and Kowalska 2018). In the study, the

most successful biological agent used as an alternative to chemical control in the control of fungal disease pathogen *S. sclerotiorum* in soybean plants was *C. minitans*. In a study conducted in Canada, it was reported that application of *Coniothyrium minitans* to soil in spring reduced apothecium production from sclerotium of *S. sclerotiorum* in the soil and increased parasitism in sclerotium produced on diseased beans (McLaren et al. 1996). In the study conducted on the control of *S. sclerotiorum*, which is a problem in soybeans and sunflowers grown in greenhouse and field conditions; it has been reported that *C. minitans* is effective when applied to soil (Sesan and Csep 1993). It was reported that β-1, 3-glucanase secreted by *C. minitans* disrupts and breaks down sclerotial tissues and is effective by breaking them down. The *C. minitans* is commercially applied and dried spores of the antagonist are sprayed on crop residues infested with pathogens at the end of the season or on the soil surface before planting and are quite economical in disease control. Commercial preparation containing *C. minitans* has become the preferred biopreparation against *S. sclerotiorum* in many different products (Budge and Whipps 1991, Chitrampalam et al. 2011, Melo et al. 2011). Bora and Özaktan (1998) reported that *C. minitans* attacks the sclerotia of *S. sclerotiorum* and forms numerous pycnidia on the surface of the sclerotia, and the infected sclerotia soften and deteriorate. Mcquilken et al. (1997) conducted pot experiments to determine the effectiveness of *C. minitans* on the sclerotia of *S. sclerotiorum* and found that *C. minitans* completely suppressed apothecia production and no new sclerotia formation was observed. According to the results of our study; the application of the biological agent to the soil before the fungal pathogen enters the host plant increasing effectiveness of the fight against the fungal pathogen, *S. sclerotiorum*. In the study, it was observed that the disease rates in all biological agent applications significantly reduced the disease outbreak compared to the control. Zeng et al (2012) in their study on soybean, investigated the effect of biological control agents on reducing the number of sclerotia in the control of Sclerotinia stem rot disease. In the study, biological agents *Coniothyrium minitans, Streptomyces lydicus* WYEC 108, *Trichoderma harzianum* and *Bacillus subtilis* QST713 were used. In the study, biological agents were applied to the top soil before planting to the soil contaminated with *S. sclerotiorum* sclerotia in two field regions. In the study, it was reported that applying *C. minitas* to the top soil before soybean planting reduced *S. sclerotiorum* disease severity index by 68% and the number of sclerotia in the soil by 95.3%. It was determined that *S. lydicus* and *T. harzianum* reduced the disease severity by 43.1% and 38.5%, and the number of sclerotia in the soil by 90.6% and 70.8%, respectively and the lowest effect was obtained from *Bacillius subtilis* applications. These results are parallel to our studies.

The application of *B. subtilis* before disease infection, which is an alternative biological agent to chemical control in controlling the fungal disease agent *S. sclerotiorum* in soybean plants, provided effective success. Rhizobacteria, which are capable of colonizing roots, are present in the plant rhizosphere region. Rhizobacteria form a first line of defense for roots against pathogen attack. Pathogens are confronted with the antagonism of these rhizosphere microorganisms before infection, during infection or during secondary spread in roots. As a result of this interaction, disease development is prevented or slowed down (Weller 1988). *Bacillus* species have been shown in many studies to be highly effective against soil-borne pathogens (Kumar et al. 2012, Rigobelo et al. 2024, Weller 1988, Zhang and Xue 2010). *Bacillus* spp. produces endospores that are resistant to adverse environmental conditions, making it easier for them to survive for a long time. These bacteria produce different types of antibiotics and growth-promoting compounds that can play a major role in their disease-suppressing and growth-enhancing effects on plants treated with them (Hallman et al. 1997). Zazzerini et al. (1987) recorded that *B. subtilis* and *B. cereus* bacterial isolates naturally surrounded infected sunflower soil, causing the contaminated sclerotia to form fewer mycelia and die. In the study, it was also determined that the germination amount of *S. sclerotiorum* sclerotia was reduced or suppressed by *B. subtilis*. Dutta et al. (2008) investigated the effects of various antagonists against *S. sclerotiorum*, which causes rot in beans, using dual culture methods. As a result of these studies, it was determined that among all the antagonists, *Pseudomonas fluorescens* provided a 64.93% growth reduction, followed by *Bacillus subtilis* with 62.86% and *Trichoderma harzianum* with 59.08%. Canik (2020) reported that the commercial *Bacillus subtilis* QST 713 preparation was effective against disease pathogens at all application times before inoculation and had a disease infection reducing effect at all application times. Soylu et al. (2020) reported in their study that some *Bacillus* spp. isolates caused significant morphological changes on the hyphae of the fungal disease agent *S. sclerotiorum*. In the study, *in vitro* antagonism activity of antagonist bacteria increased depending on the pre-incubation period before fungal inoculation. They reported that *Bacillus* spp. isolates can be used as effective biocontrol agents alternative to chemicals in the control of soilborne diseases to promote organic and sustainable agriculture due to their high antagonistic properties. In many studies of the *Bacillius* genus; it has been reported that it promotes plant growth by producing hormones such as auxin and cytokinin and converting phosphate and iron into useful forms,

thus enabling the plant to develop healthy structure and resistance to diseases, as well as increasing yields (Etesami et al. 2023, Glick 1995, Orozco-Mosqueda et al. 2013, Rigobelo et al. 2024, Smolinska and Kowalska 2018).

Among all treatments in the experiment, the highest infectious disease severity and the lowest % effect were obtained from the application of biological agent *T. harzianum* to the soil 10 days after *S. sclerotiorum* disease inoculation. The differences in the success of biological preparations may vary depending on the soil type, application environment and time, type of biological agent, plant interactions, and disease pathogen (Smolinska and Kowalska 2018). *Trichoderma* species are among the most isolated fungi from plant root zones and are known as biological agents that either parasitize or antagonis fungal pathogens that cause significant yield losses in many plants. They are also widely used against many plant pathogens (Harman et al. 2004, Kumar et al. 2021). *Trichoderma* species can produce antibiotics and are very strong in terms of hyperparasitism and nutrient competition, and can be used as effective antagonists that can prevent soil pathogens (Bora and Özaktan 1998). *Trichoderma harzianum*, which is known to be effective against soil-borne microorganisms, is quite difficult to biologically control aggressive pathogens with high epidemics, but *Trichoderma* species are quite successful against pathogenic fungi with various cell wall-degrading enzymes and secondary metabolites (Agrios 2001). Menendez and Godeas (1998) reported that in greenhouse and field trials where they applied *Trichoderma harzianum* to the biological control of *S. sclerotiorum* in soybean, the plants survived 40% more compared to other applications. In the study, a significant decrease of 62.5% in the number of germinated sclerotia was found in *T. harzianum* applied plots. Kumar et al. (2017) evaluated them against some soilborne plant pathogens using potential biological control agents. The maximum reduction in sclerotia production of *Sclerotinia sclerotiorum* was obtained from *Trichoderma harzianum* with 88.20%. Visser (2007) reported in his study that *T. harzianum* could reduce the effects of Sclerotinia on soybean plants. In soybean; 120 *Trichoderma* spp. tested *in vitro* to control *S. sclerotiorum*; it was observed that 22 species inhibited sclerotia germination by 100% and two species increased seed germination up to 67% in the greenhouse. The species with the highest performance were determined to be *T. koningiopsis, T. asperelloides, T. atroviride* and *T. virens* (Haddad et al. 2017). In the study conducted by Jones et al. (2014) in the control of *Sclerotinia sclerotiorum* pathogen in cabbage, *C. minitans, T. hamatum, T. rossicum, T. virens, T. crassum* and *Clonostachys rosea* biological agents were evaluated for their ability to control the disease by applying them to field soil. In the study, it was reported that the bioagent fungi, especially *Coniothyrium minitans* and *Trichoderma hamatum*, have the potential to control the disease by reducing *Sclerotinia sclerotiorum* infection by 46–52% and 31–57%, respectively. Krutova (1987) determined in field and laboratory studies that *C. minitans, T. hamatum, T. harzianum* and *G. catenulatum* found on the sclerotia with a hyperparasitic effect on the pathogen and could destroy the sclerotia in the soil, and stated that *Coniothyrium minitans* was the most active of these hyperparasites. Huang et al. (2000) tested five fungal species for biological control of white rot in bean. One of them was the antagonist *Epicoccum purpurascens*, and four of them were mycoparasites *C. minitans, T. virens, T. flavus* and *Trichothecium roseum*; they found that these biocontrol agents significantly reduced the disease and *C. minitans* and *E. purpurascens* were the most effective biocontrol agents.

Biological control methods may be partially sufficient to completely reduce the populations of plant disease pathogens. It is very difficult to control diseases caused by *S. sclerotiorum* with a single control method. Due to factors such as excessive yield and quality loss caused by the fungus, its ability to infect many plants and its difficulty in control, it is important to research integrated control methods together to provide long-term and effective biological control.

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Author's Contributions

Authors declare that each author's contribution is equal.

Statement of Conflict of Interest

The authors have declared no conflict of interest.

ÖZET

Soya (*Glycine max*) yetiştiriciliğinde *Sclerotinia* kök çürüklüğü (SR) hastalığına neden olan hastalık etmeni *Sclerotinia sclerotiorum* (Lib.) de Bary'dir. Hastalık etmeni tohum ve toprak kökenli olması nedeniyle önemli ürün kayıplarına neden olmaktadır. Bu çalışmada SR'nin kontrolünde kimyasal mücadeleye alternatif olabilecek biyolojik kontrol ajanlarından *Coniothyrium minitans* (CM), *Trichoderma harzianum* (TH) ve *Bacillus subtilis* (BS)'nin birlikte ve 10 gün arayla yapılan uygulamalarındaki etkinliği araştırılmıştır. Çalışmada fungal hastalık ve biyolojik ajanların birlikte ve 10 gün arayla uygulamalarında en düşük enfekteli bitki oranları hastalık inokulasyonundan 10 gün önce biyolojik ajan uygulamalarından elde edilmiştir. Bu kapsamda uygulamalarda en düşük hastalık oranı ve en

yüksek % etki oranı, %31.68 ve %61.94 ile CM+10 gün sonra SR uygulamasından elde edilmiştir; bunu sırasıyla BS+10 gün sonra SR ve TH+10 gün sonra SR izlemiştir. Çalışmada tüm biyolojik ajanların uygulamalarındaki hastalık oranları kontrole göre hastalık çıkışını önemli düzeyde azalttığı tespit edilmiştir. Çalışmada soya bitkisinde fungal hastalık etmeni *Sclerotinia sclerotiorum* ile mücadelede kimyasal mücadeleye alternatif biyolojik ajanlardan en başarılısı *Coniothyrium minitans* olmuş, bunu sırasıyla *Bacillus subtilis* ve *Trichoderma harzianum* izlemiştir. Çalışma sonuçları biyolojik preparatların uygulanmasında; fungal hastalık enfeksiyondan önce, enfeksiyon sırasında veya sonrasındaki uygulamaların patojen ile mücadelede başarıyı artırmada önemli bir etken olduğunu ortaya koymuştur.

Anahtar kelimeler: soya, *Sclerotinia sclerotiorum, Coniothyrium minitans, Trichoderma harzianum, Bacillus subtilis*

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