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Radish (*Raphanus sativus* L.), a New Host of *Pseudomonas solanacearum* in Taiwan

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ABSTRACT

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In the recent years, a wilt disease was observed on radish in summer plantings in Shuiching area of central Taiwan. The characteristic symptoms of the disease were wilting of plants and water-soaking and brownish discoloration of the internal tissue of taproots. The disease was identified as a bacterial wilt caused by *Pseudomonas solanacearum*. Strains of this bacterium from radish induced wilting not only on radish, but also on solanaceous, cruciferous and other host plants, and could be classified as race 1. Two biovars occurred among these strains; the majority were biovar 4, with few biovar 3. Race 1, biovar 3 or biovar 4 strains of *P. solanacearum* were also detected from the roots of symptomless weed plants of black nightshade, green amaranth and common purslane. High temperature favored the disease development on radish. Under high temperature regime (35/30 C, day/night), the disease incidence was high over a wide range of soil inoculum density ($2.5 \times 10^3 - 3.3 \times 10^7$ cfu/g dry soil), whereas under low temperature regime (25/20 C), the disease incidence was low even at high inoculum densities. Thirteen commercial cultivars of radish were susceptible to *P. solanacearum* by the stem inoculation method, but showed some resistance by the soil infestation method in the greenhouse tests.

Key words: Radish, bacterial wilt, *Pseudomonas solanacearum*.

INTRODUCTION

Bacterial wilt caused by *Pseudomonas solanacearum* (Smith) Smith affects a wide variety of important crops throughout the world (14). In Taiwan, the disease occurs primarily on solanaceous plants, though a few other hosts have also been reported (9). In 1989, a wilt disease was observed on radish (*Raphanus sativus* L.), a cruciferous crop, in several fields in Shuiching area (Hsinshe, Taichung). The infected plants showed wilting; and the internal tissues of taproots became water-soaked and light brown discolored. Isolation of the suspect pathogen from the infected tissues yielded colonies characteristic of *P. solanacearum* on Kelman's tetrazolium chloride agar (TZC) medium (15), suggesting that the disease was a bacterial wilt caused by this bacterium. Presently, the disease on radish occurs only on the summer crop. Crucifers are rarely affected by the bacterial wilt caused by *P. solanacearum* (14), however, the disease affecting radish has been briefly described in Japan (19) and India (16). In this paper, we report the occurrence of bacterial wilt on radish in Taiwan and confirm that radish as a natural host of *P. solanacearum*.

MATERIALS AND METHODS

Bacterial strains

Twenty strains of *P. solanacearum* were isolated from infected radish plants collected from several fields located at Shuiching area during 1990-1992. Six strains of the bacterium were isolated also from the root systems of symptomless weed species collected from the radish fields (Table 1). Strains of *P. solanacearum* from other hosts (Table 6) were obtained from the collection maintained in our laboratory. All strains were stored as suspensions in sterile distilled water in screw-capped test tubes at room temperature.

Phenotypic characterization

Strains of *P. solanacearum* from radish and weeds were characterized by various physiological and biochemical tests. These tests were performed essentially as previously described (11), except nitrate reduction was determined by the method described by Fahy and Hayward (5). In addition, utilization of carbon sources, oxidative/fermentative test and production of 3-ketolactose were also tested according

to Fahy and Hayward (5). Lecithinase was determined by the method described by Dickey and Kelman (4). Levan formation was tested on nutrient agar containing 5% sucrose (5). Motility was determined by the method described by Leary and Chun (17).

Plants

The crop plants used for inoculations were radish (cv. Gantsai), tomato (cv. Known-you 301), tobacco (cv. Taiwan Tobacco No. 5), sweet pepper (cv. Blue Star), potato (cv. Kennebec), eggplant (cv. Pingtung Long), peanut (cv. Tainan No. 9), perilla (cv. Suo Mien Tzu Su), unknown cultivars of cabbage (*Brassica oleracea* var. *capitata* DC), pak-choi (*B. chinensis* L.), mustard (*B. juncea* Cosson), field mustard (*B. campestris* L.) and turnip (*B. rapa* L.). Three weed species were also used for inoculations. They were black nightshade (*Solanum nigrum* L.), green amaranth (*Amaranthus viridis* L.) and common purslane (*Portulaca oleracea* L.). Seeds of radish were sown directly in plastic bottles (8 cm in diameter, 18 cm in height) or plastic bags (10.5 cm wide and 23 cm long) containing loam soil, with three seeds in each bottle or bag. Twenty-one days old seedlings were used for stem inoculations. Tuber seed pieces of potato and seeds of peanut, cabbage, pak-choi, field mustard, mustard and turnip were also planted directly in loam soil in pots (11 cm in diameter, 9 cm in height); and one to three plants were allowed to develop per pot. Black nightshade, green amaranth and common purslane plants were collected from the fields and transplanted in pots with one plant per pot. Other host plants were seeded in BVB substrate (Bas Van Buren, No. 4, Maasland, Netherlands), and seedlings at 3–4 leaf stages were then transplanted into pots. All plants were grown in a greenhouse at 25–37 C or in a growth chamber with a photoperiod of 12 hr and regulated at day temperature of 30 C and night temperature of 25 C. Plants of about 15 cm high were used for inoculations.

Inoculation

Bacterial cells were streaked on TZC medium (15) and incubated at 30 C for 48 hr. Single fluidal colonies were used as the inocula for the stem-prick inoculation; or the colonies were further transferred to agar plates of medium 523 (13). After incubation at 30 C for 24 hr, bacterial cells were suspended in sterile distilled water and adjusted to an optical density of 0.3 at the wavelength of 620 nm. The resulting suspension contained about 10^8 cfu/ml when determined by dilution plating on TZC medium, and was used as the inoculum for soil infestation tests. For stem-prick inoculation, bacterial cells from colonies were picked with a sterile toothpick and inserted into the stem at the axil of the third or fourth leaf below the stem apex. The toothpick was left in position. Plants similarly pricked with sterile toothpicks were used as controls. For soil infestation test, the bacterial suspension was added to the loam soil at the ratio of 1:10 (v/w) and mixed thoroughly. With radish, seeds were planted directly in the plastic bags containing the infested soil, three seeds per bag. With other host plants, seedlings were transplanted into pots containing the infested soil, one seedling per pot. Seeds or seedlings planted in soil without *P. solanacearum* were served as controls. Ten to 15 plants of each host were inoculated with each strain of the bacterium for both inoculation methods. The experiment was repeated at least once. All inoculated plants were placed in the growth chamber at temperatures described above. Percentages of wilted plants were recorded 14 or 21 days after inoculation.

Detection of the pathogen in weeds

Weed species commonly found in the radish fields were collected from the Shuiching area during fallow period. None of the weeds showed visible symptoms. The root systems of each plant were washed with tap water, blotted dry, excised and macerated in 5 ml sterile distilled water. The prepared samples were then spread onto a modified SM 1 agar medium (6,20). Colonies of *P. solanacearum* formed on the agar plates were confirmed by streaking them on TZC medium for observing colony morphology characteristic of the bacterium, and by inoculating them to radish plants for observing the development of wilt symptoms.

Effect of temperature on disease development

Seeds of radish (cv. Gantsai) were planted directly in the plastic bags containing the loam soil infested with different concentrations of *P. solanacearum* Rd 4, three seeds in each bag. The infested soils were prepared by adding the bacterial suspension of 10^3, 10^4, 10^5, 10^6, 10^7, or 10^8 cfu/ml to the soil at a ratio of 1:10 (v/w); and the density of the bacterium in soil was determined by dilution plating on the modified SM 1 medium. All bags were placed in the growth chamber with a 12-hr photoperiod and regulated at 25/20, 30/25 and 35/30 C (day/night), respectively. The wilt symptom was recorded four weeks after seeding. The experiment was done twice, with 10–15 seedlings for each treatment in one experiment.

Susceptibility of radish cultivars to *P. solanacearum*

Disease reaction of various radish cultivars to the pathogen was tested by the stem inoculation and the soil infestation methods. For stem inoculation, seeds were sown directly in plastic bags (23 cm long, 10.5 cm wide) with three seeds per bag. Ten to 15 seedlings for each cultivar were inoculated by the stem-prick method as described above at one month after sowing. The percentage of wilted plants was recorded four weeks after inoculation. For soil infestation, seeds were also

planted directly in the bags containing soil infested with *P. solanacearum* Rd 4 at a density of about 10^7 cfu/g dry soil. Thirty seeds were sown for each cultivar, and percentages of wilting of germinated seedlings were determined four weeks after sowing. The experiment was conducted in a greenhouse at 25–37 C, and repeated once.

RESULTS

Symptoms

In the field, the symptoms of the disease usually appear at the taproot developing stage. Above-ground symptoms are manifest first in the water deficiency of lower leaves. As the disease progresses, the leaves become flaccid, drooped and discolored; and finally the whole plant wilted and died (Fig. 1A). Below-ground symptoms are first visible in the tip of taproot by showing browish discoloration in the vascular region and water-soaked. As the disease advances, the symptoms progress upward and finally most internal tissues of the taproot become water-soaked and brownish discolored (Fig. 1B). By squeezing, extensive bacterial ooze can be forced out of the cut surface of the infected taproot. *P. solanacearum* was consistently isolated from the infected tissues. Symptoms similar to those described for the naturally infected plants were produced by artificial inoculations with the isolated pathogen (Fig. 1C and 1D).

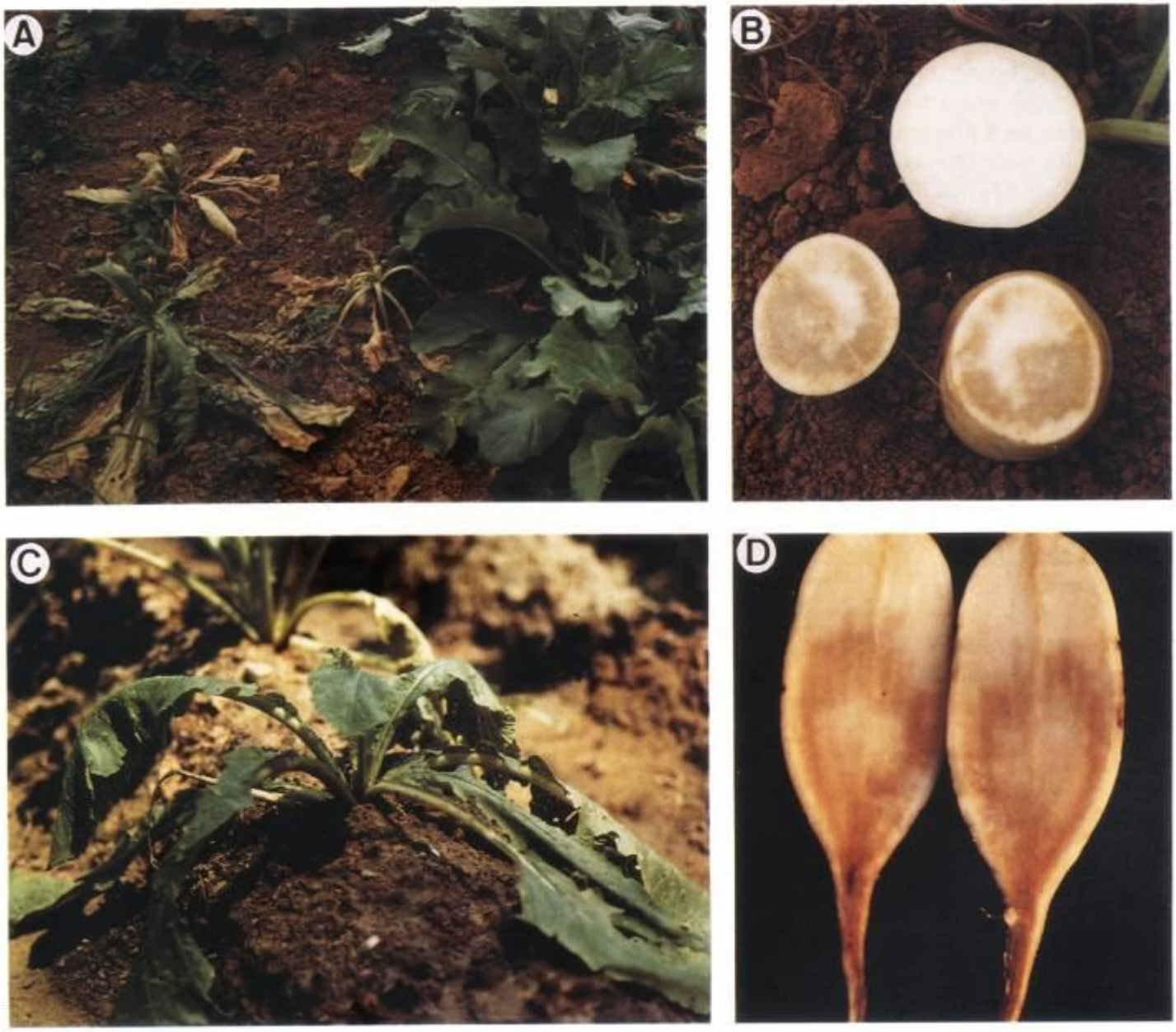

Fig. 1. Symptoms of bacterial wilt of radish. A, Wilting of naturally infected plants (left: wilted plant, right: healthy plants); B, Slices of naturally infected taproots showing water-soaking and brownish discoloration (upper: healthy taproot, lower: diseased taproot); C, Wilting of plants grown in soil infested with *Pseudomonas solanacearum* Rd9, and D, Slices of a taproot showing water-soaking and browning discoloration from an infected plant grown in soil infested with *P. solanacearum* Rd9.

Detection of the pathogen in weeds

Among 146 samples of weeds collected, *P. solanacearum* was detected in the roots from 2 of 17 samples of *A. viridis*, 3 of 24 samples of *P. oleracea* and 1 of 16 samples of *S. nigrum* (Table 1). Strains of the bacterium isolated from the six positive samples were designated as Gal and Ga2 from *A. viridis*, Cp1, Cp2 and Cp3 from *P. oleracea*, and Bnl from *S. nigrum*. These strains were used in some experiments.

Phenotypic characteristics

Twenty strains of *P. solanacearum* from radish and six strains from weeds were Gram-negative rods. They produced irregularly round or elliptical, fluidal, white colonies with pink or red centers on TZC medium. All strains showed the same reaction in the utilization of most substrates tested for growth. However, strains from radish were variable in the utilization of xylose; and strains from weeds were variable in the utilization of tryptophan and propionate (Table 2). Strains from radish in the substrate utilization test were also the same as strain Ps95 of *P. solanacearum* from tomato used for comparison, except that propionate was not utilized and tryptophan was utilized by strain Ps95. All strains metabolized carbohydrates (glucose and sucrose) oxidatively in the oxidative/fermentative test. Other physiological and biochemical properties of strains from radish and weeds were similar to those of strains Ps75 and Ps95 from tomato, which were used as reference strains (Table 3).

Among the 20 strains from radish, 3 strains (Rd2, Rd9, Rd15) were biovar 3 and the other 17 strains, were biovar 4 based on acid production from the oxidation of six carbohydrates (Table 4). Among the six strains from weeds, 4 strains (Bn1, Cp1, Cp2, Cp3) belonged to biovar 3, and the other 2 strains (Ga1, Ga2) belonged to biovar 4 (Table 4).

TABLE 1. Detection of *Pseudomonas solanacearum* in roots of weeds collected from radish fields in Shuiching, Taichung

Weed	No. of samples examined	No. of samples containing *P. solanacearum* in roots
Amaranthaceae 莧科		
Amaranthus viridis 野莧	17	2
Compositae 菊科		
Ageratum conyzoides L. 霍香薊	21	0
Bidens pilosa L. var. *minor* (Bl.) Sherff 咸豐草	8	0
Emilia sonchifolia (L.) DC. 紫背草	7	0
Cyperaceae 莎草科		
Cyperus rotundus L. 香附子	18	0
Cyperus compressus L. 莎田草	7	0
Gramineae 禾本科		
Cynodon dactylon Pers. 狗牙根	8	0
Eleusine indica (L.) Gaertn. 牛筋草	10	0
Portulacaceae 馬齒莧科		
Portulaca oleracea L. 馬齒莧	24	3
Rubiaceae 茜草科		
Borreria latifolia K. 闊葉破得力	10	0
Solanaceae 茄科		
Solanum nigrum L. 龍葵	16	1

TABLE 2. Utilization of substrates for growth of strains of *Pseudomonas solanacearum* isolated form radish and weeds

Substrate	Strain from		
	Radish (20 strains)	Weed (6 strains)	Tomato (Ps95[2])
D-Arabinose, LD-arabinose, fructose, D(+)-galactose, glucose, D(+)-mannose, saccharate, sucrose, D(+)-trehalose	+[1]	+	+
D(+)-Raffinose, α-L-rhamnose	−	−	−
D(+)-Xylose	V	+	+
β-Alanine, L-histidine, L-serine	+	+	+
Hippurate	−	−	−
L-Tryptophan	−	V	+
Benzonate, glycerol, lactate, D-tartrate	+	+	+
Ethyleneglycol, malonate	−	−	−
Propionate	+	V	−
Salicin	−	−	−
Azelate	+	+	+
Inositol	+	+	+
Levulinate	+	+	+

[1] +, positive reaction; −, negative reaction; V, variable reaction.

[2] strain Ps95 was used as a reference strain.

Pathogenicity

All 20 strains from radish were pathogenic to radish when the stems were inoculated. Depending on strains, 53.5–93.3% plants wilted 21 days after the stem inoculation. Inoculation of radish with two test strains Rd4 and Rd15 by the soil infestation method resulted in 30.8–38.5% wilted plants. Pathogenicity of strains from radish was also determined on several other hosts by the stem-prick inoculation. All strains were highly

TABLE 3. Physiological and biochemical properties of strains of *Pseudomonas solanacearum* isolated from radish and weeds

Property	Radish[1]		Weed[1]		Reference[2]	
	Biovar 3 (3 strains)	Biovar 4 (17 strains)	Biovar 3 (4 strains)	Biovar 4 (2 strains)	Biovar 3 Ps95	Biovar 4 Ps75
Accumulation of poly-β hydroxybutyric acid	+[3]	+	+	+	+	+
Arginine dihydrolase	−	−	−	−	−	−
Brown pigment	+	V	+	V	+	+
Catalase	+	+	+	+	+	+
Citrate utilization	+	+	+	+	+	+
Denitrification (gas from nitrate)	−	+	+	+	+	+
Fluorescent pigment	−	−	−	−	−	−
Gelatin hydrolysis	−	−	−	−	−	−
Growth at 4 C	−	−	−	−	−	−
41 C	−	−	−	−	−	−
H_2S from cysteine	−	−	−	−	−	−
Indole production	−	−	−	−	−	−
3-Ketolactose test	−	−	−	−	−	−
Kovas' oxidase	+	+	+	+	+	+
Lecithinase activity	−	−	−	−	−	−
Levan formation	−	−	−	−	−	−
Methyl red test	−	−	−	−	−	−
Motility test	−	−	−	−	−	−
Nitrate reduction	+	+	+	+	+	+
Pectate degradation	+	+	+	+	+	+
Phenylalanine deaminase	−	−	−	−	−	−
Phosphatase	−	−	+	−	+	+
Salt tolerance 1.0% NaCl	+	+	+	+	+	+
1.4% NaCl	+	+	+	+	+	+
1.7% NaCl	−	−	−	−	−	−
2.0% NaCl	−	−	−	−	−	−
Starch hydrolysis	−	−	−	−	−	−
Sulphatase activity	−	−	−	−	−	−
Tween 80 hydrolysis	+	+	+	+	+	+
Urease	+	+	+	+	+	+
Voges-Proskauer test	−	−	−	−	−	−

[1] See Table 4 for the biovar strains.
[2] Strains Ps75 and Ps95 from tomato were used as reference.
[3] +, positive reaction; −, negative reaction; V, variable reaction.

TABLE 4. Biovars of strains of *Pseudomonas solanacearum* isolated from radish and weeds from radish fields

Strain	Acid production from carbohydrates[1]						Biovar
	Lac.	Mal.	Cel.	Man.	Sor.	Dul.	
Radish							
Rd2, Rd9, Rd15	+[3]	+	+	+	+	+	3
Rd3, Rd4, Rd5, Rd6, Rd7, Rd8, Rd10, Rd11, Rd12, Rd13, Rd14, Rd16, Rd17, Rd18, Rd19, Rd20, Rd21	−	−	−	+	+	+	4
Weed							
Bn1, Cp1, Cp2, Cp3	+	+	+	+	+	+	3
Ga1, Ga2	−	−	−	+	+	+	4
Reference[2]							
Ps48, Ps95	+	+	+	+	+	+	3
Ps60, Ps75	−	−	−	+	+	+	4

[1] Lac, lactose; Mal, maltose; Cel, cellobiose; Man, mannitol; Sor, sorbitol and Dul, dulcitol.
[2] Strain Ps48 was isolated from tobacco, Ps60 from potato, Ps75 and Ps95 from tomato.
[3] +, positive reaction; −, negative reaction.

virulent to tomato, sweet pepper, eggplant and potato, but varied in virulence to tobacco and peanut. None of the strains were virulent to perilla (Table 5). Two of these strains, Rd4 and Rd15, were also pathogenic (inducing wilting) on cabbage, pak-choi, mustard, field mustard, turnip, black nightshade, green amaranth and common purslane.

Twenty-eight strains from 10 other hosts caused wilting of radish plants with the stem-prick inoculation; however, strains Ps9 and Ps73 from tomato and strains Ps103 and Ps104 from peanut did not induce wilting with the soil infestation method (Table 6).

The six strains from weeds were pathogenic to black nightshade, green amaranth and common purslane, and induced wilting of plants after stem inoculation. They were also highly virulent (induced 70–100% wilted plants) to radish, tomato, sweet pepper, eggplant and potato, but were not virulent to peanut and perilla.

Effect of temperature on disease development

The disease development was directly related to temperature and soil inoculum concentration. In general, the disease incidence increased with increasing temperature and inoculum density. Under a high temperature regime (35/30 C), over 80% of plants wilted with the soil inoculum concentration at or above 3.7×10^4 cfu/g dry soil; whereas under a low temperature regime (25/20 C), less than 30% wilted plants were obtained even with the inoculum concentration as high as 3.3×10^7 cfu/g dry soil (Fig. 2).

TABLE 5. Virulence of strains of *Pseudomonas solanacearum* from radish on other host plants

Strain	Virulence rating[1]						
	Tomato	Sweet pepper	Eggplant	Potato	Tobacco	Peanut	Perilla
Rd2, Rd9, Rd15	H	H	H	H	H	H	O
Rd16, Rd21	H	H	H	H	H	L	O
Rd5, Rd7, Rd8, Rd10, Rd11, Rd12, Rd14, Rd17, Rd20	H	H	H	H	H	O	O
Rd18, Rd19	H	H	H	H	M	VL	O
Rd3, Rd4, Rd6, Rd13	H	H	H	H	M	O	O

[1] Based on percentage of wilted plants 21 days after the stem inoculation. H, high virulence (more than 70% plants wilted); M, moderate virulence (more than 40% but less than 70% plants wilted); L, low virulence (more than 20% but less than 40% plants wilted); VL, very low virulence (less than 20% plants wilted); O, not virulent (none of plants wilted).

TABLE 6. Pathogenicity of strains of *Pseudomonas solanacearum* from solanaceous and other host plants to radish by the stem inoculation and the soil infestation methods

Strain	Host origin	Pathogenicity[1]	
		Stem inoculation	Soil infestation
20 strains	Radish	+	+
6 strains	Weeds	+	+
Ps76, Ps99, Ps100	Eggplant	+	+
Ps60, Ps92	Potato	+	+
Ps84, Ps87	Sweet pepper	+	+
Ps38, Ps44, Ps48	Tobacco	+	+
Ps9, Ps73	Tomato	+	−
Ps61, Ps69, Ps75, Ps95	Tomato	+	+
Ps103, Ps104	Peanut	+	−
Ps109	Peanut	+	+
Bp4, Bp6	Bird-of-paradise	+	+
Sa1, Sa7	Sugar apple	+	+
Pr40, Pr52, Pr55	Perilla	+	+
Bs10, Bs21	Strawberry	+	+

[1] +, pathogenic (induced wilting 14 days after stem inoculation or 21 days after planting into infested soil); −, not pathogenic.

Cultivar susceptibility

Inoculation of radish plants by the stem-prick method resulted in more than 76% wilted plants on 13 commercial cultivars tested. Therefore, all these cultivars appeared to be susceptible to stem inoculation. However, these cultivars showed some resistance to *P. solanacearum* when inoculated by the soil infestation method (Table 7). Cultivar Tapaigan-A2 had lowest disease incidence among the 13 cultivars tested.

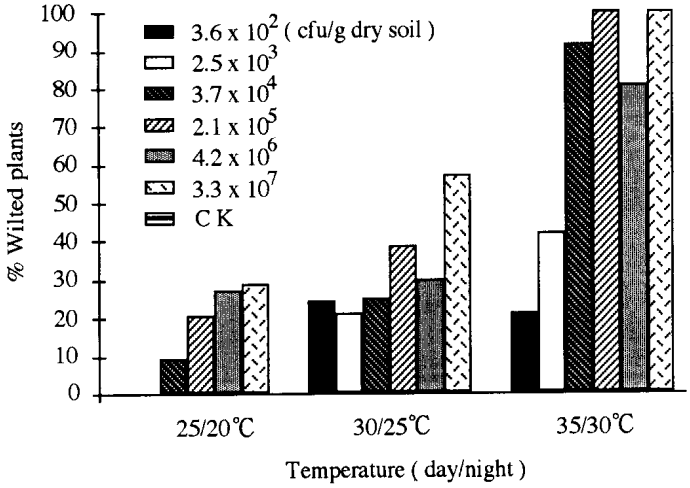

Fig. 2. Effect of temperature and soil inoculum concentration on development of bacterial wilt on radish 28 days after sowing in the soil infested with *Pseudomonas solanacearum* Rd4.

TABLE 7. Reaction of radish cultivars to *Pseudomonas solanacearum* Rd4 determined by the stem inoculation and the soil infestation methods

Cultivar	Percentage of plants wilted[1]	
	Stem inoculation	Soil infestation
KS (夏豐2號)	76.67 a[2]	36.35 abc
Chotou (卓頭)	90.00 abc	28.40 ab
Gantasi (矸仔)	96.67 bc	51.00 bc
Chunhsueh (春雪)	83.33 ab	34.95 abc
Tapaii (大白朴)	90.00 abc	48.65 abc
Minghoi (明和朴)	96.67 bc	53.10 bc
Tameihua (大梅花)	93.33 bc	35.00 abc
Meinung Tsaosheng (美濃早生)	96.67 bc	60.30 c
Nungsheng Gani (農生矸朴)	90.00 abc	27.75 ab
Tapaigan-A1 (大白矸A1)	96.67 bc	38.00 abc
Tapaigan-A2 (大白矸A2)	100.00 c	19.15 a
Tameihua-No1 (大梅花1號)	96.67 bc	29.60 ab
Tameihua-No2 (大梅花2號)	96.67 bc	37.30 abc

[1] Recorded 28 days after inoculation.

[2] Data are the mean of two experiments. Means within the same column followed by the same letter are not significantly different (P=0.05) by Duncan's multiple range test.

DISCUSSION

The results of this study revealed that the wilt disease of radish found in Shuiching area, the central part of Taiwan was caused by *P. solanacearum*. This bacterium has been reported previously to cause bacterial wilt on many solanaceous and other crops (9), but this is the first report of radish as a host in Taiwan. The bacterial wilt of radish at present occurs only on the summer crop and has not been widespread in other areas since the disease was first observed. However, in the past few years, radish was severely affected by the disease in several fields around Shuiching area so that the growing of radish crop has to be discontinued. The disease that has been observed in Shuiching was mostly in those fields where radish was rotated with sweet pepper and also was grown adjacent to the infected eggplant fields; thus, the high incidence of disease on radish probably was attributed to the high bacterial wilt pressure existing in the fields.

Strains of *P. solanacearum* have been separated into different races based on the hosts affected (1). All strains from hosts that have been reported previously in Taiwan belonged to race 1 (2,3,11,12,21), which has a wide host range. Strains from radish infected not only radish, but also tomato, sweet pepper, eggplant, potato, tobacco and peanut. Strains from solanaceous and other hosts tested also caused disease on radish. Thus, strains from radish and strains from other hosts were alike and could be classified as race 1. Although radish was the only cruciferous crop naturally affected by bacterial wilt in Taiwan, several crucifers tested were susceptible to artificial inoculation with *P. solanacearum*. Hence, bacterial wilt will be a potential problem on these crucifers if the inoculum density in soils is sufficient and the environmental condition in fields is favorable. Strains of *P. solanacearum* have also been classified into different biovars on the basis of the ability to oxidize three disaccharides and three hexose alcohols (7). Strains from radish could be separated into biovars 3 and 4. Biovar 3 was the predominant biovar reported in Taiwan (2,3,10,11,21), however, majority (17 out of 20) of strains collected from radish were biovar 4.

A number of weed plants are known to be infected with *P. solanacearum* with or without symptom expression. They may play an important role in the long-term survival of the bacterium in the absence of a susceptible crop (8,18). In this study, *P. solanacearum* was detected from root systems of plants of black nightshade, green amaranth and common purslane among 11 weed species collected from the radish fields during fallow periods. No visible symptoms were observed on all weeds found in the fields. However, wilting was induced on the three weeds when they were stem-inoculated with strains of *P. solanacearum* isolated from these weeds. These strains also were pathogenic

to radish and several other hosts tested. Furthermore, strains from weeds belonged to biovars 3 and 4, which was similar to the strains obtained from radish. The significance of the weed species in serving as a source of inoculum for the bacterial wilt of radish or in the survival of *P. solanacearum* in the absence of radish and other susceptible crops requires further study.

The results of this research showed that under the high temperature regime (35/30 C), the disease incidence was high over a wide range of soil inoculum density. Whereas under the low temperature regime (25/20 C), the disease incidence was low even at high inoculum densities, indicating that high temperature favored the development of bacterial wilt on radish. This was also true for bacterial wilt on many other crops, being most severe in the summer or warm-wet seasons in Taiwan (9).

Breeding or selection of cultivars for disease resistance is the main approach for managing bacterial wilt on various crops. The radish cultivars reacted differently to *P. solanacearum* with the two inoculation methods in the greenhouse experiment. All commercial cultivars tested were very susceptible when determined by the stem inoculation, but showed some degrees of resistance when tested by the soil infestation. Because radish plants are infected through roots by *P. solanacearum* in soils under the natural condition, the soil infestation is a better method of testing cultivar resistance than the stem inoculation. With the soil infestation, certain cultivars expressed resistance to *P. solanacearum*. They should be selected among the cultivars available for the summer planting to reduce the loss due to bacterial wilt.

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LITERATURE CITED

1. Buddenhagen, I., Sequeira, L., and Kelman, A. 1962. Designation of races in *Pseudomonas solanacearum*. Phytopathology 52:726. (Abstr.).
2. Chen, W. Y. 1978. Characteristics of strains of *Pseudomonas solanacearum* from tobacco plants in Taiwan. Bull. Taiwan Tob. Res. Inst. 9:71-80.
3. Denq, Y. J., Hsu, S. T., and Tzeng, K. C. 1993. Bacterial wilt of sugar apple: characteristics of the pathogen and susceptibility of Annonaceae to the pathogen. Plant Prot. Bull. 35:354-355. (Abstr.).
4. Dickey, R. S., and Kelman, A. 1988. Erwinia: "carotovora" or soft rot group. Pages 44-59 *in*: Laboratory Guide for Identification of Plant Pathogenic Bacteria. 2nd ed. N. W. Schaad, ed. American Phytopathological Society, St. Paul, Minnesota.
5. Fahy, P. C., and Hayward, A. C. 1983. Media and methods for isolation and diagnostic tests. Pages 337-378 *in*: Plant Bacterial Diseases, A Diagnostic Guide. P. C. Fahy and G. J. Persley, eds. Academic Press, Sydney, Australia.
6. Granada, G. A., and Sequeira, L. 1983. A new selective medium for *Pseudomonas solanacearum*. Plant Dis. 67:1084-1088.
7. Hayward, A. C. 1964. Characteristics of *Pseudomonas solanacearum*. J. Appl. Bacteriol. 27:265-277.
8. Hayward, A. C. 1991. Biology and epidemiology of bacterial wilt caused by *Pseudomonas solanacearum*. Annu. Rev. Phytopathol. 29:65-87.
9. Hsu, S. T. 1991. Ecology and control of *Pseudomonas solanacearum* in Taiwan. Plant Prot. Bull. 33:72-79.
10. Hsu, S. T., and Chen, J. Y. 1977. Physiological variation among isolates of *Pseudomonas solanacearum* from Taiwan. Plant Prot. Bull. 19:124-132.
11. Hsu, S. T., Hong, W. F., Tzeng, K. C., and Chen, C. C. 1993. Bacterial wilt of perilla caused by *Pseudomonas solanacearum* and its transmission. Plant Dis. 77:674-677.
12. Hsu, S. T., Tsai, T. T., and Tzeng, K. C. 1979. Pathovars of *Pseudomonas solanacearum* in Taiwan and their interaction in tobacco plants. Nat. Sci. Counc. Mon. ROC. 7:609-620.
13. Kado, C. I., and Heskett, M. G. 1970. Selective media for isolation of *Agrobacterium, Corynebacterium, Erwinia, Pseudomonas,* and *Xanthomonas*. Phytopathology 60:969-976.
14. Kelman, A. 1953. The bacterial wilt caused by *Pseudomonas solanacearum*. N. C. Agric. Exp. Sta. Tech. Bull. 99. 194 pp.
15. Kelman, A. 1954. The relationship of pathogenicity in *Pseudomonas solanacearum* to colony appearance on a tetrazolium medium. Phytopathology 44:693-695.
16. Kishun, R. 1983. *Raphanus sativus* and *Cosmos bipinnatus*-new hosts for *Pseudomonas solanacearum*. Indian Phytopathol. 36:742-743.
17. Leary, J. V., and Chun, W. W. C. 1988. *Bacillus*. Pages 120-127 *in*: Laboratory Guide for Identification of Plant Pathogenic Bacteria. 2nd ed. N. W. Schaad, ed. American Phytopathological Society, St. Paul, Minnesota.
18. Persley, G. J. 1986. Ecology of *Pseudomonas solanacearum*, the causal agent of bacterial wilt. Pages 71-76 *in*: Bacterial Wilt Disease in Asia and the South Pacific. G. J. Persley, ed. Proc. Int. Workshop PCARRD. ACIAR Proc. 13.
19. Takimoto, S. 1930. The bacterial wilt of radish. J.

Plant Protect. (Byochugai Zasshi) 17:794-797.
20. Tsai, J. W., Hsu, S. T., and Chen, L. C. 1985. Bacteriocin-producing strains of *Pseudomonas solanacearum* and their effect on development of bacterial wilt of tomato. Plant Prot. Bull. 27:267-278.
21. Yang, T. H., Hsu, S. T., and Tzeng, K. C. 1980. Studies on bird-of-paradise strains of *Pseudomonas solanacearum*-physiological characteristics, pathogenicity and lectin induction. J. Agric. For. 29:119-133.

摘　　要

林志鴻、徐世典、曾國欽．1994．蘿蔔 — 青枯病菌在台灣之新寄主．植病會刊3:147-155．(台中市　國立中興大學植物病理學系)

近年來在台中縣新社鄉水井地區，夏季種植的蘿蔔發生一種萎凋病害，其主要病徵爲植株發生萎凋及塊根內部呈現水浸狀及淡褐色之變化。本病害經鑑定爲 *Pseudomonas solanacearum* 引起的青枯病。由蘿蔔分離的青枯病菌菌株不僅引起蘿蔔植株的萎凋，也能造成多種茄科、十字花科及其他寄主植物的萎凋，因此可歸類爲第一生理小種 (race 1)。這些菌株含有兩種生物型，大多數爲第四生物型 (biovar 4)，少數爲第三生物型 (biovar 3)。由蘿蔔田間採集的龍葵、野莧及馬齒莧等三種雜草，雖無病徵，但由其根部也可分離到屬於第一生理小種、第三生物型或第四生物型的青枯病菌。高溫有利於蘿蔔青枯病的發生，在日夜溫度分別爲35及30 C時，土壤含病菌濃度在 $2.5 \times 10^3 - 3.3 \times 10^7$ cfu/g dry soil 範圍內引起之萎凋發生率均高，而在日夜溫度爲25及20 C時，即使土壤含病菌量高，萎凋發生率仍低。在溫室試驗，以莖部穿刺接種時，供試13種蘿蔔栽培品種的發病率均高，呈感病性，但以土壤混菌接種時，則發病率不高，表現某些程度的抗病性。

關鍵詞：蘿蔔、青枯病、青枯病菌。