# Pythium spp. Associated with Crown Rot of Cucumbers in British Columbia Greenhouses

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**ABSTRACT** 

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Studies were undertaken to determine the causal agent responsible for a root disease ("crown rot") of cucumbers occurring in British Columbia greenhouses. Three Pythium spp.—P. aphanidermatum, P. irregulare, and Pythium sp. Group G—were regularly isolated from infected root crown tissue. A fourth species, P. coloratum, was isolated from grower propagation mix. Isolates representative of species were tested for pathogenicity against cucumber plants in greenhouse and growth chamber studies. All four species were pathogenic, as shown by their ability to reduce plant growth or cause death. Symptoms observed in commercial greenhouses were reproduced. P. aphanidermatum caused the most deaths in all studies, followed by P. irregulare and then Pythium sp. Group G. A disease survey in a commercial greenhouse revealed that plant-to-plant transmission of crown rot within sawdust bags occurred at a relatively low frequency. Of several potential sources of Pythium inoculum examined, peat-based propagation mixes used in B.C. greenhouses commonly contained Pythium spp., including those isolated from cucumber tissues infected with crown rot.

In British Columbia, Long English cucumbers (Cucumis sativus L.) are grown in greenhouses using a sawdust culture technique (1,10). In early January, the cucumber seeds are germinated in wooden flats containing either commercially prepared or custommade soilless potting mixes. When the seedlings have emerged (3-4 days after sowing), they are transferred to individual pots (12-15 cm diameter) until well rooted (14-25 days). Some growers prefer to seed directly into the pots, thus omitting the first transplant.

The propagation mixes used for germination and potting are composed mainly of peat and perlite. Varying amounts of vermiculite, sand, and/or sawdust may be added, depending on grower preference. Most growers use different mixes for germination and potting.

The young potted plants are transplanted in late January into 10-L plastic bags filled with sawdust and are fed a complete nutrient solution via a centrally supplied dripper system. Cucumbers are produced from March through October. After the plants are removed, the greenhouses are fumigated in preparation for the next growing season.

As sawdust culture became established in British Columbia during the late

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1970s, so did a new root disease, known in the local industry as "crown rot." Crown rot of cucumbers is distinguished by rotting symptoms confined mainly to the root crown area and basal 8-10 cm of the stem. The basal stems of infected plants appear chlorotic and whitish yellow during the early stages of infection. Brownish orange necrotic lesions develop along the stem as the disease progresses. Stem symptoms are accompanied by a rotting and yellow-to orange-brown discoloration of the root crown. The affected stems and roots are usually dry rather than water-soaked in appearance. Infected plants tend to produce few lateral roots from the crown region, but this is not easily noticed on older plants. Severely infected plants are weakly anchored and can be easily lifted out of the sawdust, indicating a general reduction of the root system.

Crown rot typically appears as a serious problem at early fruit set (8-12 wk after seeding) and/or during the late season on old plants. Plants rarely show symptoms before being transplanted into sawdust.

In general, the disease is sporadic both within and among greenhouses, and year-to-year variations are common. There also appears to be a host stress component to crown rot. Severe infections and patterns of infection often coincide with environmental conditions that can debilitate the plants.

The symptoms are often overlooked by growers, especially in greenhouses with large numbers of plants. As a result, infected plants are usually first noticed by a sudden wilting, particularly during

warm, sunny weather when water demands are greatest. Prolonged wilting usually results in death, although less severely infected plants occasionally recover

During the 1983 growing season, personnel from the B.C. Ministry of Agriculture and Food collected a number of infected root crowns from commercial greenhouses and isolated *Pythium aphanidermatum* (Edson) Fitzp. from some of the plants. Since that time, no further work on crown rot was carried out until the studies presented here were undertaken. The following studies were designed to clarify the etiology of cucumber crown rot in B.C. greenhouses.

## MATERIALS AND METHODS

Isolation of Pythium spp. Infected cucumber root crown tissues were collected from commercial greenhouses and examined under the light microscope. Oogonia, antheridia, and sporangia suggestive of Pythium spp. were often found. Infected tissues were washed thoroughly in distilled water, surfacedisinfested in 1% NaOCl for 30 sec, then plated onto water agar. Isolation of Pythium spp. was also attempted using the baiting technique of Waterhouse (22). Infected tissues were rinsed thoroughly, then placed in petri dishes containing tap water. Blades of lawn grass (Poa spp.) cut into 2-cm segments were boiled for 10 min, placed in the dishes containing the infected tissues, and incubated at 25 C. After incubation for 24-48 hr, the grass segments were examined for fungal growth and subcultures were made from hyphal tips.

The isolates were identified by J. S. Barr at the Biosystematics Research Institute, Ottawa, Canada. Three species were isolated from infected tissues and grower potting mixes: P. aphanidermatum, P. irregulare Buisman, and Pythium sp. Group G, Van der Plaats-Niterink. A fourth species, P. coloratum Vaartaja, was obtained only from grower potting mixes. The Group G isolate belongs to a group of Pythium spp. that do not produce sexual structures (oogonia and antheridia) in culture and do not appear to be heterothallic (21).

Pathogenicity of Pythium isolates. Pathogenicity of the isolates was evaluated in four separate experiments. Experiments 1-3 were conducted in

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growth chambers on cucumber seedlings, cv. Straight Eight, grown individually in disposable rectangular plastic pots ( $7 \times 6 \times 7$  cm) containing a medium consisting of Sunshine Mix, Blend No. 1 (Fison's Western Corp., Vancouver, B.C.) and a sandy loam soil, pH 6.8, mixed 2:1, v/v. In experiment 4, gynecious plants were grown individually in plastic bags containing sawdust, under conditions similar to those found in commercial greenhouses. The growth medium for experiments I and 2 was autoclaved for 1 hr before use. Unsterilized Sunshine Mix was used in experiment 3.

For experiment 1, inocula of P. aphanidermatum, P. irregulare, and Pythium sp. Group G were prepared from freshly harvested 2-wk-old mycelial mats grown in 100 × 15 mm plastic petri dishes containing 15-20 ml of V8cholesterol broth (V8CB) (2). Individual harvested mats were drained of broth, rinsed twice with distilled water, and then homogenized for 10 sec with 20 ml of distilled water in a blender at moderate speed. A preliminary experiment revealed that dry weights of 2-wk-old mycelial mats grown on V8CB were not significantly different (P=0.28) among the four Pythium spp. Therefore, a standard inoculum concentration of one 2-wk-old mat per 20 ml of distilled water was constant for all four species. Plants were grown from seed in sterilized growth medium contained in a  $60 \times 40 \times 7$  cm plastic tray. When 7 days old the seedlings were transplanted into individual pots containing the same medium. Fungal inoculum, containing mycelium plus oospores, was applied as a drench (10 ml per pot) to the surface of the growth medium around the base of each plant 8 days after transplanting. Distilled water was applied to control plants. There were 20 pots for each treatment, contained in four replicate plastic trays  $(17 \times 15 \times 7 \text{ cm})$  each containing five

For experiment 2, seeds were sown directly into the  $7 \times 6 \times 7$  cm pots; otherwise, plant growth conditions and time and method of inoculation were similar to those of experiment 1. Inocula, prepared as described for experiment 1, included *P. coloratum* and the three species used in experiment 1. In addition, inoculum levels of  $0.5 \times$  and  $0.25 \times$  for *P. aphanidermatum* were obtained by diluting the fungal homogenate with distilled water, and levels of  $2 \times$  for *Pythium* sp. Group G were obtained by doubling the number of mats homogenized per unit volume of water.

For experiment 3, plants were sown into potting mix previously infested with *Pythium* oospores. *P. aphanidermatum* and *P. irregulare* were chosen for study because of the relative ease with which oospores of these species could be produced in culture. Fungal mats produced on V8CB were comminuted in

distilled water, and the resulting suspension was filtered through four layers of cheesecloth. Oospore populations in the filtrate were estimated with a hemacytometer. The oospore suspensions were added separately to unsterilized mix and mixed thoroughly. The infested mixes were then incubated in plastic bags for 7-10 days in the dark at 25 C. An equal volume of distilled water was used for the control mix treatment. The oospore concentration for each mix was estimated by dilution plating on Mircetich's Pythium-selective medium (MVPM) (11), and the inoculum level adjusted to approximately 2,000 propagules per gram dry weight by dilution with uninfested mix previously analyzed to confirm the absence of Pythium spp. The mixes were placed in pots similar to those used in the previous two experiments. Half of the pots containing infested mix were then drenched with 20 ml of metalaxyl solution (10 µg a.i./ml, prepared from Ridomil 2.4 EC). Control pots and Pythium-alone treatments received 20 ml of distilled water. The following five treatments were compared in experiment 3: unamended control mix, P. aphanidermatum-infested mix with and without a metalaxyl drench, and P. irregulareinfested mix with and without a metalaxyl drench. Two days after drenching, cucumber seeds were sown individually into the plastic pots, which were then placed in trays as described earlier. For each treatment, there were eight replicate trays containing six pots of a given treatment. The trays were arranged in a randomized complete block design.

Plants used in experiments 1, 2, and 3 were grown on a 16-hr photoperiod at 25 C and watered from below every 2-3 days. Mortality was recorded at intervals, and surviving plants were harvested and shoot dry weights determined 40 and 45 days after inoculation in experiments 1 and 2, respectively, and 60 days after sowing in experiment 3. Also, at harvest, pieces of root crown tissue were dissected from one or two plants selected at random per replication from each treatment, surface-sterilized in 1% NaOCl for 1 min, and plated onto water agar. Transfers from hyphal growth occurring on water agar plates were made to V8CB for production of pure cultures for identification.

For experiment 4, seeds of gynecious Long English cucumber, cv. Farona, were sown directly into 10-cm-diameter plastic pots containing a mixture of peat and softwood sawdust (1:1, v/v). Plants were grown under a 16-hr photoperiod on greenhouse benches at 24 C and were inoculated 17 days after sowing (at the two- to four-true-leaf stage) with a suspension of mycelium-free *P. aphanidermatum* oospores. For inoculation, four wells approximately 1 cm deep were

made in the potting medium around the base of each plant 1-2 cm from the stem. Each treated plant received approximately 8,000 oospores in a 10-ml suspension distributed evenly among the four wells. Controls received 10 ml of distilled water. Plants were transplanted into 10-L plastic bags of sawdust 4 days after inoculation. The bags were arranged in a randomized complete block design with 12 plants per treatment. Cucumber fruit was picked when ripe at regular intervals for 24 days, and yields were recorded. At final harvest, root tissues were collected, surface-sterilized, and plated on water agar and MVPM.

Survey of disease distribution in a commercial greenhouse. During August 1985, a survey was conducted in a commercial cucumber greenhouse with a high incidence of crown rot to estimate the relative virulence of the crown rot pathogen(s). The greenhouse culture was atypical in that the grower had placed two plants in each sawdust bag rather than one. The presence of two plants in each bag presented an opportunity to compare the number of bags with zero, one, and two plants with crown rot with the number in each class predicted by the hypothesis that the probability of a plant becoming infected was independent of the condition of its "bagmate." Thus, each bag received a rating of 0, 1, or 2. Only those bags containing the originally transplanted two plants, or remnants thereof, were rated. A total of 2,914 bags were examined and rated according to the number of plants unambiguously killed by crown rot or showing obvious symptoms of the disease.

Sources of *Pythium* inoculum in greenhouses. Three potential sources of *Pythium* inoculum were examined: propagation mixes, nutrient solutions/irrigation water, and dark-winged fungus gnats (Diptera: Sciaridae).

Samples of propagation mixes were collected from 17 cucumber growers in Fraser Valley at four times between seeding and transplanting into sawdust bags. In order to detect Pythium in mix samples, 20 ml of mix was placed into 60 × 20 mm petri dishes and saturated with distilled water. There were four such dishes for each sample. Disks (1 cm) were then cut from cotyledons of 7-day-old cucumber seedlings and placed into the saturated mix. There were five disks (baits) per dish (20 per mix sample). The baits were incubated in the mix for 24 hr at 24 C, then removed, rinsed individually with sterile distilled water, blotted dry on filter paper, and placed directly onto MVPM. Five baits from each dish of mix were placed onto one plate of the Pythium-selective medium. Plates containing baits were incubated for 24 hr at 24 C, then examined under a dissecting microscope for mycelium growing from the edges of the baits. The number of bait disks showing growth after 24 hr was

recorded for each sample. Plates were reexamined after 48 hr, at which time isolations were made from hyphal tips for identification. *Pythium* isolates were identified from transfers placed onto V8CB and further identified to species.

Three growers in the lower Fraser Valley with a history of crown rot were selected for analysis of the water used in their greenhouses for the possible presence of Pythium spp. Submersible traps were made by placing autoclaved cucumber, sesame, and sunflower seeds (10 of each) into the bottom halves of 60 × 20 mm plastic petri dishes. Nylon mesh was then stretched over the open side of the dishes and tied with plastic-coated wire. Traps were taken to the greenhouses and submerged in nutrient tanks. For one grower, traps were also placed in an open, outdoor irrigation pond from which water was taken for use in his greenhouses. The other two growers used city water supplies or wells. The temperature of nutrient solutions in the tanks ranged between 28 and 30 C. Fifteen traps were submerged in each tank and the irrigation pond for 48 hr. The contents of the traps were then removed, rinsed with distilled water for 30 min, blotted dry on filter paper, and plated onto MVPM and water agar. Plates were incubated at 20 C in the light for 24-72 hr, and transfers were made from hyphal tips onto fresh plates of MVPM.

The possibility of dissemination of *Pythium* propagules by adult darkwinged fungus gnats (Diptera: Sciaridae) was also examined because there is concern in the B.C. greenhouse industry

regarding the role of fungus gnats as potential pests, including their possible influence on fungal root pathogens. Gnats were collected with an aspirator from the sawdust surface of bags that either contained or were in the vicinity of plants affected by crown rot. Approximately 250 gnats were collected from each of the greenhouses where water sampling was conducted. The gnats were stunned with CO<sub>2</sub> before being plated onto water agar. Plates were incubated at 25 C in the light. Open plates of MVPM were also placed on the sawdust surface of the bags containing plants previously showing crown rot symptoms. The plates were left exposed in situ for 24 hr before collection and examination.

### RESULTS

Pathogenicity experiments 1 and 2. Both experiments using the mycelium/ oospore inoculum drench in sterilized potting mix gave similar results. Plants began to wilt and die within 10 and 14 days after inoculation with P. aphanidermatum and P. irregulare, respectively. Additional plants died until at least 4 wk after inoculation with either fungus. None of the plants in uninoculated control pots died. P. aphanidermatum was more pathogenic than P. irregulare in both experiments. In experiment 1, plant mortality in P. aphanidermatum and P. irregulare treatments was 100 and 65%, respectively. In experiment 2, mortality was observed in P. aphanidermatum, P. irregulare, and Pythium sp. Group G 2× dose treatments (Fig. 1). No mortality occurred in Group G 1× dose, P. coloratum, and P. aphanidermatum

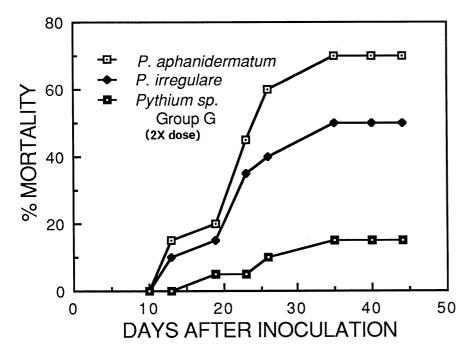


Fig. 1. Percent mortality over time of cucumber plants grown in sterilized potting medium and inoculated with *Pythium* spp. using mycelium/oospore drench at varying dosages (experiment 2).

 $0.5 \times$  dose and  $0.25 \times$  dose treatments.

A brownish orange discoloration was apparent on the roots of most of the plants in the P. aphanidermatum and P. irregulare treatments. This discoloration often extended along the basal 2 cm of the stem; it was usually preceded by a slight swelling and chlorosis of the basal stem and the production of small adventitious root primordia. Surviving plants in P. aphanidermatum and P. irregulare treatments were stunted and had reduced root growth compared with controls. Only a slight basal stem chlorosis and swelling occurred on a few of the plants in the Pythium sp. Group G treatments; most of the plants appeared symptomless. Despite the lack of symptoms, shoot dry weights of plants receiving Group G treatments were significantly reduced by the end of the experiment (Table 1). Plants inoculated with P. coloratum and those receiving the  $0.5\times$  and  $0.25\times$  doses of P. aphanidermatum also showed only slight or no visible symptoms, but shoot dry weights of these plants were all significantly less than those of uninoculated control plants (Table 1).

Pathogenicity experiment 3. At 10 days, there were no significant differences in seedling emergence among the treatments (P = 0.38) but wilting and discolored stems were apparent on some of the *Pythium*-treated plants.

Plant mortality over time was less than was observed in experiments 1 and 2, but the relative difference in level of

Table 1. Mean shoot dry weights of surviving *Pythium*-inoculated cucumber plants 40 (experiment 1) and 44 (experiment 2) days after inoculation with mycelium/oospore inoculum drench

Treatment	Mean shoot dry weight <sup>x</sup> (g)
Experiment 1	
Control	4.34 a <sup>y</sup>
Pythium sp. Group G	2.87 b
P. irregulare	1.81 c
P. aphanidermatum	×
Experiment 2	
Control	2.85 a
P. aphanidermatum $(0.25 \times dose)$	2.31 b
P. aphanidermatum $(0.5 \times dose)$	2.30 b
P. coloratum	2.20 b
Pythium sp. Group G	1.94 b
Pythium sp. Group G ( $2 \times dose$ )	0.80 c
P. irregulare	0.64 c
P. aphanidermatum	0.53 c

<sup>\*</sup>Mean weight per plant for surviving plants from four replicate trays each containing five pots of one plant per pot. Coefficients of variation for treatments in experiments 1 and 2 are 23.8 and 28.1%, respectively.

y Means followed by the same letter for a given experiment are not significantly different (P = 0.05) according to Student-Newman-Keuls test.

<sup>&</sup>lt;sup>z</sup> All plants died.

mortality between the two species was similar to that observed in the previous experiments (Fig. 2). No mortality occurred in uninoculated controls or the metalaxyl + inoculated treatments. Twenty days after sowing, however, a number of metalaxyl-treated plants showed some basal stem chlorosis or necrosis. Control plants showed no overt signs of infection throughout the experiment.

Dry weights of Pythium-treated plants were significantly less than those of control plants and plants treated with metalaxyl + Pythium (Table 2). P. irregulare-inoculated plants suffered the greatest reduction in growth, although few plants actually died. Surviving plants from the P. aphanidermatum treatment also showed reduced growth compared with controls. Treatment with metalaxyl enhanced the growth of plants beyond that observed in controls. Both Pythium spp. were reisolated from infected tissues of inoculated plants by plating on MVPM. Pythium spp. were not recovered from control plants plated on MVPM, whereas they were recovered from plants treated with metalaxyl + Pythium.

Pathogenicity experiment 4. About half of the *Pythium*-treated plants began to show symptoms within 15 days after inoculation. Infected basal stems were initially chlorotic and eventually showed a yellow-orange necrosis. Necrotic basal stem lesions retained turgidity and did not become water-soaked. Stem symptoms were accompanied by wilting in about half of the infected plants. Two infected plants previously showing wilt symptoms

completely collapsed and died 24-27 days after inoculation. After that time and throughout first fruit set (approximately 32-40 days after inoculation), no further symptoms developed. Surviving plants did not wilt during sunny weather, and stem infections gradually became calloused and dry. Student's t test analysis showed a significant difference (P = 0.04) between yields of Pythiumtreated plants and controls. Mean numbers of fruit per plant for control and Pythium treatment were 12.08 and 8.1, respectively. These figures include yields of zero for the two treated plants that died just before first picking. P. aphanidermatum was reisolated from root crown tissue of nine of the 10 surviving inoculated plants, whereas Pythium spp. were not isolated from root tissue of control plants.

Greenhouse survey of disease distribution. Of the 2,914 bags examined, 2,617 had no plants infected with crown rot, 221 had one plant infected, and 76 had both plants infected (Table 3). The proportion of infected plants (p) of the 5,828 plants examined was 0.064. The null hypothesis that infection was random and noncontagious was tested using a binominal expansion for a sample of n = 2 for each bag. Thus, expected proportions of infection were calculated using the formula  $p^2 + 2p(1-p) + (1-p)^2$ = 1, where  $p^2$  = the probability of finding two infected plants per bag, 2p(1-p) =the probability of finding one infected plant per bag, and  $(1-p)^2$  = the probability of finding zero infected plants per bag. On the basis of the chisquare analysis of the observed and expected frequencies, the hypothesis of independent infection can be rejected (*P* <0.001), suggesting that crown rot is either contagious or is associated with an unidentified "bag factor." If crown rot is assumed to be contagious, it is not highly so, given the relatively high ratio of single-plant to double-plant infections found in the survey.

Sources of inoculum. Of 48 propagation mix samples analyzed by baiting, 20 were positive for the presence of *Pythium* spp. All four *Pythium* spp. described earlier were isolated during this analysis, and a number of mix samples contained more than one species. Of the 11 different mix types used by the 17 growers sampled, 10 were peat-based, of which eight contained detectable levels of Pythium. The only nonpeat propagation medium used was vermiculite, and Pythium spp. were not detected in that sample. All but one of the commercially prepared mixes used by Fraser Valley cucumber growers, as well as three peat-based custom mixes, contained Pythium spp. The mixes that were positive for Pythium were obtained from 11 of the 17 growers surveyed. The use of thiram-treated seed by some growers appeared not to influence the

**Table 2.** Mean shoot dry weights of surviving cucumber plants grown in media infested with *Pythium irregulare* or *P. aphanidermatum* oospores with and without metalaxyl drenches

Treatment	Mean shoot dry weight <sup>x</sup> (g)	
P. irregulare + metalaxyl <sup>y</sup>	1.48 a <sup>z</sup>	
P. aphanidermatum + metalaxyl	1.48 a	
Uninfested controls	1.26 b	
P. aphanidermatum	1.11 c	
P. irregulare	0.90 d	

<sup>&</sup>lt;sup>x</sup>Mean weight per plant for 60-day-old plants from eight replicate trays each containing six pots of one plant per pot. Coefficient of variation for treatments is 20.2%.

Table 3. Summary of greenhouse survey data showing observed and expected frequencies (with associated chi-square values) of the number of sawdust bags containing zero, one, or two plants with crown rot

Plants/bag with			
crown rot	Observed	Expected	$\chi^2$
0	2,617	2,553	1.64
1	221	349	47.02
2	76	12	343.84
Total	2,914	2,914	392.50
			(P < 0.001)

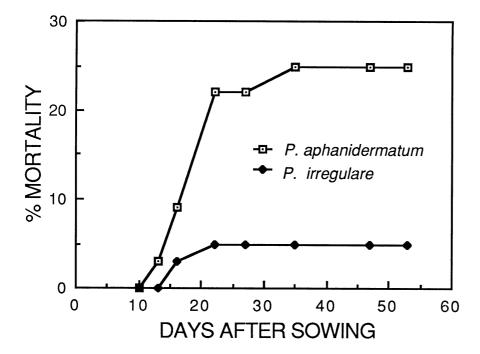


Fig 2. Percent mortality over time of cucumber plants after seeding into nonsterilized potting mix previously infested with *Pythium* spp. oospores (experiment 3).

<sup>&</sup>lt;sup>y</sup>Each plant was drenched with 200 μg a.i. of metalaxyl (prepared from Ridomil 2.4 EC).

<sup>&</sup>lt;sup>2</sup> Means followed by the same letter are not significantly different (*P* = 0.05) according to Student-Newman-Keuls test.

ability to detect *Pythium* spp. in these samples.

Fungal growth from trap baits used to assess water systems was observed on MVPM and water agar plates representing two of the three locations sampled. Colonization was lower in samples collected from growers using city water supplies (0-11%) than in samples collected from the one grower using an open irrigation pond (86%). Isolations from these plates onto MVPM and V8CB failed to show conclusively that Pythium spp. had colonized the baits. Sexual structures (oogonia, antheridia) were not observed, nor were hyphal swellings typical of Pythium sp. Group G when subsequent transfers were made onto V8CB. Most of the fungi growing on MVPM were identified as Mortierella spp. Isolation and identification of baittrapped fungi were also attempted by placing 5-mm disks of agar containing hyphal tips into dishes of Petri salt solution. Fungal growth from these disks, observed 24–48 hr later, contained no structures (sporangia, oogonia) that would allow positive identification of Pythium spp. Pythium spp. were also not isolated from plated fungus gnats. Plates mainly contained species of Penicillium, Rhizopus, and Aspergillus. Open plates of MVPM placed on sawdust bags failed to show any evidence of colonization by Pythium spp.

## DISCUSSION

The experimental work described demonstrates that at least four Pythium spp. isolated from the commercial sawdust culture system are able to cause reductions in cucumber growth with or without rotting symptoms similar to those observed in commercial greenhouses. Of the four species studied, P. aphanidermatum appeared to be the most aggressive under the given experimental conditions. Whether the difference in pathogenicity of P. aphanidermatum and P. irregulare in experiment 3 compared with experiments 1 and 2 reflects a difference in the diseaseproducing potentials of mixed oosporemycelium inoculum, a difference in the disease suppressiveness of the unsterilized vs. the sterilized growth medium, or some other factor is unknown.

The observed reduction in plant growth in the absence of symptoms is noteworthy. Recent work (17) indicates that such "subclinical" infection by *Pythium* may be relatively common. Yield reductions as high as 54% were reported in hydroponically grown lettuce not showing visible symptoms (17).

The sampling survey to identify potential sources of *Pythium* inoculum showed that these fungi occur commonly in the mixes used for cucumber seedling propagation in commercial greenhouses. In a similar survey of randomly selected

horticultural peat products, Kim et al (9) isolated *Pythium* spp. (including *P. aphanidermatum* and *P. irregulare*) from 15 of 52 mixes sampled. The occurrence of *Pythium* spp. in water systems in commercial greenhouses or in fungus gnats occurring in these greenhouses was not confirmed; the number of sites sampled was low, however.

A combination of factors present during propagation, such as reduced levels of antagonism in the mixes (4), juvenile host tissues, abundant supplies of inorganic nutrients (12), and high moisture and temperatures, provides an ideal environment for the proliferation of Pythium. Despite these factors, disease is rarely seen on immature plants in the local industry. The sporadic occurrence of relatively low levels of Pythium propagules in the early stages of propagation and/or the possibility that the Pythium spp. present are not highly pathogenic may at least partially explain why growers rarely see damping-off. Throughout these early stages, however, Pythium spp. may, under suitable conditions, colonize root cortical tissues as "minor pathogens" (14), without causing any apparent damage. Under conditions of host stress and physiological change, such as the initiation of fruit production, plant defenses may weaken to such an extent as to allow the rapid spread of these minor infections to produce overt crown rot symptoms and wilting. The contention that weak or "minor" pathogens also cause crown rot is indirectly supported by the results of the disease epidemiology survey.

Greenhouse environments are generally conducive to the growth of Pythium spp. High soil temperatures and abundant moisture, identified as the two most important factors for infection by Pythium (6), are common in greenhouses. P. aphanidermatum has caused extensive losses in hydroponically grown tomatoes (7,19), cucumbers (8,13), and spinach (3)and also in soilless and rock wool culture of cucumbers (16,18), poinsettia (5), and bedding plants (20). P. irregulare and P. coloratum were isolated from nutrient film technique (NFT) culture in the United Kingdom, where these species had previously caused disease on a number of crops (13). P. aphanidermatum also caused root and basal stem disease on tomatoes and cucumbers grown on NFT in the Federal Republic of Germany (15).

Crown rot in British Columbia most closely resembles the disease described in Saudi Arabia on cucumbers grown in sand culture (18) and in Florida where cucumbers are produced in polyethylene bags of commercial potting mix (16). Disease in these cases appears to be caused primarily by *P. aphanidermatum* and is similar to crown rot in that it is prevalent on mature plants and produces

rot and wilting symptoms similar to those occurring in the British Columbia commercial greenhouse industry.

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