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MANAGEMENT OF SUNFLOWER SCLEROTINIA ROT WITH CHEMICALS[1]

WEN-SHI WU

Department of Plant Pathology and Entomology
National Taiwan University, Taipei,
Taiwan 107, Republic of China

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Abstract

Sunflower was grown in a field heavily infested by *Sclerotinia sclerotiorum* on October, 1979. The amount of inoculum was approximately 22-40 apothecia/ M^2 during the growing stage of sunflower. Each apothecium was able to produce 1.3×10^6 ascospores totally. The apothecia discharged ascospores continuously for 11-12 days under laboratory condition.

DCNA, mancozeb, PCNB, and vinclozolin at the concentration of 1,000 ppm of active ingredient were sprayed once every 10 days after 14 days of planting. Ten sprayings were carried out totally in the field. Percent germination, height, disease index, survivability and yield of sunflower were recorded and analysed statistically. Vinclozolin seemed to be the best chemical among these fungicides to control the disease. Besides, DCNA was also effective in preventing sunflower from infection. PCNB and mancozeb were unable to reduce the harmful effect caused by *Sclerotinia sclerotiorum*.

Ascospores germinated readily on the surface of both leaves and stems of sunflower which were treated with sterilized distilled water. Ascospores were unable to germinate or gave a limited growth on sunflowers whenever the plants were pre-sprayed with either vinclozolin or DCNA.

Introduction

Sclerotinia sclerotiorum has a very wide range of hosts. Besides, sclerotia of this pathogen can survive long time in the soil. Hence it is not suitable and uneasy to apply crop rotation to reduce the threatening of sclerotinia disease in Taiwan where farmers grow economic crops intensively in their land. Although *Coniothyrium minitans* (Huang, 1977; Huang and Hoes, 1976), *Sporidesmium sclerotivorum* (Ayers and Adams, 1979a, b) have been proven to be able to infect the sclerotia of *S. sclerotiorum*, it may not succeed to control sclerotinia disease in the field without understanding the ecology of these antagonists. Planting resistant varieties of crops is a very effective method

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to control plant diseases. However, there are no profitable vegetables and oil crops resistant to *S. sclerotiorum* in Taiwan. Disease management with chemicals still provides direct, prompt and effective results.

It is important to investigate the source and dispersal of inoculum of a disease which are generally the two critical factors associated with the outbreak of disease. It is necessary to understand the rhythm of discharging infective unit of pathogens, then the critical spraying time can be setted.

This paper studies on the effectiveness of chemicals to control sclerotinia disease, the effect of chemicals upon the germination and penetration of *S. sclerotiorum* on the surface of sunflower, and the spore discharging rhythm of this pathogen.

Materials and Methods

DCNA (Dicloran, 2, 6-dichloro-4-nitroaniline; Boots), mancozeb (Dithane M-45, a coordination product of zinc ion and manganese ethylene bisdithiocarbamate; Rohm and Haas), PCNB (Terraclor 75%, pentachloronitrobenzene; Olin International) and vinclozolin (Ronilan 50%, 3-(3, 5-dichlorophenyl) 5-ethyl-5-methyl-2, 4 oxazolidinedione; BASF) were selected to spray onto sunflower in the field. This field was infested heavily by *S. sclerotiorum* naturally since it was used to grow cruciferous plants and sunflower continuously for many years. Sunflower (*Helianthus annus* L.) was planted in a 0.05 ha field. There were four plots. Each plot consisted of nine treatments which were control and plants treated with either of the four fungicides. Each chemical consisted of two treatments. One was placing 50 g of chemical (a. i.) along the seeding sites plus spraying with the same chemical 14 days after planting. The other was spraying only 14 days after planting. The concentration of each spraying chemical was 1,000 ppm of active ingredient. Spraying was once every 10 days. There were 10 sprayings totally, and it was harvested after 120 days. Randomized completely block design was used in this study. There was one row of oats growing beside every treatment in order to prevent the drift of chemical to the sunflower which should not be treated with it. Each treatment consisted of 40 plants which were planted in two adjacent rows. Each row was 25 cm apart from the next one, and it was 6 M long. Disease ratings were recorded separately for stem and flower parts. Disease ratings of stem were 0, healthy; 1, leaves being infected; 2, stem being infected; 3, both leaves and stem being infected but without wilting; 4, wilting. Disease ratings of flower were 0, healthy; 1, 1/4 of flower part being infected; ...; 4, 4/4 of flower part being infected.

1,000 ppm of both DCNA and vinclozolin were selected to spray onto sunflower at 5, 2 and 0 days before and after inoculation with the ascospore of

S. sclerotiorum. After inoculation of two days, the inoculated leaves and stems were detached from sunflower. These tissues were fixed with 2.5% glutaraldehyde and 1% osmic acid, then dehydrated with a series of alcohol. After rinsing with isoamyl acetate for 10 minutes, the specimens were dryed by critical point drying method, coated with gold. The specimens were observed with scanning electron microscope (Hitachi S-550).

A 24-hour spore trap (Kramer and Pady, 1966) has been used to trap the released ascospores every hour from the apothecium. The apothecium was collected either from the field or the culture. Spore trap was started from the very beginning of initiated apothecium to the time of decaying. The collected apothecium with sclerotium was placed in wetted cotton pad in a petri plate which was covered with a glass funnel which was connected with the spore trap by rubber tube. One apothecium was tested each time and repeated five times. The number of ascospore releasd every hour from every apothecium was counted.

Results

Applying PCNB in the soil increased the germination significantly ($P=0.05$) than control after 7 days of planting (Table 1). Plants grew from the soil treated with PCNB and sprayed with the same fungicide afterward grew significantly ($P=0.05$) higher than control after 55 days of planting. At the time of harvesting, vinclozolin and DCNA provided significanly ($P=0.05$) better disease control than the other treatments to prevent the stems and leaves from the infection by *S. sclerotiorum* in the field. In spite of rotted flower head, vinclozolin and DCNA kept the significantly ($P=0.05$) more green and standing sunflower than the other treatments in the field. However, only plants treated with vinclozolin had significantly ($P=0.05$) more yield than the other treatments.

Ascospores germinated readily on the untreated leaves and stems after two days of inoculation (Fig. 1). However, they seemed unable to germinate on vinclozolin-treated tissues, even the ascospore located beside the stoma. DCNA allowed the ascospore to germinate, but the germination was limited.

After 20 days of planting, apothecia were appeared in the field. There were about 22–40 apothecia persisting on each M^2 of experimental field during the growing period. Since the initiation of apothecia from the sclerotia, the apothecia can last for 16 to 22 days without decaying. Each apothecium can produce ascospores continuously for 11 to 12 days (Fig. 2). Most of these ascospores released from the fifth to the ninth days ever since the apothecium started to release ascospores. The maximum daily production of ascospores was 3×10^5, and 1.3×10^6 ascospores totally during the period of

Table 1. *The effectiveness of different chemicals to control sclerotinia disease of sunflower*

Characteristics of Sunflower	Treatments								
	Control	DCNA		Mancozeb		PCNB		Vinclozolin	
		Soil incorporation + Spray	Spray	Soil incorporation + Spray	Spray	Soil incorporation + Spray	Spray	Soil incorporation + Spray	Spray
Percent germination after 7 days of planting	54.50^{bc1}	52.25^{bc}	52.25^{bc}	47.50^{bc}	53.25^{bc}	64.50^{a}	47.00^{c}	55.25^{bc}	57.00^{ab}
Height (cm) after 55 days of planting	35.72^{cde}	40.53^{bcde}	40.09^{bcd}	31.63^{e}	44.31^{bc}	54.13^{a}	34.69^{de}	40.09^{bcd}	46.06^{ab}
Disease rating[2] (Leaves & Stems) before harvesting time	3.66^{a}	2.16^{b}	2.50^{b}	3.50^{a}	3.75^{a}	3.50^{a}	3.28^{a}	0.75^{c}	1.19^{c}
Disease rating[3] (floral parts) before harvesting time	3.94^{a}	3.25^{c}	3.31^{bc}	3.84^{ab}	4.00^{a}	3.63^{abc}	3.66^{abc}	3.50^{abc}	3.66^{abc}
Percent survival before harvesting time	7.50^{c}	21.00^{b}	19.25^{b}	4.75^{c}	3.00^{c}	4.75^{c}	7.25^{c}	32.50^{a}	32.75^{a}
Average yield (kg) of 4 replicates	163.75^{b}	220.13^{b}	176.94^{b}	91.25^{b}	110.63^{b}	130.00^{b}	163.13^{b}	546.25^{a}	576.25^{a}

1 Values followed by common letter in the same line do not differ significantly at the 5% level by the Duncan's new multiple-range test. Data were the average of 160 plants.

2 0: healthy, 1: leaves infected, 2: stem being infected, 3: both leaves and stem being infected but without wilting, 4: wilting.

3 0: healthy, 1: 1/4 of flower part being infected,..., 4: 4/4 of flower part being infected.

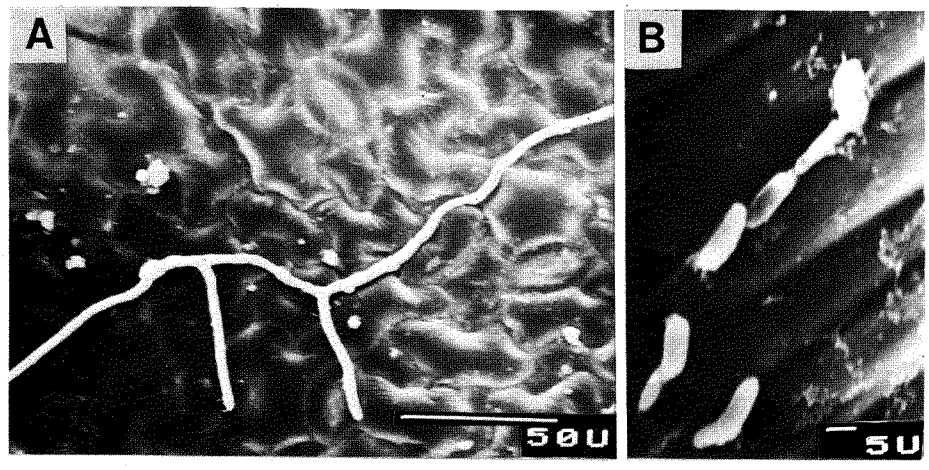

Fig. 1. The behavior of ascospores of *Sclerotinia sclerotiorum* on (A) Chemical-untreated leaf and (B) Vinclozolin-treated stem of sunflower.

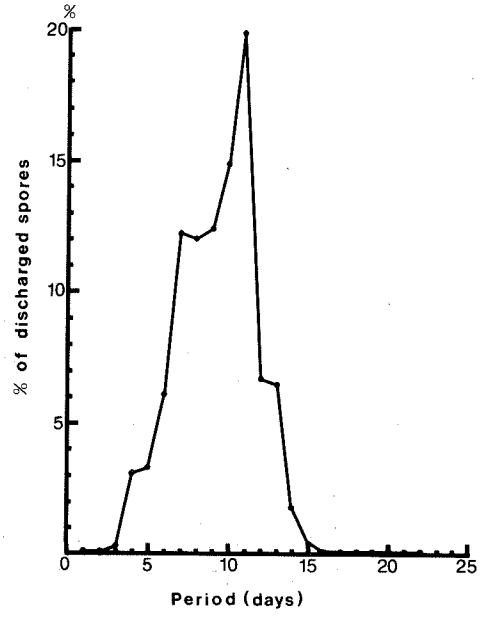

Fig. 2. Percent discharging ascospores of *Sclerotinia sclerotiorum* during its spore-releasing period.

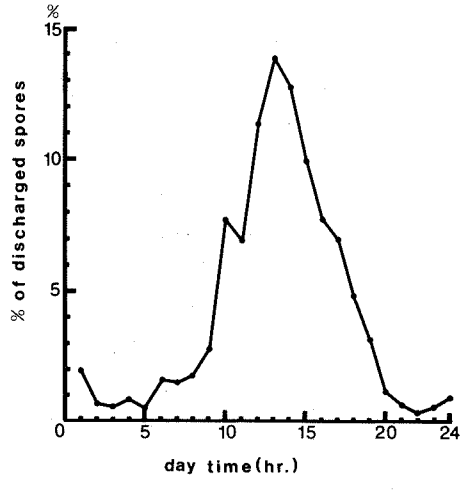

Fig. 3. Percent discharging ascospores of *Sclerotinia sclerotiorum* during 24 hours of spore-releasing days.

releasing. Apothecium discharged ascospores every hour, but the maximum ascospores releasing was during the period between 12 AM and 2 PM (Fig. 3). Within this period of time, one apothecium can release about 2×10^5 ascospores.

Discussion

There were almost no significant difference among these different treatments from control in germinability. Sunflower planted in the soil treated with PCNB germinated more significantly than control. This was probably due to PCNB prevented these seedlings from infection by *Rhizoctonia solani*, since seedlings with girdling necrotic lesion and ungerminated seeds yielded a high percentage of *R. solani* on water agar. This beneficial effectiveness extended to promote the early development of plants, hence sunflowers grown from the soil treated with PCNB showed significantly higher than control. Plants sprayed with vinclozolin also grew significantly higher than control. Although PCNB has been demonstrated to control sclerotinia disease (Beute *et al.*, 1975; Kruger, 1973), PCNB was failed to protect sunflower continuously from the infection by *S. sclerotiorum* which infested with the field intensively (22–40 apothecia/M^2 of field) in this study. DCNA and vinclozolin showed the ability to control sclerotinia disease on sunflower. Soil incorporation with DCNA or sprayed of DCNA can control lettuce drop caused by *S. sclerotiorum* and the effectiveness of DCNA was better than benomyl and PCNB (Marcum *et al.*, 1977). The same was true in this study. DCNA provided significanly more survival plants than plants treated with PCNB or mancozeb. Mancozeb showed a promising effective to inhibit the growth of *S. sclerotiorum* (Charifi-Tahrani, 1974). However, mancozeb was proven not suitable to control sclerotinia disease of sunflower in this investigation. Vinclozolin was a relative new product of BASF and has shown its promising effect to control sclerotinia disease as DCNA did. Vinclozolin could almost inhibit the germination of ascospores completely on the leaves and stems of sunflower. DCNA was also able to delay or stop the germination of ascospores on leaves and stems.

The first application of fungicides was 14 days after planting this time, whereas the first application of chemical was 30 days after planting in 1979 (Lee and Wu, 1979). Since the delay spraying in 1979, there were no significant difference among those chemicals to control sclerotinia disease due to early infection. Early spraying of chemicals protected seedlings from infection and provided normal growth. Consequently, there were some difference between these chemical treatments and control.

Kramer-Collins 24-hour spore sampler can collect air-borne spores every hour (Kramer and Pady, 1966) and was used in this study. Each apothecium

released 1.3×10^6 ascospores in average in laboratory condition by direct counting from these sampling slides, whereas 2.32×10^6 ascospores were estimated by Schwartz and Steadman (1978) by counting from a Spencer hemacytometer. Maximum discharge of ascospores was between 12 AM and 2 PM, and between the fifth and ninth day since the apothecium started to release ascospores. Hence protective chemicals had to be sprayed before the maximum spore releasing time and dates for the purpose of keeping the crops in healthy condition.

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Literature Cited

Ayers, W. A. and P. B. Adams. 1979a. Mycoparasitism of sclerotia of *Sclerotinia* and *Sclerotium* species by *Sporidesmium sclerotivorum*. Can. J. Microbiol. **25**: 17-23.

Ayers, W. A. and P. B. Adams. 1979b. Factors affecting germination, mycoparasitism, and survival of *Sporidesmium sclerotivorum*. Can. J. Microbiol. **25**: 1021-1026.

Beute, M. K., D. M. Porter and B. A. Hadley. 1975. Sclerotinia blight of peanut in North Carolina and Virginia and its chemical control. Plant Dis. Reptr. **59**: 697-701.

Charifi-Tehrani, A. 1974. Effect of several fungicides on mycelial growth and pathogenicity of *Sclerotinia sclerotiorum* (Lib.) de By. agent of sclerotinia disease of sunflower. Phytratrie-Phytopharmacie **23**: 189-192. (*IN* RPP 54: 548).

Huang, H. C. 1977. Importance of *Coniothyrium minitans* in survival of sclerotia of *Sclerotinia sclerotiorum* in wilted sunflower. Can. J. Botany **55**: 289-295.

Huang, H. C. and J. A. Hoes. 1976. Penetration and infection of *Sclerotinia sclerotiorum* by *Coniothyrium minitans*. Can. J. Botany **54**: 406-410.

Kramer, C. L. and S. M. Pady. 1966. A new 24-hour spore sampler. Phytopathology **56**: 517-520.

Kruger, W. 1973. Measures for controlling *Sclerotinia sclerotiorum* (Lib.) de Bary on rape. Phytopathologische Z. **77**: 125-137.

Lee, Y. A. and W. S. Wu. 1979. Management of the sclerotinia disease with biological and chemical methods. Memoirs of the College of Agric. (NTU) **19**: 96-108.

Marcum, D. B., R. G. Grogan, and A. S. Greathead. 1977. Fungicide control of lettuce drop caused by *Sclerotinia sclerotiorum* 'minor'. Plant Dis. Reptr. **61**: 555-559.

Schwartz, H. F. and J. R. Steadman. 1978. Factors affecting sclerotium populations of and apothecium production by *Sclerotinia sclerotiorum*. Phytopathology **68**: 383-388.

向日葵菌核病之化學防治

吳 文 希

國立臺灣大學植物病蟲害學系病理組

栽種向日葵的田中，每平方公尺的地面上散佈著22～40個子囊盤，每一子囊盤平均總共可產生 1.3×10^6 個子囊胞子，而且每一子囊盤可以連續 11～12 天放出子囊胞子。

向日葵栽種兩週後，以 1,000 ppm 有效濃度的 DCNA, mancozeb, PCNB 及 vinclozolin 每隔十天噴灑植物一次，至收穫前共噴灑藥劑十次。DCNA 及 vinclozolin 能够有效地防治菌核病，而 PCNB 及 mancozeb 却不能達到防治的目的.

菌核病菌的子囊胞子在未經藥劑處理的向日葵上，可以很容易地發芽，但却不能在噴灑過 DCNA 及 vinclozolin 的植物體表自由地發芽、生長。