

## Phytophthora-Induced Diseases on Citrus in Jeju Island

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*Phytophthora*-induced diseases on citrus in Jeju island have been considered of minor importance because of the use as rootstock of trifoliolate orange, which is immune to *Phytophthora*. However, brown rot on fruit, which severely occurred in 1998 and 1999, has become a great threat to citrus production in the island. About one-half of the surveyed orchards were infected in 1998 and 4 out of 19 infected fields showed over 20% fruit infection rate. The disease was less severe in 1999, with an estimated infected area and total fruit reduction of 3,155 ha and 15,300 tons, respectively. Typical gummosis was also occasionally observed on cv. Shiranuhi, which is mostly cultivated under plastic film houses. Two types of *Phytophthora* were consistently isolated from various plant parts, identified as *P. citrophthora* and *P. nicotianae*. The former was isolated from the aerial parts of the fruit, young leaf, and shoot in the fields. Meanwhile, the latter was only isolated from the basal stem showing gummosis in plastic film houses.

**Keywords :** brown rot, citrus, gummosis, *Phytophthora citrophthora*, *P. nicotianae*.

*Phytophthora* causing foot and root rot, crown rot, gummosis, and brown rot on citrus is known as the most important soilborne pathogen worldwide (Erwin and Ribeiro, 1996). As soilborne diseases of citrus, gummosis and rots on foot, root, and crown have been causing serious damage when susceptible scions and rootstocks are used. Meanwhile, the brown rot of fruit, which is largely affected by storm occurs not only during the growing season on the tree but also in packing houses or storage resulting in further losses (Graham and Menge, 2000).

Among the genus *Phytophthora*, *P. nicotianae*, *P. citrophthora*, *P. palmivora*, and *P. citricola* have been reported as the causal agents of gummosis on the trunk and of brown rot on fruit in citrus in many countries (Graham et al., 1998; Zitko et al., 1991). However, there is no report on *Phytophthora*-induced diseases on citrus in Jeju Island, which is the

major citrus production area in Korea. The diseases have been considered of minor importance in the island because of the use as rootstock of trifoliolate orange which is immune to *Phytophthora*. However, it is possible that *Phytophthora* diseases have now been occurring in the island due to the long cultivation and the more diversified cultivars of citrus being used recently.

However, outbreak of brown rot on fruit in 1998 and 1999 became a great threat to the production of citrus in Jeju island. Incidence of infected fruit was over 50% in some orchards, which led to abandoned harvesting (Table 1 and Fig. 1B). About one-half of the surveyed orchards were infected in 1998, with 4 out of 19 infected fields showing over 20% fruit infection rate (Table 1). The disease was less severe in 1999, with an estimated infected area and total fruit reduction of 3,155 ha and 15,300 tons, respectively (data not shown). Typical gummosis on basal trunk of citrus was also occasionally observed, especially on cv. Shiranuhi, which is mostly grown under plastic film houses (Fig. 2A). Characteristic symptoms of the gummosis are gum exudation from the trunk, discoloration of inner bark, and loss its vigor (Fig. 2A). The disease was only observed in cultivar, of which cultivation acreage has increased in Jeju island

In normal year, typhoons hit Jeju island from August to September. During this period, brown rot of fruit is most severe. The disease is usually observed 5-7 days after a heavy rainfall at the fruit maturing stage. However, it is occasionally observed on young fruits, twigs and leaves during prolonged rainy days. The brown rot of fruit is first

**Table 1.** Survey of citrus brown rot caused by *Phytophthora citrophthora* in Jeju island, 1998-2000

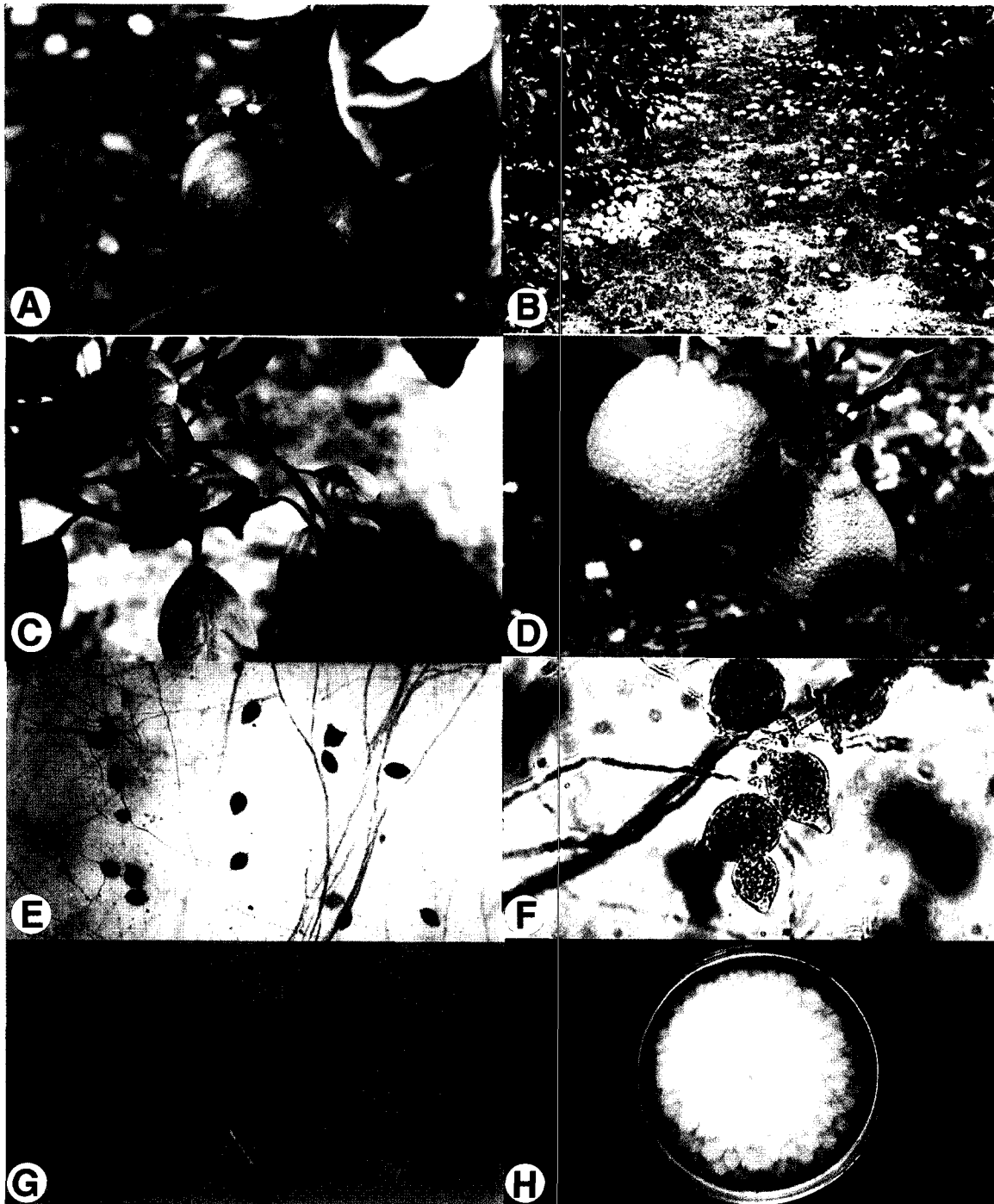
Surveyed year	No. of fields surveyed	No. of fields					
		Degree of infection rate (%)					
		0%	<1%	2-5%	6-20%	21-50%	>50%
1998	37	19	13	1	(1) <sup>a</sup>	1	(2)
1999	46	38	3	1	1	2(1)	(1)
2000	57	55	2	0	0	0	0

<sup>a</sup>No. of fields in parenthesis was due to the reports by growers.

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**Fig. 1.** Brown rot of Satsuma mandarin (A, B and C) and cv. Shiranuhi (D) caused by *Phytophthora citrophthora*, and sporangial features of the causal pathogen (E, F, and G) and colony pattern on PDA (H).

observed as a small light brown discoloration of the rind. The firm and leathery rot lesions enlarge irregularly without sunken or elevation, and become undistinguishable from that of adjacent healthy rind. At times, delicate white mycelia are formed on the rind surface under humid conditions. Infected fruits acquire characteristic pungent, rancid odor

and abscised soon after (Fig. 1A).

The causal agent of brown rot on fruit and gummosis on the citrus trunk was isolated and identified. For isolation, fruits, young leaves, and twigs showing brown rot were collected from the fields and cut into small pieces. After a short disinfection by using 70% ethanol, the plant tissues

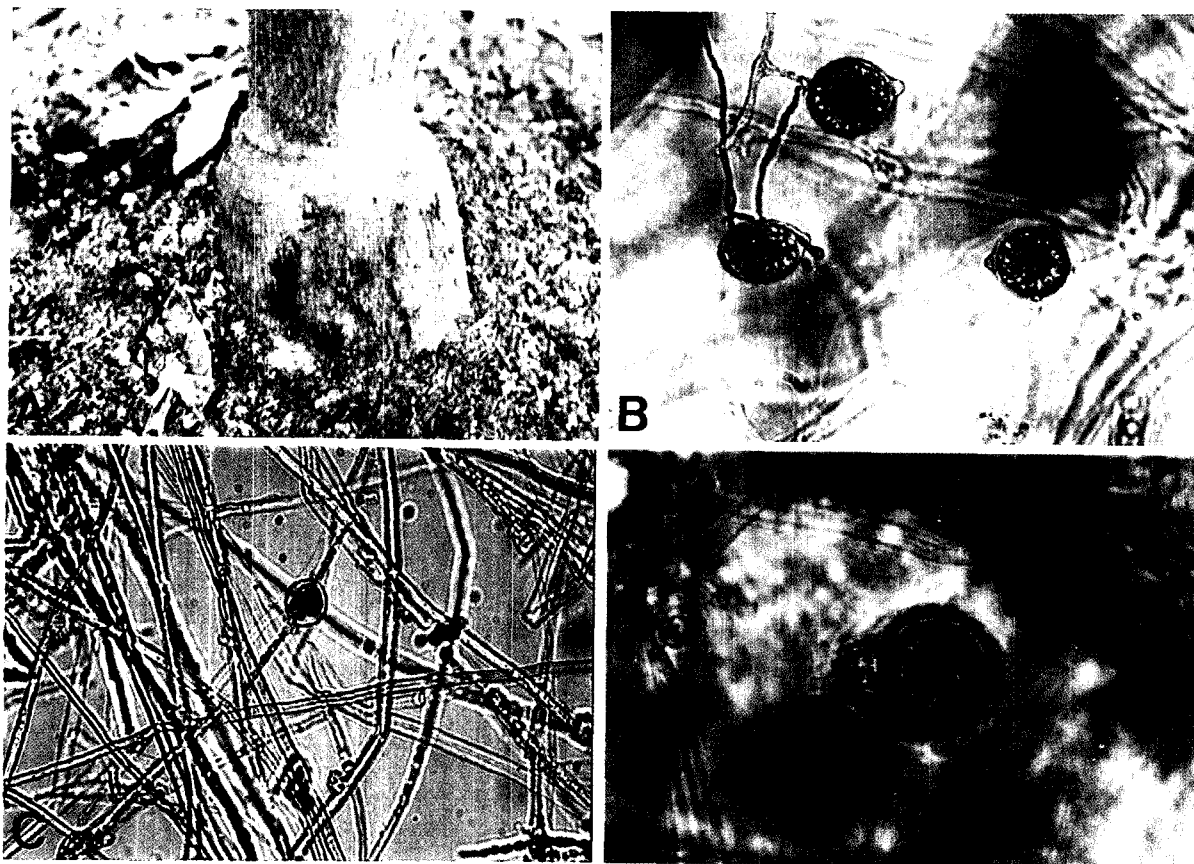


Fig. 2. Gummosis on trunk of citrus, cv. Shiranuhi (A) and the causal pathogen, *Phytophthora nicotianae*: sporangia (B), chlamydospore (C), and oospore (D).

were placed on a *Phytophthora* semi-selective medium containing pimarin 5 mg pimarin, 10 mg rifampicin, 100 mg rifampicin, 25 mg hymexazol, 100 mg PCNB, and 20 mg thiophanate-methyl in 1.0 L of corn meal agar. The plates were incubated at 25°C for 2-3 days and mycelia growing out from the tissues were cut and transferred into 10% V8 juice agar (100 ml commercial V-8 juice; 0.5 g CaCO<sub>3</sub>; 17 g agar; 900 ml distilled water) for further study.

To investigate morphological characteristics of the fungus, isolates were cultured on V-8 juice agar for 5-7 days, and agar disks (1 cm × 1 cm) were made by using a sterilized scalpel. The disks were transferred into a new Petri plate and soaked in sterilized water, then incubated at 25°C for 48-72 h for the production of sporangia as described by Jee et al. (1997). Effect of temperature on mycelial growth was examined on V-8 juice agar in the dark at 5°C intervals from 5 to 35°C. Mating types and sexual reproduction structures of the isolates were examined as described by Jee et al. (1997). Mating type standards of *P. nicotianae* and *P. capsici* were supplied by the Plant Pathology Division, National Institute of Agricultural Science and Technology (NIAST), RDA, Korea. After mating between the standard

and present isolates by dual culture at 20°C for 10-14 days, oospores were examined under a light microscope.

For the pathogenicity test, Satsuma mandarin was used. The fruit were surface-sterilized in 0.5% sodium hypochlorite for 5 min, washed three times with sterilized water, and air-dried under a clean bench. The fruits non-wounded or wounded with a needle were inoculated with an agar plug (5 mm in diameter) of each isolate, covered with a sterile cotton soaked with sterilized water, and sealed with parafilm to maintain enough moisture for infection. The fruits were incubated at 25°C in the dark and observed for disease development daily.

Two morphologically distinctive types of *Phytophthora* were isolated from the diseased tissues. The first type of isolates was collected from rotten fruits, young leaves and shoots of Satsuma mandarin and Shiranuhi, while the other type was from the gummosis on trunk. The first type produced petaloid colony patterns on PDA (Fig. 1H). Sporangia were semipapillate, variable in shape (ellipsoid, ovoid, globose etc), round base, and with a range of 35-60 × 27.5-35 μm (av. 48.3 × 26.3 μm) (Figs. 1E, F and G). The isolates did not produce oospores and grew between 4°C and

**Table 2.** Characteristics of *Phytophthora* causing brown rot or gummosis on citrus in Jeju island

Investigated characteristics	Features of representative isolates from		<i>P. citrophthora</i>	<i>P. nicotianae</i>
	Rots on young leaf, shoot, and fruit	Crown rot		
Sporangium formation	Single	Single	Single or in a loose sympodium	Single or in a loose sympodium
Papillium	Semipapillate	Papillate	Papillate and some semipapillate	Prominent papilla
Shape	Variable (ellipsoid, ovoid, globose etc.)	Variable (ovoid, spherical, etc.)	Extremely variable (ellipsoid, ovoid, globose, etc.)	Variable (ellipsoid, ovoid to spherical)
Base	Round	Round	Round	Round
Caducity	Some	Some	None	None
Size (µm)	35-60 × 27.5-35 (48.3 × 26.3)	36-50 × 24-40 (42.0 × 32.0)	27.0-65.3 × 18.9-40.4	40.2 × 28.5
L/B ratio	1.5	1.3	1.3-1.8	1.34
Chlamydospore	Intercalary, spherical	Abundant	Rare	Abundant
Cultural pattern (PDA)	Petaloid	Fluffy or cottony	Rosette or stellate on V8 agar	Arachnoid
Sexuality	No occurrence	Heterothallic, A1	Not occurrence	Heterothallic
Oogonium				
Shape		Smooth, spherical		Smooth, spherical
Size (µm)		25-30		
Oospore size (µm)		21-23		15-64 (26.8)
Antheridium		Amphigynous		Amphigynous
Growth at				
4°C	+	-	+	-
35°C	-	+	-	+

35°C (Table 2). These characteristics of the isolates are similar with those of *P. citrophthora* (Erwin and Ribeiro, 1996). The gummosis isolates produced fluffy aerial mycelia on PDA. Sporangia were papillate, variable in shape as ovoid or spherical, round base, and with a range of 36-50 × 24-40 µm (av. 42.0 × 32.0 µm) (Fig. 2B). They grew well at 35°C but did not thrive at 4°C, and produced abundant chlamydospores and oospores (Fig. 2C, D) when mated with A2 mating type standard. Antheridia were all amphigynous and sizes of oogonia and oospores were measured as 25-30 µm and 21-23 µm, respectively (Table 2). These characteristics of the isolates were similar with those of *P. nicotianae* (Erwin and Ribeiro, 1996) (Table 2).

All inoculated fruits of Satsuma mandarin either wounded or non-wounded showed rots within 7 days. The rot lesions enlarge more rapidly on the wounded than non-wounded fruits and *P. citrophthora* revealed stronger pathogenicity than *P. nicotianae* in the pathogenicity test on fruit.

Results indicated that two species of *Phytophthora* caused the diseases on citrus in Jeju Island. These are: *P. citrophthora*, which induced the brown rot on fruit and rots on young leaves and twigs; and *P. nicotianae*, which induced gummosis on the trunk. Although, *Phytophthora*-

immune trifoliolate orange is used as rootstock in the island, aboveground parts of most cultivars including Satsuma mandarin (Onshu) and Shiranuhi are not safe from *Phytophthora* diseases. Since outbreak of brown rot is largely affected by typhoon that often hits Jeju island during the growing season, development of control strategies for the diseases is urgently needed.

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