Bot. Bull. Acad. Sin. (2002) **43:** 219-225

Integrated control of black rot disease of wasabi, caused by *Phoma wasabiae*

Chaur-Tsuen Lo[1,*], Kuei-Mai Wang[1], Ming-Fu Hu[2], and Chong-Ho Wang[3]

[1]*Department of Plant Pathology, Taiwan Agricultural Research Institute, Wu-Feng, Taichung 413, Taiwan, ROC*

[2]*Department of Agronomy, Taiwan Agricultural Research Institute, Wu-Feng, Taichung 413, Taiwan, ROC*

[3]*Department of Agricultural Chemistry, Taiwan Agricultural Research Institute, Wu-Feng, Taichung 413, Taiwan, ROC*

(Received November 28, 2001; Accepted January 15, 2002)

Abstract. Soaking with difenoconazole, thiabendazole, benomyl, and polyoxin eliminated the pathogen as an initial inoculum source in rhizome-tillers and root-plantlets of wasabi. The effective period was 2, 4, 8, and 10 h, respectively, for difenoconazole, thiabendazole, benomyl, and polyoxin. Because thiabendazole and difenoconazole caused injury to wasabi tillers, only benomyl was used to control disease in tillers in the field. When plots were covered with PE sheets, leaf spot disease of wasabi was significantly reduced. The average disease incidence of black rot was reduced from 90% to 5.9%. The pathogen was effectively prevented by benomyl from spreading from the parent rhizomes to tillers in the field. Treating wasabi tillers only with benomyl, however, did not significantly reduce the disease incidence on wasabi leaves or petioles. In contrast, the tillers from rhizomes treated with benomyl and covered with a polyethylene sheet on the cultivated plot significantly reduced the disease severity on wasabi rhizomes, petioles and leaves. The treatments reduced disease severity from 75% to 12.5% in rhizomes, from 73.5% to 37.5% on petioles, and from 72.6% to 33.5% on leaves of wasabi. The results of this study suggest that using pathogen-free plantlets and preventing secondary infection with polyethylene cover could be effective in controlling black rot disease of wasabi in the field.

Keywords: Black rot; Fungicide; Integrated control; *Phoma wasabiae*; Rhizome.

Introduction

Wasabi (*Wasabia japonica* [Miquel] Matsu.), is a perennial crucifer introduced into Taiwan from Japan in 1915 and has become an economically important export commodity (Cheng and Tung, 1986; Lo et al., 1990; Hu et al., 1991; Lo et al., 1996). Unfortunately, black rot disease, caused by *Phoma wasabiae* Yokogi has caused considerable losses in yield and market value of the crop since first report of the disease in Taiwan in 1986 by Cheng and Tung (Hu et al., 1986; Lo and Wang 2000a). The disease results in leaf spot, petiole blight, root rot, and black rot or streak in rhizomes. Approximately 30 to 70% of the total production of wasabi rhizome has been affected by the disease annually, resulting in downgrading of the rhizome and lower market prices (Hu et al., 1986; Lo and Wang, 2001).

Transmission of black rot disease through vegetative propagation has been demonstrated as the primary cause of wasabi black rot disease and the most likely means of introducing the pathogen into previously uninfested areas, because farmers in Taiwan are accustomed to planting wasabi via vegetative propagation, such as tillers from parent rhizomes or plantlets from fibrous roots of a parent plant (Lo et al., 1990, 1991; Lo and Wang, 2001). This cultural practice has caused the disease to become endemic to all areas of Taiwan where wasabi is cultivated.

In addition to transmission by propagative materials, pycnidia produced by *P. wasabiae* either on infected plant debris or survival in soils may provide initial inoculum as pycnidiospores are released from pycnidia exposed to high humidity (>97% RH) and spread by splashing and wind-blown rain or sprinkler irrigation (Lo and Wang, 2000a, 2000b). Our previous study identified pycnidia and pycnidiospores as important sources of primary and secondary inoculum for black rot disease of wasabi (Lo et al., 1991; Lo and Wang, 2001). Lo and Wong reported that rainfall and irrigation may be important factors increasing the incidence of black rot of wasabi in the field, especially the leaf spot phase of the disease (Wang et al., 1992; Lo and Wang, 2000a, 2001). Consequently, reducing primary and secondary inoculums of *P. wasabiae* should be considered an important strategy for controlling wasabi black rot disease.

Management of wasabi black rot is a challenge in commercial production fields due to the limited efficacy of disease management strategies in Taiwan and Japan (Ozoe et al., 1971; Takuda et al., 1973; Goto and Matsumoto, 1986; Lo and Wang, 2000a). Takuda developed some control methods including fungicides against this disease sev-

*Corresponding author. Tel: +886-4-2877021; Fax: +886-4-2877585; E-mail: sdyeh@nchu.edu.tw

eral years ago (Takuda et al., 1973). However, these methods have not been used against black rot disease in the field because some fungicides were not effective and others were not approved for use in Taiwan (Cheng and Tung, 1986; Lo et al., 1990). Therefore, development of an effective strategy for controlling black rot disease of wasabi is urgently needed in Taiwan.

The objectives of this study were (a) to determine which fungicide would prevent the pathogen from spreading from the parent plants to plantlets, and (b) to assess whether the tillers from parent rhizomes of wasabi and the plantlets from fibrous roots of the parent wasabi treated with fungicide can effectively reduce the wasabi streak disease in the field. In addition, disease severity on pathogen-free plantlet transplants covered with a plastic sheet on the cultivated plots was also evaluated.

Materials and Methods

Plant Materials and Fungicides

The cultivar 70-I-1 of wasabi was used in this study (Hu et al., 1991). Vegetative materials, including plantlets such as tillers from parent rhizomes and root plantlets from fibrous roots of parent wasabi plants were provided by wasabi growers in Ali-shan and Ta-pang unless indicated otherwise. Rhizomes and root fragments of parent plants and plantlets from tissue culture provided by Hsin-Guo Co. (Hsin-Chu, Taiwan) were also used. Unless stated otherwise, rhizomes and fibrous root fragments from parent wasabi were grown in sterile sand (10 kg) in boxes (45 × 30 × 10 cm) and covered with 1 cm of sterile peat moss (2% Nitrogen, pH 6.0). Each box was moistened with 1800 ml of sterile distilled water at the beginning of each experiment. Water was added as needed during the 3-month incubation in a growth chamber at 15-18°C with a 12 h photoperiod to produce the tillers and root plantlets. The tillers and root plantlets were then transplanted to pots with sterile sandy loam and peat moss (1:2, pH 6.0). These plantlets were grown at 15-18°C with a 12 h photoperiod for 6 months in a growth chamber to obtain adult plants. Fertilizer (N:P:K=15:10:10) was added to the medium as needed. The fungicides used were thiabendazole [40% WP; 2-(4-thiazoly)-benzimidazole] (Merck & Co., Inc., Germany), difenoconazole [24.9% E.C; 1-(2-(4-(4-chlorophenolxyl)- 2-chlorophenyl)-4 methyl-1,3-dioxolan-2-yl-methyl)-1H-1,2,4-triazol] (Ciba-Geigy, Switzerland), polyoxin [2.2% WP; 1-5-n-carbamoyl-2-amino-2-deoxy-L-xylonyl-5-amino- 5-deoxy-(-D-allofulanosyluzonic acid)-5-hydroxyl methyl-uracil (Nihon Nohyaku Co., Ltd., Japan), and benomyl [50% WP; methyl-1-(butylcarbamoy)-2-benzimidazolecarbamate] (Dupont de Nemours & Co., Inc., Taipei, Taiwan).

Media

Potato dextrose agar supplemented with streptomycin sulfate (PDSA) and 2% water agar (WA) were used for isolating *P. wasabiae*. PDSA consists of potato dextrose broth 24 g (Difco Laboratories, Detroit, MI. USA), agar 20 g (Difco), streptomycin sulfate 300 mg (Sigma, Co., MO. USA), and distilled water 1000 ml.

Effect of Fungicide Treatment on Disease Incidence of Wasabi Plantlets Infected with Phoma Wasabiae

The efficacy of fungicides for eradicating the pathogen was analyzed by soaking the infected plantlets in fungicide solutions. The rhizomes and the fibrous roots of parent wasabi were cut with sterile knives into lengths of 0.8-1.0 and 3-5 cm, respectively. Samples infected by *P. wasabiae* were individually divided into five groups. Each group contained 100 rhizomes and 100 fibrous roots to be separately treated with either thiabenazole, difenoconazole, polyoxin or benomyl at their recommended concentrations of 400, 249, 55 and 1000 μg/ml, respectively. Each group was then soaked in each fungicide (200 ml) for either 0, 2, 4, 6, 8, or 10 h. Control (untreated) plants were treated similarly with sterile distilled water. After treatment, these test materials were air-dried in a hood for 1 h, then cut into fragments. Five tissue fragments (0.5 × 0.3 cm) taken randomly from each fibrous root or rhizome were placed onto a PDSA plate and incubated at 25°C for 3 days. The efficacy of the fungicides was evaluated by the presence or absence of the pathogen on PDAS-plates. The experiment was performed twice; data presented are from a representative test.

Effect of Fungicide Treatment on Black Rot Disease Control in Wasabi

Two hundred wasabi tillers were divided into four groups for treatment with either thiabendazole, difenoconazole, benomyl or sterile water as described earlier. Preliminary tests were performed to determine the best treatment duration for each fungicide. Treated tillers were then planted in sterilized peat soils as described and placed into a growth chamber at 15-20°C with a 12 h photoperiod provided by cool-white fluorescent lights. The peat soil was moistened to 18% water content and additional water was added as needed. The incidence of streak disease in wasabi tillers treated with fungicides was examined at 6 months after planting. Each tiller was cut into 0.5 × 0.3 cm cross sections. Five sections randomly taken from each tiller were incubated on a PDSA plate at 25°C for 3 days. When *P. wasabiae* from any section of a tiller appeared on PDSA media, the pathogen was considered to be present in the tiller. Mortality of the wasabi tillers after each fungicide treatment was also evaluated 14 days after planting. Analysis of variance and Duncan's $LSD_{0.05}$ test were used to analyze the data. The experiments were repeated twice. Data presented are from a representative test.

Disease Control with PE Cover in the Field

To understand the effect of pathogen dissemination by water on disease severity of black rot on wasabi leaves, polyethylene sheet covers (PE) were designed as tunnel to prevent the pathogen spread by windblown rain or water drops from the sprinklers in the field. The experimen-

tal field was located at Ta-pong, Chia-yi County, Taiwan. The plots were arranged in a randomized complete block design with three replications. The PE cover and control (uncovered) were treated as subplots (4×1 m). Each subplot contained 40 wasabi tillers. All plots were evaluated for leaf spot severity two months after planting. Ten wasabi plants were randomly selected to survey the disease severity and incidence of wasabi leaves. The disease incidence of leaf spot of wasabi were rated as previously described Lo and Wang, 2000a. The disease severity was measured as described below.

Disease Control with Tissue-Culture Plantlets in the Greenhouse

The tissue-culture plantlets of wasabi were transplanted into sterilized soils and infested field soils in pots in a plastic house in Mei-fong, Na-tou County, Taiwan. Each 15-cm pot had four tissue culture plantlets. Each treatment had three replicates. The disease incidence and severity of wasabi on leaves, petioles, and rhizomes were then recorded for six months after transplanting. Disease incidence was assessed by symptom appearance on rhizomes, petioles, and leaves of whole plants. Disease severity of leaf spot was rated on a scale of 0-4 where 0 meant no disease, 1, 1-10%; 2, 11-25%; 3, 26-50% and 4, ≥51% diseased area. Disease severity on the rhizome was recorded as the number of rhizomes with discolored vascular tissue /total number of wasabi rhizomes × 100%. The discolored vascular tissue of rhizomes was calculated as the total length of discolored vascular tissue minus the initial length of the rhizome black rot. Disease incidence on wasabi petioles was measured as the number of infected petioles/total number of wasabi petioles × 100%. The severity on infected petioles of wasabi was measured as the total length of the black tissue on petioles/ total length of the entire wasabi petiole.

Evaluation of Integrated Control of Disease in the Field

Two field tests were performed during 1991 to 1992 and 1993 to 1994 to evaluate the efficacy of fungicide treatment for control of black rot disease on wasabi platelets. Wasabi tillers were soaked in benomyl (1000 µg/ml) for 8 h, and then transplanted to the field. Control (untreated) tillers were similarly treated with sterile distilled water. To prevent pathogen dissemination by water droplets from sprinkler or rainfall, polyethylene sheet covers were used as tunnel in this experiment. The four treatments were benomyl-treated tillers covered with polyethylene sheets (PE); uncovered, benomyl-treated tillers; uncovered, untreated tillers; and covered, untreated tillers. Disease severity of rhizomes, tillers and petioles of adult plants (full-grown plants) of wasabi was rated 10 months after transplanting as described previously. In 1993 to 1994, the experiment was repeated, except that tissue culture plantlets were used instead of the benomyl-treated tillers. Data were recorded 18 months after transplanting. A completely randomized design with four replicates was used in 1991 to 1992 and 1993 to 1994.

Experimental Design and Data Analysis

All data were submitted to analyses of variance and Duncan's multiple range tests to separate the means using the Statistical Analysis System (SAS Institute Inc., Cary, NC) program.

Results

Effect of Fungicide Treatment on Spread of Pathogenin Wasabi Plantlets

The efficacy of fungicides for eradicating the pathogen was analyzed by soaking the infected plantlets in different fungicide solutions. All tested fungicides effectively eliminated the pathogen from tiller or root plantlets after a 10 h soaking. The minimum effective treatment periods for difenoconazole, thiabenazole, benomyl, and polyoxin were 2, 4, 8, and 10 h, respectively (Table 1).

Thiabenazole, difenoconazole, and benomyl were selected for further study to develop safe, efficient, and effective plantlet treatments. The effectiveness of the fungicides in agar plate tests was positively correlated with their ability to eradicate *P. wasabiae* from infected plant-

Table 1. Effect of different fungicide soakings on disease incidence of wasabi tissues infected by *Phoma wasabiae*.

Fungicide	Concentration (µg/ml)	Disease incidence (%)[1]											
		Treatment duration (h) for wasabi tissue											
		0		2		4		6		8		10	
		tiller	root	tiller	root	tiller	root	tiller	root	tiller	root	tiller	root
Thiabenazole	400	90	100	10	50	0	0	0	0	0	0	0	0
Polyoxin	55	90	100	70	80	30	50	10	10	10	10	0	0
Difenoconazole	249	90	100	0	0	0	0	0	0	0	0	0	0
Benomyl	1000	90	100	40	40	20	20	20	20	0	0	0	0
Control/(water)		90	100	90	100	90	100	90	100	90	100	90	100

[1]Each treatment consisted of 100 wasabi tillers and 100 root sections. The experiments were repeated twice, and data presented are from a representative test.

lets when difenoconazole, thiabenazole, and benomyl were used to treat wasabi plantlets for 2, 4, and 8 h, respectively. These fungicides significantly reduced the incidence of wasabi black rot disease. However, thiabenazole and difenoconazole increased the mortality of wasabi plantlets as compared with benomyl and control (Table 2).

Evaluation of Disease Control of Leaf Spot by PE Cover

From January to February, the disease incidence and severity of leaf spot of wasabi showed no difference between plots covered with PE sheet and untreated plots. However, the treatment of PE cover significantly reduced the disease severity and incidence of wasabi compared with uncovered plots after March, particularly after June (Figure 1).

Evaluation of Integrated Method for Disease Control in the Field

Tissue-culture plantlets were used to detect the infection sources of *P. wasabiae* in May-feng, Natou County. Wasabi rhizomes became infected by *P. wasabiae* when pathogen-free plantlets were planted in naturally infested soils, whereas no streak disease developed in wasabi rhizomes when plantlets were planted in sterilized soils (Table 3). No disease incidence occurred on wasabi leaves and petioles when the plantlets were covered with a PE sheet. However, the disease occurred on wasabi leaves and petioles without a PE cover (Table 3).

In field evaluations, benomyl effectively reduced the disease severity in rhizomes of tillers during the growing period (Table 4). However, treating of plantlets with benomyl alone was not sufficient to reduce the disease

Table 2. Effect of fungicide treatment on mortality and disease incidence in wasabi plantlets in growth chamber.

Fungicide	Concentration (μg/ml)	Mortality (%)[1]	Disease incidence of tiller (%)[2]
Thiabenazole	400	52a[3]	18b
Difenoconazole	249	28b	17b
Benomyl	1000	8c	21b
Control	0	4c	86a

[1]Data were taken 2 weeks after fungicide treatment.
[2]Data were taken 6 months after transplanting.
[3]Means followed by the same letter do not differ significantly (p=0.05) according to Duncan's multiple range test.

Table 3. Suppression of wasabi streak disease using tissue culture plantlets and polyethylene sheet (PE) cover in Mei-fong[1].

Treatment	Disease incidence (%)		
	Rhizomes	Petioles	Leaves
Sterilized soil + PE cover	0.0	0.0	0.0
Sterilized soil	0.0	4.2	12.5
Natural soil + PE cover	50.0	0.0	0.0
Natural soil	66.7	13.2	43.3

[1]The data were taken 6 months after transplanting.

Table 4. Effect of planting pathogen-free plantlets and a polyethylene sheet cover on disease severity on wasabi in a field in Tapang.

Treatment	Disease severity (%)							
	Adult plants							
	Tillers		Rhizomes		Petioles		Leaves	
	1992[1]	1994[2]	1992	1994	1992	1994	1992	1994
Treated plantlets with PE cover[3]	18.2d[4]	19c	12.5b	7.2c	37.5b	-	33.5b	37.1b
Treated plantlets without PE cover	43.1c	33b	12.5b	15.0b	73.5a	-	71.5a	68.7a
Untreated plantlets with PE cover	62.2b	38b	68.2a	54.0a	45.5b	-	40.2b	41.5b
Untreated plantlets without PE cover	91.0a	50a	75.0a	58.0a	73.5a	-	72.6a	74.5a

[1]Data were taken 10 months after transplanting tillers treated with benomyl.
[2]Data were recorded 18 months after transplanting tissue-culture plantlets. "-" = No data.
[3]Polyethylene sheet cover.
[4]Means followed by the same letter do not differ significantly (p=0.05) according to Duncan's multiple range tests.

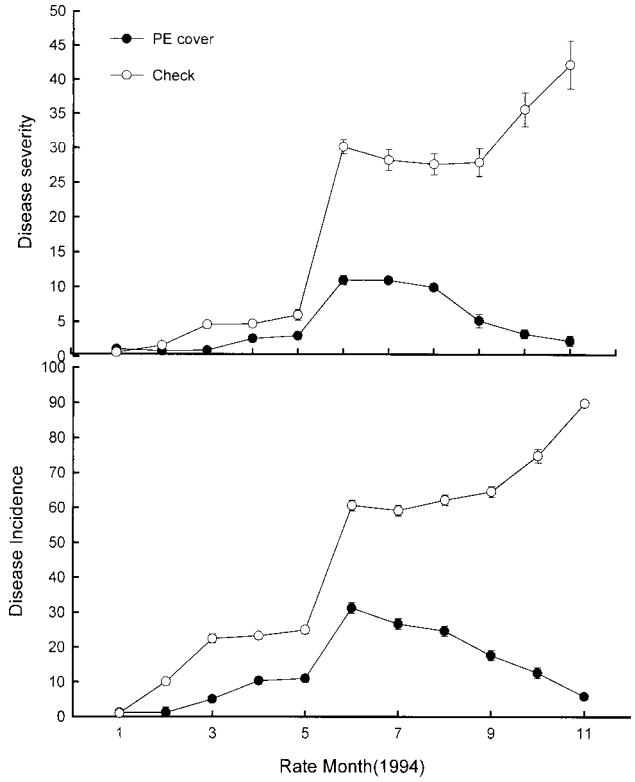

Figure 1. Effect of polyethylene sheet cover on disease severity and incidence of black rot of wasabi during 1994.

on wasabi leaves and petioles without the additional protection of a PE sheet cover in 1991-1992 (Table 4). Similar results were also found for tissue-culture plantlets planted in the field in Ta-pang that were not covered with PE sheets in 1993-1994 (Table 4). The best control was obtained by planting pathogen-free plantlets (benomyl treated tillers and tissue-culture plantlets) and covering them with a PE sheet (Table 4).

Discussion

Reducing of initial plant infection by soilborne pathogens is a reasonable strategy for controlling monocyclic diseases (Lo et al., 1996). Adams (1980) and Cooke and Logan (1984) indicated that tuber inoculum was more important than stem inoculum in the transfer of *Phoma foveata* to progeny tubers of potatoes. Our previous study also indicated that plantlet transmission of wasabi such as tillers and root plantlets was an important inoculum source for rhizome black rot disease (Lo and Wang, 2001). Results from this study demonstrated that usage of pathogen-free plantlets was an effective method for control of black rot disease of wasabi rhizomes.

Eliminating infected stems or tubers reduces the incidence of tuber contamination by *P. foveata* (Logan, 1974; Carnegie and Cameron, 1991). Planting the symptomless tiller plantlets was not effective in reducing disease incidence on rhizomes in fields, probably because *P. wasabiae* is a systemic pathogen and was present in wasabi tissues before the appearance of symptoms (Lo et al., unpublished data). Mercury and lime-sulfur were used to suppress disease caused by *P. wasabiae* (Nozu and Yokogi, 1932; Yokogi, 1952; Ozoe et al., 1971). However, these compounds may cause injury to wasabi plantlets and seeds (Ozoe et al., 1971). Results from this study indicated that thiabenazole and difenoconazole were as effective as benomyl in eradicating the pathogen from tillers or fibrous roots. However, difenoconazole and thiabenazole caused injury to the tiller and root plantlets at the recommended dosage. When benomyl was used to treat the infected wasabi plantlets, it also effectively reduced black rot disease on wasabi rhizomes but not leaves in the field.

Secondary infection with polycyclic diseases is also an important factor in disease progress when primary inoculum is low (Lo, 1996). Logan (1974) suggested that pycnidia of *P. foveata* could be an important source of inoculum for potato tuber contamination (Logan, 1974). Lapwood et al. (1978) asserted that the pathogen produced a watery brown rot in aging seed tubers in the ground, and pycnidia were formed rapidly when rotten tissue was placed in water. Pycnidia of *P. wasabiae* were quickly produced on PDA and rapidly released pycnidiospores when the culture was sprayed with water on PDA discs (Wang et al., 1992; Lo and Wang, 2000b). Similarly, water droplets from sprinklers or rainfall dispersed the pathogen and increased the disease development on wasabi in fields (Lo et al., unpublished results). In addition, presence of the pathogen in other hosts also affected the disease development (Lo and Wang, 2001). Consequently, preventing dispersal by splash droplets of rainfall may reduce disease incidence. Walker and Tisdal (1922) demonstrated that spread and disease development of black leg in cabbage seed beds depended on rainfall and that the average disease incidence of black leg was reduced from 50% to 2% when cabbage seedbeds were covered during rains in Wisconsin. Our previous studies showed that the disease development of wasabi streak disease was associated with rainfall and rainy days and that high relative humidity (over 97%) increased pycnidiospore release of *P. wasabiae* (Wang et al., 1992; Lo and Wang, 2000b). Results from these studies indicated that leaf spot of wasabi caused by *P. wasabiae* could be reduced by covering the plant plot with a polyethylene sheet (Figure 1). A small amount of disease occurring under the cover was probably due to splash-dispersal pycnidiospores from untreated plots during the rainy periods. Similarly, leaf spot and petiole blight of wasabi was significantly reduced in integrated control plots whereas there was no difference without a PE cover, even when tissue-cultured or benomyl-treated plantlets were used. Thus, reduction of secondary infection is also important in suppressing *Phoma* disease of wasabi.

Phoma wasabiae can survive as mycelia in plant residues or pycnidia in soils (Lo and Wang, 2001; Lo, unpublished data). The pathogen in soil may thus have a chance to serve as the source of initial inoculum. To reduce black rot disease of wasabi, it is necessary to eliminate the pathogen from soils (Table 3).

Based on our results, a possible strategy for control of wasabi black rot disease begins with soil fumigation to reduce the soil inoculum. Pathogen-free planting materials such as tissue-culture plantlets or fungicide treated tillers or root plantlets should be used for planting. Cultivation beds should be covered with PE sheets. Sprinker should also be replaced by a drip irrigation system to prevent pathogen dispersal.

Acknowledgements. This work was supported by a grant from the Council of Agriculture, Executive Yuan, Taiwan, ROC.

Literature Cited

Adams, M.J. 1980. The role of seed-tuber and stem inoculum in the development of gangrene in potatoes. Ann. App. Biol. **96:** 17-28.

Carnegie, S.F. and A.M. Cameron. 1991. The role of soil, seed-potato tuber and haulm in the transmission of *Phoma foveata* from infected seed to daughter tubers. Plant Pathol. **40:** 352-358.

Cheng, M.F. and B.K. Tung. 1986. Etiology and fungicide control of black rot of wasabi caused by *Phoma wasabiae.* J. Nat. Chia-Yi Inst. Agri. **13:** 1-10.

Cooke, L.R. and C. Logan. 1984. Further experiments with fungicide sprays for control of potato gangrene. Record Agri. Res. **32:** 43-46.

Goto, M. and K. Matsumoto. 1986. Causal agents associated with the internal black rot syndrome of Japanese horseradish. Ann. Phytopath. Soc. Japan **52:** 59-68.

Hu, M.F., S.M. Chiu, and S.Y. Liu. 1986. Effect of different environments on the growth and yield of *Wasabia japonica* Matsum. J. Agri. Res. China **35:** 291-299.

Hu, M.F., S.M. Chiu, C.T. Lo, S.Y. Liu, and C.C. Tu. 1991. Development of wasabi variety Tainung No. 1. J. Agri. Res. China **40:** 13-21.

Lapwood, D.H., R.I. Harris, and G.A. Hide. 1978. Isolation of *Phoma exigua* var. *foveata* from, and pycnidial formation on, brown watery rots of planted seed potato tubers. Trans. Brit. Mycol. Soc. **71:** 319-320.

Lo, C.T., K.M. Wang, C.C. Tu, and W.H. Tsai. 1991. Inoculum sources of wasabi streak disease and its control. Plant Prot. Bull. **33:** 434.

Lo, C.T. 1996. Biological Control of Turfgrass Diseases and Ecological Studies with *Trichoderma harzianum* Strain 1295-22. Ph.D. thesis, Cornell University, NY.

Lo, C.T. and K.M. Wang. 2000a. Survey of fungal diseases on aboveground parts of wasabi in Taiwan. Plant Pathol. Bull. **9:** 17-22.

Lo, C.T. and K.M. Wang. 2000b. Factors affecting pycnidial production and pycnidiospore germination of *Phoma wasabia,* the causal agent of wasabi. Plant Pathol. Bull. **9:** 99-106.

Lo, C.T. and K.M. Wang. 2001. Inoculum sources of black rot of wasabi, caused by *Phoma wasabiae*. Plant Pathol. Bull. **10:** 88-92.

Lo, C.T., E.B. Nelson, and G. E. Harman. 1996. Biological control of turfgrass diseases with a rhizosphere competent strain of *Trichoderma harzianum*. Plant Dis. **80:** 736-741.

Lo, C.T., K.M. Wang, C.C. Tu, and W. H. Tsai. 1990. Studies on streak disease of wasabi. Plant Prot. Bull. **32:** 345-346.

Logan, C. 1974. The effect of soil- and tuber-borne inoculum on the incidence of potato gangrene. Ann. Appl. Biol. **78:** 251-259.

Nozu, R. and K. Yokogi. 1932. Streak disease of wasabi. Ann. Phytopath. Soc. Japan **2:** 6.

Ozoe, S., T. Takuda, and T. Hirosawa. 1971. Effect of several fungicides for controlling the spread of *Phoma wasabiae* on wasabi leaves. Chugoku Nogyo Kenkyu **43:** 55-56.

Takuda, T., S. Ozoe, and T. Hirosawa. 1973. Streak disease control of wasabi. Ann. Phytopath. Soc. Japan **39:** 166.

Walker, J.C. and W.B. Tisdal. 1922. Further notes on the occurrence of cabbage black leg. Phytopathology **12:** 43.

Wang, K.M., C.T. Lo, C.C. Tu, and W.H. Tsai. 1992. Effect of temperature and humidity on the development of wasabi streak disease caused by *Phoma wasabiae*. Plant Pathol. Bull. **1:** 96-103.

Yokogi, K. 1952. Studies on the disease of wasabi and their control. Res. Bull. Shiamane Pref. Agr. Exp. St. Comm. Issu. 77th Aniv., Found. **3:** 1-70.

山葵黑心病之綜合防治

羅朝村　王貴美　胡敏夫　王鐘和

行政院農業委員會農業試驗所

利用炭克（24.9% E.C.）、腐絕（40% W.P.）、萬力（50% W.P.）及保力黴素（2.2% W.P.）等化學藥劑浸泡山葵根莖分孽苗與根苗，發現均可去除病原菌，惟所需浸泡時間不同；在其推薦濃度下有效時間分別爲2、4、8及10小時。由於炭克及腐絕二藥劑會影響山葵植株存活以及時間考量，僅萬力被用作田間之處理試驗。另外爲防阻雨水飛濺傳播病原及病害，隧道式塑膠布覆蓋被處理於田間觀察比較，結果顯示塑膠布覆蓋可降低山葵黑心病菌引起之葉斑病，病害可從90%降至5.9%。在田間綜合防治試驗，亦顯示若與對照組相較，單獨使用藥劑處理分孽苗雖可減少根莖黑心病但卻無法減輕葉斑點病。相反的，僅塑膠布覆蓋可減少葉、葉柄病害，但無法顯著降低根莖黑心病；唯有二者同時使用才能明顯降低山葵地下根莖及地上黑心病菌引起之病害。由以上資料建議農民可利用組織培養苗或經藥劑處理後之健康苗(pathogen-free plantlets) 栽種，並於雨季時節以塑膠布覆蓋等措施來減少山葵黑心病的危害。

關鍵詞：綜合防治；殺菌劑；山葵黑心病；山葵根莖。