# Etiology and Control of Seed Decay and Preemergence Damping-Off of Chickpea by Pythium ultimum

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#### ABSTRACT

Kaiser, W. J., and Hannan, R. M. 1983. Etiology and control of seed decay and preemergence damping-off of chickpea by Pythium ultimum. Plant Disease 67:77-81.

Emergence of several cream-colored chickpea (Cicer arietinum) Plant Inventory (PI) accessions in field trials at various locations in the Palouse region of eastern Washington was erratic and/or reduced because of a seed rot and preemergence damping-off disease caused by Pythium ultimum. Fusarium solani and P. ultimum were isolated most frequently from decayed seeds, while fungi isolated occasionally were Mucor sp. and other Pythium spp. Seedling emergence of cream-colored PI 458870 was reduced from 86% in the uninfested control to 58, 24, and 2% in soil artificially infested with 5, 50, and 500 propagules per gram of soil (ppg) of P. ultimum, respectively. In the same test, brown- and black-seeded PI accessions 273879 and 458869, respectively, were highly resistant to seed rot and preemergence damping-off compared with PI 458870. Untreated seeds of PI 458870 failed to emerge in natural field soil at 10, 15, 20, or 25 C, whereas emergence of metalaxyl-treated seeds was >80% at these temperatures. Seedling age greatly influenced susceptibility to seed decay and damping-off. When pregerminated in moist vermiculite, transplanted seedlings developed resistance to P. ultimum in naturally infested soil after 48-96 hr. In soils collected in the top 16 cm of 12 cropped and fallowed fields in eastern Washington and northern Idaho, populations of P. ultimum ranged from 56 to 1400 ppg and emergence of untreated seeds was reduced by >90%. Soil fumigation with methyl bromide and treatment of seeds with different fungicides significantly increased emergence and yields of PI 458870 in field trials at Central Ferry, WA. Metalaxyl (Ridomil) and captan were the most effective seed-treatment fungicides for preventing seed rot and preemergence damping-off and in increasing yields. In soil artificially infested with P. ultimum, seeds treated with metalaxyl, captan, ethazol, ethazol + thiophanate, thiram, or fenaminosulf significantly increased emergence over the untreated control. Fenaminosulf (Dexon) delayed germination and stunted seedlings. Metalaxyl-treated seeds stored at 4 C for up to 464 days before planting were protected when planted in field soil heavily infested with P. ultimum.

Additional key words: fungicide seed treatment, soilborne

The U.S. Department of Agriculture Plant Inventory (PI) germ plasm collection for chickpea (Cicer arietinum L.), also called garbanzo or gram, has been located at the Regional Plant Introduction Station, Pullman, WA, since 1979. Chickpea PI accessions must be increased periodically in the field or greenhouse to replenish seed supplies. Diseases affecting seed increase cause great concern because important Cicer

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Accepted for publication 26 May 1982.

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germ plasm, if lost, may be impossible to

In the United States, chickpeas are grown commercially on more than 5,000 ha (12,500 acres), primarily in the central coastal areas of California (3); however, the United States was still importing seeds of large, cream-colored chickpeas valued at more than \$9 million in 1980 (16). Chickpeas are a potential crop for the dryland areas of eastern Washington and northern Idaho (2,8,9). The climate and soils in this region appear to be ideally suited to the cultivation of chickpeas. Seed yields of >3,400 kg/ha (3,000 lb per acre) were obtained with different black, brown, and/or creamcolored lines in field trials at Central Ferry and Pullman, WA (unpublished), and Moscow, ID (2).

During the 1979 and 1980 growing seasons, emergence of untreated seeds of many large (>25 g/100 seeds), creamcolored PI lines at Central Ferry and Pullman was erratic or poor when compared with untreated seeds of brown or black lines. The primary soilborne pathogen responsible for this damage in the Palouse region of eastern Washington and northern Idaho appeared to be

Pythium ultimum Trow (9). Information on P. ultimum as a pathogen of chickpea is scarce but it has been reported to cause preemergence and postemergence damping-off and root rot of chickpeas in California (17) and Iran (10).

The objectives of this study were to examine the etiology of the seed rot and preemergence damping-off disease of cream-colored chickpeas in the Palouse region, particularly the role of P. ultimum; to study the influence of inoculum density of P. ultimum in naturally and artificially infested soils and of soil temperature on disease incidence; and to evaluate the effectiveness of different seed-treatment fungicides in controlling the disease under field and greenhouse conditions. A preliminary report on part of this work has been published (9).

## MATERIALS AND METHODS

Isolation from chickpea seeds. Unless stated otherwise, all field and greenhouse experiments were carried out with a large (39 g/100 seeds), cream-colored chickpea line (PI 458870, USA). At intervals up to 14 days after planting, ungerminated seeds were removed from soil in the field or greenhouse, placed in running tap water for 15-30 min, and surface sterilized in 0.25% NaOCl for 5 min. Seed pieces were plated on 2% water agar (WA), fresh potato-dextrose agar (PDA), or P<sub>10</sub>VP-selective medium (15). Plates were incubated at 24-26 C. Fungi isolated from chickpea seeds were stored on Difco cornmeal agar (CMA) or PDA at 25 or 4 C.

Isolation of P. ultimum from soil. A Pythium-selective medium (11,12) was used to estimate the inoculum density of Pythium spp., including P. ultimum, in naturally infested field and artificially infested greenhouse soils. One milliliter of each soil dilution suspended in 0.2% WA was spread on the surface of the medium. Plates were incubated at 20 C in the dark for 48 hr.

Greenhouse experiments. Soil from Central Ferry, WA, naturally infested with P. ultimum, was used in most greenhouse experiments. This soil was classified as a Spofford silt loam with the following characteristics: pH 6.6; conductivity, 0.6 mmhos/cm; 27% sand; 53.8% coarse silt; 2.8% fine silt; 16.4% clay; and 1.6% organic matter. The soil was amended with peat moss (10-14%) v/v) to improve friability and reduce soil compaction. Experiments were performed in a greenhouse at  $20 \pm 5$  C or in growth chambers at 10, 15, 20, and 25 C with a 16-hr photoperiod (11,750 lux). Plastic pots, 15 cm in diameter and 15 cm deep, were used in all tests. Ten fungicide-treated or untreated seeds were sown in each pot to a depth of 1.5-2.0 cm. Pots were watered to saturation after planting and thereafter every 2-4 days when the soil moisture reached -0.3 bars. Soil moisture was monitored with a tensiometer (Irrometer Co., Inc., Riverside, CA 92506).

Soils were collected from the top 16 cm of fields throughout the Palouse region. Each sample was thoroughly mixed and planted to captan-treated (3 g a.i./kg seed) or untreated seeds. The population of *Pythium* spp. was determined for each soil using Mircetich's selective medium (11). Stand counts were made 10–15 days after planting.

Pathogenicity tests. Pathogenicity tests were carried out in the greenhouse with fungi isolated from rotted seeds or from seedlings that had damped-off before emergence. The method used to test pathogenicity was one of the following:

1) Fungal isolates were grown on PDA or CMA plates for 5-10 days at 23-25 C. Cultures were cut into small pieces (~1.0 cm²) and one plate was incorporated into the top 4-6 cm of greenhouse potting medium (55% peat moss, 35% pumice, 10% sand) contained in a 15-cm-diameter plastic pot. Uninfested controls containing sterile PDA or CMA were included in all tests. Untreated chickpea seeds were planted in all pots.

2) Fungal isolates were grown in a

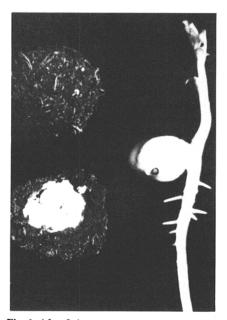


Fig. 1. After 9 days, untreated chickpea seeds (left) planted in field soil from Central Ferry, WA, were rotted by *Pythium ultimum*, while seed treated with metalaxyl (Ridomil) (right) has germinated. Soil usually adheres to decayed seeds, giving them the appearance of a brown ball

sterilized mixture of sand-cornmeal (97% sand and 3% cornmeal, w/w, with 20% water, w/v) for 14 days. Inoculum of each isolate was incorporated into greenhouse potting soil that was then air-dried for 5-7 days before being placed in pots. Uninfested control pots received the sterile sand-cornmeal mixture without fungus. Untreated chickpea seeds were planted in all pots.

Pathogenicity of each fungal isolate was based on percentage emergence of untreated seeds in infested soil and on necrosis of roots of emerged plants. Pathogenicity of fungi on roots was based on a root rot index (RRI) of 0-4 where 0 = healthy roots, 1 = 1-10%, 2 = 11-25%, 3 = 26-50%, and 4 = >50% of the root system necrotic.

Effects of temperature and seedling age on damping-off. The effects of soil temperature on Pythium seed rot and preemergence damping-off were tested by planting untreated and metalaxyl-treated (0.3 g a.i./kg) seeds in field soil that was incubated at 10, 15, 20, or 25 C in growth chambers. Stand counts were taken daily for 20 days and isolations were made at periodic intervals from selected seeds that failed to emerge. The effect of seedling age on preemergence and postemergence damping-off also was tested in naturally infested field soil in the greenhouse. Untreated seeds were germinated in moist vermiculite at 15 or 25 C. After rinsing in tap water, ungerminated and germinating seeds of different ages were planted in field soil in the greenhouse (15-25 C).

Seed treatment. Fungicides used to treat chickpea seeds in the different greenhouse and field experiments were benomyl (Benlate 50W), thiram (Thiram 75W), thiram (60W) + dieldrin (12.75W)(Delsan A-D), E. I. du Pont de Nemours & Co., Wilmington, DE 19898; metalaxyl (Ridomil 25W or CGA-48988), Ciba-Geigy Corp., Greensboro, NC 27409; captan (Orthocide 50W), Chevron Chemical Co., Ortho Division, Richmond, CA 94804; ethazol (Truban 30W), ethazol (15W) + thiophanate methyl (25W) (Banrot 40W), Mallinckrodt, Inc., St. Louis, MO 63147; fenaminosulf (Lesan 35W), Mobay Chemical Corp., Kansas City, MO 64120; quintozene (Terraclor 75W), Olin Corp., Agricultural Division, Little Rock, AR 77203.

The effect of various fungicide seed treatments on emergence was tested in the greenhouse with naturally infested field soil and greenhouse soil artificially infested with *P. ultimum*. Fungicide seed slurries were prepared by mixing each fungicide in 3-4 ml of water in 500-ml Erlenmeyer flasks. One hundred grams of seed was added to flasks, agitated for 2 min, and air-dried.

In greenhouse trials with naturally infested Spofford silt loam soil, seeds were treated with metalaxyl applied at various dosage rates to determine its phytotoxic effect on emergence and

growth of seedlings. Treated seeds were stored at 4 C for various intervals to determine the effectiveness of metalaxyl for disease control.

Field experiments. Field trials were conducted in Spofford silt loam soil at Central Ferry. Fungicide-treated and untreated seeds were planted 2.5–3.0 cm deep with a V-belt planter in fumigated and unfumigated soil on 17 April 1980. Seeds were planted in rows 6.1 m long with a spacing of 1.5 m between rows. Each row constituted a replicate and treatments were replicated four times, with 50 seeds per replicate in a randomized complete block design.

For fumigation, tarped plots were treated with a mixture of methyl bromide (98%) and chloropicrin (2%) at a rate of 55 g/m<sup>2</sup>. A row of untreated seeds was planted down the middle of each fumigated plot, and emergence counts were made after 11, 18, and 26 days. Seed yields were determined for each plot.

### RESULTS

Isolation from seeds. Isolations were made on WA and P<sub>10</sub>VP medium from 85 pieces of tissue taken from 17 rotted seeds that failed to emerge after 9 days in Spofford silt loam soil in pots incubated in the greenhouse. Soil frequently adhered to rotted seed, giving it the appearance of a small brown ball (Fig. 1). The cotyledons were decayed and at times discolored. Fusarium solani (Mart.) Appel & Wr. was isolated from >94% of the seed pieces on both media; P. ultimum was isolated from 41.2 and 77.6% of the seed pieces on WA and P<sub>10</sub>VP medium, respectively. Mucor sp. and another Pythium sp. were isolated from <10% of the seed pieces.

Isolations also were made from 300 seeds plated at 12-96 hr after planting in unsterile Spofford silt loam soil that was incubated at 10, 15, or 25 C in the greenhouse. P. ultimum accounted for >95% of the fungi isolated from 2,400 seed pieces (each seed cut into eight pieces) (Table 1). P. ultimum infected seeds most rapidly at 25 C. Within 12 hr at 25 C, the fungus was isolated from 7% of the seed pieces (Table 1). At 72 hr after planting, P. ultimum was isolated from 66, 91, and 100% of the tissues from seeds that had been incubated in soil at 10, 15, or 25 C, respectively. At that time most of the seeds were turning soft and the seed coats were beginning to slough off.

Pathogenicity tests. Pythium ultimum was the only fungus pathogenic to chickpea that was isolated consistently from rotted seeds in controlled-inoculation studies. Emergence in soil artificially infested with P. ultimum was reduced by 92% over the uninfested control. The fungus also was pathogenic to the roots of chickpea seedlings (RRI 3) that emerged in infested soil. Seedlings in P. ultimum-infested soil were stunted and the larger roots were necrotic, discolored, and

devoid of feeder rootlets. Frequently >40% of the stunted plants died before flowering. Emergence in soil infested by *F. solani, Pythium* sp. or *Mucor* sp. was not statistically different from that in uninfested soil; moreover, these fungi were not pathogenic to the roots of inoculated chickpeas (RRI = 0).

The pathogenicity of *P. ultimum* to chickpea PI accessions with cream, brown, or black seed coats varied greatly in greenhouse inoculation trials (Table 2). Emergence of cream-colored PI 458870 was reduced significantly compared with the uninfested control at inoculum levels of 5, 50, and 500 propagules per gram of soil (ppg) (Table 2). As inoculum levels of *P. ultimum* increased from 0 (uninfested) to 500 ppg of soil, emergence decreased from 86 to 2%. The brown and black PI lines used in this inoculation study were highly resistant to *P. ultimum*, even at the highest inoculum level (Table 2).

Temperature and seedling age. The emergence of untreated and metalaxyltreated seeds was tested in greenhouse studies using naturally infested field soil with a *P. ultimum* population of 288 ppg at 10, 15, 20, or 25 C. Untreated seeds failed to emerge at the temperatures tested. After 10–15 days, *P. ultimum* was isolated from >60% of untreated seeds at the different temperatures. Emergence of metalaxyl-treated seeds after 15 days at 10, 15, 20, or 25 C was 82, 84, 86, and 86%, respectively.

Table 1. Time required for *Pythium ultimum* to infect chickpea seeds in naturally infested field soil<sup>a</sup> at specified temperatures<sup>b</sup>

Temperature (C)	Seed removed from soil after (hr)	Isolation from seed pieces <sup>c</sup>	Infection (%)
10	24	0/160	0
	36	3/160	1.9
	48	25/160	15.9
	72	105/160	65.6
	96	149/160	93.1
15	24	1/160	0.6
	36	9/160	5.6
	48	69/160	43.1
	72	146/160	91.3
	96	160/160	100
25	12	11/160	6.9
	24	74/160	46.3
	36	202/240	84.2
	48	159/160	99.4
	72	80/80	100

<sup>&</sup>lt;sup>a</sup> Spofford silt loam soil from Central Ferry, WA. Natural population of *P. ultimum* of 860 propagules per gram of soil.

Transplanted seedlings (radicle growth  $\geq$ 0.3 cm) were less susceptible to preemergence damping-off than ungerminated seeds (no visible radicle growth) (Table 3). Seeds planted in moist vermiculite at 15 or 25 C and held for 0-72 or 0-24 hr, respectively, were highly susceptible to preemergence damping-off caused by P. ultimum when transplanted to naturally infested field soil. Dry seeds (0 hr), or those without visible radicle growth when planted in Central Ferry soil, failed to emerge. The incidence of preemergence damping-off was significantly less as radicle length increased and shoot growth began before planting in infested soil. The incidence of postemergence damping-off was low (<20%) in seedlings that had just initiated shoot growth when planted.

Emergence of seeds in Palouse soils. Untreated seeds failed to emerge in 10 of 12 silt loam soils collected from different areas with different cropping histories in the Palouse region (Table 4). *P. ultimum* was the only pathogen consistently isolated from rotted seeds in these soils. Emergence of captan-treated seeds was significantly higher than with untreated seeds in all soils and ranged from 56 to 92% (Fig. 2). The population of *P. ultimum* in these soils ranged from 56 to 1400 ppg.

Field experiments. Soil fumigation and several seed-treatment fungicides significantly reduced preemergence damping-off and seed rot and increased yields (Table 5). Fumigation of soil with methyl bromide before planting resulted in the most emergence and highest seed yields.

**Table 2.** Susceptibility of three chickpea lines to seed rot and preemergence damping-off in soil artificially infested with *Pythium ultimum* at three inoculum levels<sup>x</sup>

Inoculum level (propagules per gram of soil)	Chickpea line	Seed color	Emergence <sup>3</sup> (%)
500	PI 273879, Ethiopia	Brown	92 a²
	PI 458869, Israel	Black	86 ab
	PI 458870, USA	Cream	2 e
50	PI 273879	Brown	88 ab
	PI 458869	Black	84 b
	PI 458870	Cream	24 d
5	PI 273879	Brown	90 ab
	PI 458869	Black	86 ab
	PI 458870	Cream	58 c
0 (uninfested)	PI 273879	Brown	88 ab
	PI 458869	Black	84 b
	PI 458870	Cream	86 ab

<sup>\*</sup>Soil was infested with a cornmeal-sand medium of *P. ultimum*, then serially diluted with sterile soil to give the inoculum levels indicated. Uninfested soil received sterile cornmeal-sand. Mircetich's (11) selective medium (P<sub>5</sub>V<sub>300</sub>PCNB<sub>100</sub>RB<sub>10</sub>) was used to estimate the inoculum density of *P. ultimum*.

**Table 3.** Susceptibility of cream-colored chickpea seeds germinated in moist vermiculite at 15 or 25 C for different time intervals to preemergence and postemergence damping-off caused by *Pythium ultimum* when transplanted to naturally infested field soil<sup>a</sup>

	Preplant germination time (hr)	Mean length of				
Temperature (C)		Radicle (cm)	Shoot (cm)	Damping-off (%)		
				Preemergence	Postemergence	
15	0	О <sub>р</sub>	0	100	0	
	12	0	0	100	0	
	24	0	0	100	0	
	48	0.3	0	78	0	
	72	1.4	0	52	0	
	96	3.0	0.2	0	12	
25	0	0	0	100	0	
	12	0	0	100	0	
	24	0.3	0	80	0	
	48	2.0	0.2	0	20	
	72	5.0	0.8	0	0	
	96	6.2	1.8	0	0	

<sup>&</sup>lt;sup>a</sup>Seeds of cream-colored chickpea PI 458870, USA, were transplanted to a Spofford silt loam soil from Central Ferry, WA, where the population of *P. ultimum* was 860 propagules per gram of soil. <sup>b</sup> Seeds or seedlings were planted (10/pot) in 15 cm pots with five pots per treatment. Pots were incubated in the greenhouse at  $20 \pm 5$  C and disease readings were taken after 10 and 15 days.

bAfter removal from soil, seeds of creamcolored chickpea PI 458870, USA, were rinsed in tap water and surface-sterilized in 0.25% NaOCI for 5 min. Seed coats were removed before each seed was cut into eight pieces and plated on 2% water agar, one seed per I0-cm plate.

<sup>&</sup>lt;sup>c</sup>Number of pieces infected with *P. ultimum* over total number cultured.

<sup>&</sup>lt;sup>y</sup>Untreated seeds were planted (10/pot) in 15-cm-diameter plastic pots with five pots per treatment per PI line. Emergence counts were taken 11 and 16 days after planting.

<sup>&</sup>lt;sup>2</sup>Numbers within the column followed by the same letter do not differ significantly (P = 0.05) according to Duncan's multiple range test.

Preemergence damping-off and seed rot were significantly less and yields significantly higher than with the untreated control if seeds were treated with metalaxyl, captan, ethazol, thiram + dieldrin, fenaminosulf, or ethazol + thiophanate. Untreated seeds failed to emerge. Emergence and yields of seeds treated with benomyl and quintozene (PCNB) did not differ significantly from those in the untreated control. Germination of seeds treated with fenaminosulf at 3.0 g a.i./kg seed was delayed several days and plant growth was initially retarded.

Greenhouse fungicide trials. In the greenhouse, seeds treated with the same fungicides used in the field study (Table 5) were planted in soil artificially infested with *P. ultimum*. Preemergence damping-off and seed rot caused by *P. ultimum* 

were significantly reduced by all treatments tested, except benomyl and quintozene. Emergence of seeds treated with the different fungicides was similar to that observed in the field trial. These chemicals also protected seedlings against postemergence damping-off and root rot caused by P. ultimum throughout the 31/2 wk of experimentation, even at a very high inoculum level (28,600 ppg). Metalaxyl applied to the seeds resulted in the most emergence (86%), followed by captan (80%). Untreated seeds failed to emerge, while emergence of those treated with fenaminosulf was delayed 2-5 days and >75% of the seedlings were stunted with deformed foliage.

Emergence of seeds treated with metalaxyl at dosage rates of 0.075, 0.15, 0.30, or 0.60 g a.i./kg seed was 91, 92, 86, and 91%, respectively, whereas that of

Table 4. Emergence of untreated and captan-treated chickpea seeds in silt loam soils from 12 fields naturally infested with *Pythium ultimum* in the Palouse region of eastern Washington and northern Idaho<sup>a</sup>

	Crop history				No. P. ultimum
	Previous 1979	Present 1980 <sup>b</sup>	Emergence (%)		propagules
Location			Treated	Untreated	per gram of soil
Central Ferry, WA	Barley	Fallow	84	0	722
Colfax, WA	Fallow	Wheat	90	0	89
Garfield, WA	Wheat	Lentil	77	0	293
Genesee, ID	Wheat	Fallow	62	0	260
Lapwai, ID	Wheat	Chickpea	72	0	144
Moscow, ID	Wheat	Wheat	72	10	455
Pullman, WA	Pea	Lentil	56	0	422
Pullman, WA	Wheat	Pea	90	10	145
Potlatch, ID	Wheat	Wheat	80	0	1400
St. John, WA	Wheat	Lentil	92	Õ	178
Spangle, WA	Wheat	Fallow	82	Õ	56
Wilbur, WA	Wheat	Fallow	76	0	220

<sup>&</sup>lt;sup>a</sup> A total of 40-50 untreated or captan-treated seeds of cream-colored P1458870, USA, were placed in each soil sample in 15-cm-diameter plastic pots (four or five replicates with 10 seeds per pot). <sup>b</sup> At each location, soil samples were collected in the top 16 cm from several sites and mixed thoroughly before placing in pots.

<sup>c</sup>Soil dilutions were plated on the selective medium (P<sub>5</sub>V<sub>300</sub>PCNB<sub>100</sub>RB<sub>10</sub>) of Mircetich (11).



Fig. 2. Emergence of untreated (bottom row) and captan-treated (top row) chickpea seeds in field soil naturally infested with *Pythium ultimum*. The soil was collected from a field near Pullman, WA, that had been cropped to barley the previous season.

captan, thiram, and the untreated control was 91, 78, and 0%, respectively. There were no significant differences in emergence between the different metalaxyl treatments and captan. Metalaxyl at 0.075-0.6 g a.i./kg seed had no apparent phytotoxic effects on emergence or growth of chickpeas.

There were no significant differences in emergence of metalaxyl-treated seeds stored for 0, 14, 31, 209, or 464 days at 4 C before planting. Emergence of the metalaxyl-treated seeds ranged from 74 to 78%. Germination of treated seeds stored 14–464 days was not impaired compared with treated or untreated seeds planted immediately after treatment.

### DISCUSSION

As few as 5 ppg of P. ultimum reduced emergence of cream-colored chickpeas by >40%. All of the Palouse soils sampled had P. ultimum populations of >55 ppg and emergence of untreated seeds in these soils was less than 10%. In naturally infested soils, seed treatment with metalaxyl increased emergence over untreated seeds by 94%. The control afforded by metalaxyl, a water-soluble, systemic fungicide toxic only to Oomycetes (4,5) in field soil, provides convincing evidence of the importance of P. ultimum in the etiology of the seed decay and preemergence damping-off disease affecting chickpeas in the Palouse.

During 1980 and 1981, yields of captan-treated, cream-colored PI accessions were significantly increased by seeding in early spring at Pullman (unpublished). At that time of year chickpeas are planted when temperatures in the top 6 cm of soil are cold (<15 C). Soil temperature is often one of the most important factors influencing development of some diseases incited by P. ultimum (7); however, seed rot and preemergence damping-off of chickpeas in greenhouse trials with naturally infested field soil were not affected by soil temperature in the range tested. Seeds of untreated cream-colored lines failed to emerge at temperatures of 10-25 C. Planting in cold soils also slows the germination process, exposing susceptible tissues (cotyledons, radicles, and shoots) to infection by P. ultimum for a longer period. Most Pythium spp., including P. ultimum, are important pathogens of young succulent tissues (7). The age of chickpea seedlings had a profound effect on their susceptibility to P. ultimum in naturally infested soil in greenhouse trials. Ungerminated seeds or those just beginning to germinate (12-24 hr) were extremely susceptible to the pathogen. Seedlings, however, developed resistance to seed rot and preemergence dampingoff within 48-96 hr. Halpin and Hanson (6) observed a similar phenomenon with seedlings of alfalfa (Medicago sativa L.), red clover (Trifolium pratense L.), sweet clover (Melilotus sp.), and Ladino white

Table 5. Effect of soil fumigation or seed treatment fungicides on emergence and yield of chickpea in field trials at Central Ferry, WA

Treatment	Rate of application (grams active ingredient per kilogram of seed)	Emergence <sup>w</sup> (%)	Yield <sup>x</sup> (g)
Soil fumigation <sup>y</sup>	•••	95.0 a <sup>ż</sup>	1830 a
Captan	3.0	87.0 ab	1623 b
Metalaxyl	0.3	94.0 a	1580 bc
Ethazol	3.0	82.6 b	1468 bc
Thiram + dieldrin	3.0	71.6 b	1391 cd
Ethazol +			
thiophanate methyl	3.0	37.0 e	1254 d
Fenaminosulf	3.0	53.0 d	953 e
Benomyl	3.0	7.5 f	78 f
Quintozene	3.0	1.0 f	47 f
None (control)	•••	0 f	0 f

<sup>\*</sup>A total of 200 chickpea seeds were planted per treatment (four replicates with 50 seeds each). The natural population of *Pythium ultimum* was 335 propagules per gram of soil. Emergence counts were taken 11, 18, and 26 days after planting.

<sup>x</sup> Average yield per row from four, one-row plots (each row 5.3 m long).

clover (*T. repens* L.) to five species of *Pythium*. Seedlings of the four legume species were susceptible to the five *Pythium* spp. at the time of seeding, but were resistant when inoculated 3 or more days after seeding.

Metalaxyl seed treatment effectively increased emergence and yield of chickpeas in the Palouse region on land rotated with wheat and barley. This appears to be the first reported use of metalaxyl to control a serious disease of chickpea caused by P. ultimum. Seed treatment with metalaxyl offers several advantages over protective fungicides like captan and thiram for controlling Pythium seed rot and damping-off of chickpea in naturally infested Palouse soils. Metalaxyl-treated chickpea seeds are protected against infection by P. ultimum in heavily infested soils at low dosage rates (eg, 0.075 g a.i./kg seed). The protective effects of metalaxyl on chickpea seeds are long lasting. Chickpea seeds, initially treated with metalaxyl and stored at 4 C for more than 1 yr, were protected from infection by P. ultimum when planted in heavily infested soil. The fungicide was not phytotoxic to any of the chickpea accessions tested at dosage rates up to 0.6 g a.i./kg seed.

Chickpeas are commonly divided into two groups (kabuli and desi) based on color, size, and shape of the seed, in addition to other plant characteristics (1; L. J. G. van der Maesen, personal communication). Kabuli lines generally have large (>25 g/100 seeds), rounded, light-colored seeds with thin seed coats that adhere loosely to the cotyledons. Desi types usually have small, irregularly shaped, dark-colored seeds with thick seed coats that adhere tightly to the cotyledons. Early in our studies, we observed that poor emergence of chickpeas in field trials at Pullman and Central Ferry appeared to be associated with seed color. Field and greenhouse inoculation studies with untreated seeds demonstrated that kabuli lines were highly susceptible to Pythium seed rot and preemergence damping-off, whereas desi lines were resistant to the disease (9; unpublished). Resistance in pea to P. ultimum also appears to be associated with seed coat color (13,14). Treatment of desi seeds with fungicides effective against Pythium seed rot and dampingoff, eg, metalaxyl or captan, had no effect on increasing emergence over untreated seeds in soils heavily infested with P. ultimum. Chickpea cultivars grown commercially in the United States are the large-seeded kabuli type. As yet no kabuli lines with high levels of resistance to P. ultimum have been found in preliminary screening of the Cicer PI collection in the greenhouse and field. Incorporating resistance to *Pythium* in large-seeded kabuli lines will be an important objective of a breeding program in the Palouse region.

### **ACKNOWLEDGMENTS**

We thank W. L. Anliker, Ciba-Geigy Corp., Vancouver, WA 98664, for providing samples of metalaxyl (Ridomil); Sheau-fang Hwang, Department of Plant Pathology, Washington State University, Pullman 99164, and the Commonwealth Mycological Institute, Kew, Surrey, England, for identifying cultures of Fusarium and Pythium, respectively; Kathleen Rigert for her competent technical assistance, and V. E. Wilson for his help, advice, and encouragement during the course of this study.

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<sup>&</sup>lt;sup>y</sup> Soil (Spofford silt loam) was fumigated with a mixture of methyl bromide (98%) + chloropicrin (2%) at the rate of 55 g/m<sup>2</sup>. Untreated seeds of cream-colored chickpea PI 458870, USA, were planted in the fumigated soil.

<sup>&</sup>lt;sup>2</sup> Numbers in each column followed by the same letter do not differ significantly (P = 0.05) according to Duncan's multiple range test.