Incidence of Seedborne *Ascochyta lentis* in Lentil Germ Plasm

Walter J. Kaiser and Richard M. Hannan

Research plant pathologist and biological technician, Western Regional Plant Introduction Station, U.S. Department of Agriculture, Agricultural Research Service, Washington State University, Pullman 99164.

Mention of a trademark or proprietary product does not constitute a guarantee or warranty of the product by the U.S. Department of Agriculture and does not imply approval to the exclusion of other products that may also be suitable.

Accepted for publication 14 October 1985.

ABSTRACT

Kaiser, W. J., and Hannan, R. M. 1986. Incidence of seedborne *Ascochyta lentis* in lentil germ plasm. Phytopathology 76:355-360.

A foliar blight of lentil (*Lens culinaris*) was observed in June 1981 in several plant introductions in cold-tolerance trials planted in the fall of 1980 at Pullman and Central Ferry, WA. *Ascochyta lentis* was the predominant fungus isolated from discolored, necrotic lesions on the foliage and seeds of diseased lentil. Isolates of *A. lentis* were pathogenic to the foliage of lentil, but not to the foliage of chickpea (*Cicer arietinum*) or pea (*Pisum sativum*). The fungus was isolated from 1.5–3.5% of the original introduced seed from three of five lentil PI accessions included in the 1980–1981 cold-tolerance trials. Infection by *A. lentis* of seeds harvested from these trials (increase seeds) ranged from 0.5 to 68.5% and from 10 to 42.5% at Pullman and Central Ferry, respectively. Many seeds from heavily infected accessions were shriveled and discolored, and seed quality was adversely affected. Also, seed size was significantly correlated to the level of seedborne infection. A total of 17,060 original seeds from 284 accessions from 30 countries were screened for seedborne *A. lentis*. The fungus was isolated from 2.0% of the seeds which represented 16% of the accessions and 16 countries. Most severe infections were found on original seeds from Australia, India, Italy, Spain, and Turkey. Other fungi pathogenic to lentil that were also isolated, but less frequently than *A. lentis*, included: *Botrytis cinerea, Fusarium avenaceum, Macrophomina phaseolina, Phoma medicaginis* var. *pinodella*, and *Rhizoctonia solani*. Incidence of seedborne *A. lentis* from original infected seeds (5.0–61.7%) in 20 exotic PI accessions to increase seeds grown in typical spring plantings at Pullman was 0–2.5%. *Ascochyta lentis* remained viable in original seeds of several accessions stored for more than 30 yr. The fungus survived over 3 yr in naturally infected lentil pods and seeds at 4–6 C or in a shelter outdoors, and for 1.5 yr on the soil surface, but it lost viability within 29 wk at a soil depth of 16 cm.

The germ plasm collection of lentil (*Lens culinaris* Medik.) is maintained by the USDA at the Western Regional Plant Introduction Station, Pullman, WA. It currently contains 1,973 plant inventory (PI) accessions, most of which are of foreign origin. Periodically, lentil germ plasm lines must be increased in the greenhouse or field to replenish seed supplies. Diseases, both foreign and domestic, that affect seed increase and seed quality are of utmost concern because it may be impossible to replace potentially valuable lentil germ plasm once it is lost (9).

Two potentially important seedborne pathogens have recently been isolated from the USDA lentil PI collection at Pullman. In 1982, Hampton (8) isolated a seedborne virus, designated the lentil strain of pea seedborne mosaic virus, from 38 of 470 lentil PI accessions. In the same year, Kaiser and Hannan (10) reported isolating *Ascochyta lentis* Vassiljevsky from the original seeds of several lentil PI accessions.

In the Palouse region of eastern Washington and northern Idaho, lentil is planted in the spring and harvested in late summer. Lentil genotypes that tolerate severe winter conditions have been found in Turkey (5). Planting such cold-tolerant lentils in the fall produced greater yields than those of the spring-seeded types and the crop can be harvested earlier in the summer. Two cold-tolerance trials were established in the fall of 1980 at Pullman and Central Ferry, WA, with seeds of five cold-tolerant lentil PI accessions received from O. Tosun, Ankara University, Ankara, Turkey. Blight of lentils caused by *A. lentis* was first observed by the authors in June 1981, in both cold-tolerance trials (10).

The objectives of this study were to examine the symptoms and etiology of Ascochyta blight in naturally infected cold-tolerant lentils in eastern Washington, to determine the incidence of seedborne *A. lentis* in original and increase seeds of lentil PI accessions, and to study the survival of *A. lentis* on naturally infected lentil debris under field conditions.

The publication costs of this article were defrayed in part by page charge payment. This article must therefore be hereby marked "*advertisement*" in accordance with 18 U.S.C. § 1734 solely to indicate this fact.

MATERIALS AND METHODS

Isolation of *A. lentis* from diseased lentils. In June 1981, over 150 isolations were made from necrotic lesions on foliage and seeds of lentil plants in cold-tolerance trials at Pullman and Central Ferry, WA. Tissues were surface sterilized in 0.25% NaOCl for 5 min, plated on 2% water agar (WA), and incubated at 20–24 C under fluorescent lights (12-hr photoperiod, 4,300 lux). As fungi emerged, hyphal tips were transferred to natural potato-dextrose agar (PDA) slants and stored at 4 C in the dark. Fungi were identified to genus or species by the authors or by mycologists at the Commonwealth Mycological Institute (CMI), Kew, Surrey, U.K.

Pathogenicity tests. Pathogenicity tests were conducted in the greenhouse with two to five isolates of each fungus. Fungi included in the pathogenicity tests were cultured on PDA, WA, or carnation-leaf agar (for *Fusarium* spp. [18]) under fluorescent lights (12-hr photoperiod) for 7–14 days. Conidia were collected by flooding petri dishes with 10 ml of sterile distilled water and gently scraping the colony surface with a bent spatula. Conidial suspensions of each fungal isolate were counted with a hemacytometer and diluted to concentrations of 10^6–10^7 conidia per milliliter.

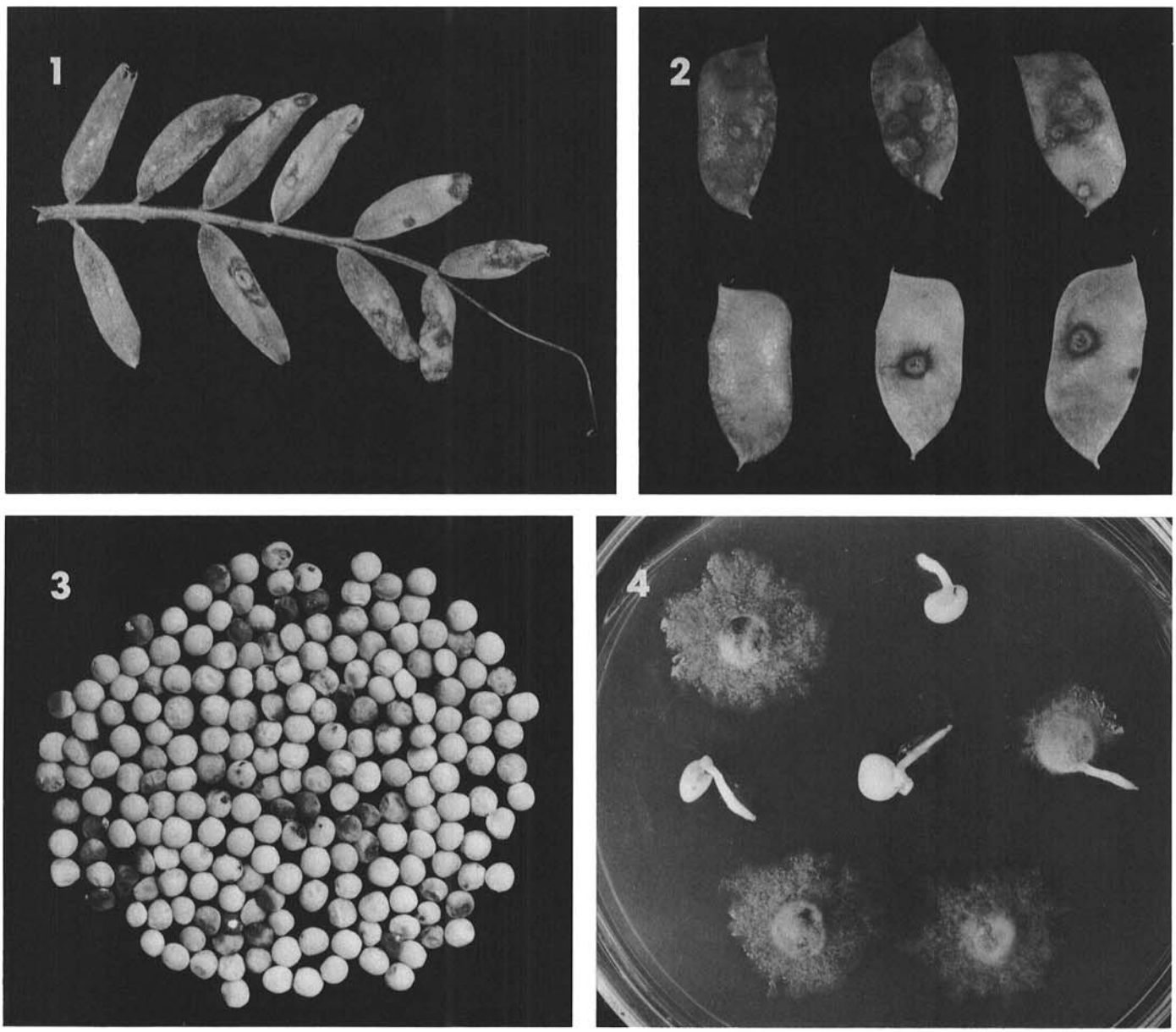
First, the foliage of four to six 20- to 30-day-old lentil (cultivar Chilean 78), chickpea (*Cicer arietinum* L., PI 458870, USA), and pea (*Pisum sativum* L. 'Perfected Wales' or 'Alaska') plants (two to three plants per 15-cm-diameter plastic pot with two to three pots per isolate) was spray-inoculated with 15–20 ml of a spore suspension of two to five isolates of each fungus applied with a DeVilbiss atomizer. After inoculation, plants were placed in a moist chamber for 72–96 hr, then the inoculated and control plants were removed and placed on a greenhouse bench. A completely random design was used to distribute pots both in the moist chamber and on the greenhouse bench. The mean disease index of pathogenicity of each isolate to the foliage was calculated for each host.

Secondly, chickpea, lentil, and pea seeds were surface sterilized in 0.25% NaOCl for 5 min and rinsed in sterile water. After drying, 50 seeds were soaked in 20-ml conidial suspensions of each fungus for 15–20 min and then planted 2 cm deep in 15-cm-diameter pots containing greenhouse potting medium (55% peat moss, 35% pumice, and 10% sand). Ten or fifteen seeds were planted per pot with two to three pots per treatment. Pots were placed in a completely random design on the greenhouse bench and disease index means were calculated as for the foliage response. Pathogenicity of each fungus to the roots and/or foliage of different crop species was based on a disease index of 1–5 in which 1 = healthy tissues, 2 = 1–10%, 3 = 11–25%, 4 = 26–50%, and 5 = more than 50% necrosis of the roots or foliage. Sterile water control treatments were included in all tests. Greenhouse temperatures ranged from 15–25 C. The foliage and roots of the three plant species inoculated with the different fungal isolates and the uninoculated controls were rated for disease after 15–20 days.

Cold-tolerance trial. Original seeds of five lentil PI accessions from Turkey were planted in September or October 1980 in cold-tolerance trials at Pullman and Central Ferry, respectively. Seeds of a local cultivar also were included as a control. Seeds were dusted with captan (50 WP) and planted 1.5–2.0 cm deep with a cone planter in nonreplicated, single-row plots 13 m long with a spacing of 1.5 m between rows. Emergence was recorded in October and November 1980 before the onset of freezing temperatures. A survival count was made in April 1981 after the plants had resumed growth. Plots were harvested in July and August 1981. At Pullman, the trial received only natural rainfall, whereas at Central Ferry the trial was sprinkler irrigated (6 mm) every 6–10 days. The percentage of seedborne *A. lentis* in both original and increase seeds was determined by surface-disinfesting 200 seeds for 5 min in 0.25% NaOCl and plating them on 2% WA (20 seeds per plate and 10 plates per PI accession).

Increase seeds of two blighted Turkish PI accessions from the Pullman cold-tolerance trial were separated by seed size by passing seeds through four metal sieves with round holes graduated into 0.8-mm increments. Two hundred lentil seeds in each size class were tested for seedborne *A. lentis* by using the procedure outlined above. The linear relationship between seed size and percent seed infection by *A. lentis* was tested by using Student's *t*-test on the regression coefficient, and correlation values were calculated.

Incidence of seedborne *A. lentis* in lentil germ plasm. Original seeds of lentil PI accessions from 30 countries were tested for seedborne *A. lentis* by using the procedures outlined previously.



Figs. 1–4. Symptoms and signs of infection of lentil by *Ascochyta lentis*. **1,** Lesions on leaflets. **2,** Lesions on infected pods (healthy pod at lower left). **3,** Lesions on naturally infected seeds. Note discoloration and whitish mycelium. **4,** Growth of *A. lentis* from infected seeds plated on potato-dextrose agar.

Seeds had been stored for up to 33 yr (since 1978 at 4–6 C and 35–40% relative humidity). Since supplies of original seeds of most PI accessions were limited, only 20–80 seeds of each PI accession were tested for seedborne *A. lentis*. Tests were also conducted with 20 PI accessions to determine the incidence of seedborne *A. lentis* in 200 seeds of original and increase seeds of each PI accession. Increase seeds were from plants that had been seeded at Pullman in April to May and harvested in July to August of different years. Plants were grown under dryland conditions.

Survival studies. Naturally infected lentil pods and seeds from the Pullman cold-tolerance trial were used in the survival studies. Survival of the fungus was studied in soil, outdoors in weather station shelters, and in regulated cold storage (4–6 C and 35–40% relative humidity). Diseased pods and seeds were placed between two pieces (10 cm^2) of nylon screen (64 squares per square centimeter) which were stapled together, and either placed on the surface or buried 16 cm deep in a silt loam soil at Pullman. Diseased tissues for the weather station shelters and cold storage treatments were placed in paper bags. Viability of the fungus was tested every 3–4 mo by plating 40–50 pieces of tissue (surface sterilized in 0.25% NaOCl for 5 min) on acidified WA plates (70 μl of 25% lactic acid per 20 ml of 2% WA) (five pieces of tissue per plate with 8–10 plates per treatment). Fungal identifications were made from hyphal-tip and germinating spore cultures on WA plates and PDA slants. The percent survival of *A. lentis* on seeds and pods was plotted against time for each survival treatment.

RESULTS

Isolation from blighted lentils. In June 1981 necrotic lesions were observed on the stems, petioles, leaves (Fig. 1), pods (Fig. 2), and seeds (Fig. 3) of several naturally infected lentil PI accessions in the cold-tolerance trials at Pullman and Central Ferry. Irregularly shaped lesions on the stems, petioles, and leaflets were tan while those on pods and seeds often were darker brown. Black pycnidia were present in most lesions on each type of tissue. Heavily infected seeds were occasionally shriveled and discolored purplish-black, with whitish mycelium and pycnidia present in the lesions.

Over 150 isolations were made from blighted foliage and seeds of these diseased lentil plants. From necrotic lesions on stems, leaves, and pods, a slow-growing pycnidial fungus accounted for more than 50% of the isolates. This fungus was subsequently identified as *A. lentis* (IMI 262298) by the CMI, Kew, U.K. Fungi isolated less frequently (less than 10%) were *Alternaria* sp., *Botrytis cinerea* Pers. ex Pers., *Cladosporium* sp., and *Stemphylium* sp. *A. lentis* was isolated from more than 80% of seeds with discolored, necrotic lesions (Fig. 4). Other fungi isolated occasionally (less than 5%) from diseased seeds were: *Alternaria* sp., *B. cinerea, Cladosporium* sp., *Fusarium avenaceum* (Fr.) Sacc., *Phoma medicaginis* Malbr. and Roum. var. *pinodella* (L. K. Jones) Boerema, and *Stemphylium* sp.

Pathogenicity tests. All isolates of *A. lentis* tested were pathogenic on the foliage but not on the roots of inoculated lentil plants (Table 1). In tests with inoculated seeds, cankers frequently formed at the crown or on the lower stems of plants. Lesions that developed on inoculated lentils were indistinguishable from those observed on naturally infected plants in field trials. Other fungi pathogenic to the roots and/or foliage of lentil were *B. cinerea, F. avenaceum*, and *P. medicaginis* var. *pinodella* (Table 1). *A. lentis* was not pathogenic to chickpea or pea.

Isolates of *Alternaria, Cladosporium*, and *Stemphylium* were not pathogenic to the foliage or roots of chickpea, lentil, and pea. These fungi occasionally were isolated from senescent foliar tissues, particularly leaf tips.

Cold-tolerance trial. *A. lentis* was detected in original seed of three of five Turkish lentil PI accessions; seed infection ranged from 1.5 to 3.5% (Table 2). However, there was a large increase (500% or greater) in the incidence of seedborne *A. lentis* in seeds of three and five Turkish PI accessions harvested at Pullman and Central Ferry, respectively. At Pullman (dryland conditions), *A. lentis* was isolated from increase seeds of four of five Turkish accessions and incidence of infection of the harvested seed ranged from 0.5 to 68.5% (Table 2). At Central Ferry (under sprinkler irrigation), the fungus was isolated from increase seeds of all cold-tolerant lentil PI accessions and incidence of infection of the harvested seed ranged from 10 to 42.5%. Although foliar infection of some Turkish accessions was evident at both locations, the main adverse effect of blight was reduction of seed quality. Shriveled, discolored seeds were particularly prevalent in increase seeds of PI 438516 and PI 438517. *A. lentis* was not isolated from original or increase seeds of cultivar Benewah lentil at either location.

There was a significant correlation between incidence of seedborne *A. lentis* and seed size (Table 3). Infection of seeds by *A. lentis* was greatest in the smallest seed size (3.2 mm) and least in the largest (5.6 mm) for both PI accessions. The difference in infection in seeds among the four size classes was most evident in PI 438516 for which it ranged from 33% in the largest seeds to 65.5% in the smallest seeds.

Seedborne *A. lentis* in lentil germ plasm. A total of 17,060 original seeds from 284 PI accessions representing 30 countries were tested for infection by *A. lentis* (Table 4). The fungus was isolated from 2.0% of the seeds and 16.2% of the PI accessions tested. Seeds from 16 of 30 countries (53%) were infected with the pathogen. The incidence of seedborne *A. lentis* varied greatly among countries and continents. Among countries from which seeds of at least 15 PI accessions were tested, incidence of infection was highest in PI accessions from Turkey (52%) and Syria (44%) and lowest in those from Iran (0%) and Chile (2%) (Table 4). *A. lentis* was isolated from more than 5% of the original seeds tested from Spain (23.3%), India (7.9%), Italy (6.5%), Turkey (6.5%), and Australia (5.5%) (Table 4). The pathogen was isolated from

TABLE 1. Pathogenicity of fungi isolated from necrotic lesions on foliage and seeds of five, cold-tolerant, naturally infected lentil accessions from Turkey to the foliage and roots of chickpea, lentil, and pea

Fungus	Isolates tested (no.)	Pathogenicity to: Chickpea		Lentil		Pea	
		Foliage[a]	Roots[b]	Foliage	Roots	Foliage	Roots
Alternaria sp.	2	1[c]	1	1	1	1	1
Ascochyta lentis	5	1	1	3	1	1	1
Botrytis cinerea	3	4	3[d]	4	4	3	2
Cladosporium sp.	2	1	1	1	1	1	1
Fusarium avenaceum	3	1	4	1	4	1	3
Phoma medicaginis var. *pinodella*	3	2	3	2	3	2	3
Stemphylium sp.	2	1	1	1	1	1	1

[a] Foliage was inoculated with a spore suspension (10^6–10^7 spores per milliliter) of each fungus.
[b] Seeds were inoculated with a spore suspension (10^6–10^7 spores per milliliter) of each fungus before they were planted in sterile potting mix.
[c] Pathogenicity of each fungus to the roots and/or foliage of the test plants was based on a disease index scale of 1–5 in which 1 = healthy tissues, 2 = 1–10%, 3 = 11–25%, 4 = 26–50%, and 5 = more than 50% necrosis of inoculated tissues. The mean disease index of each fungus species was calculated for foliage and root responses for each of the crop species tested.
[d] *Botrytis* causes pre- and postemergence damping-off of chickpea, lentils, and pea.

original seeds of one PI accession from India, one from Syria, and seven from Turkey that had been stored for over 30 yr. *A. lentis* also was isolated from lentil crop debris, particularly pieces of pod that were mixed with seeds of a few lentil PI accessions from Turkey. In one instance, the pod tissue and seeds had been collected in 1948. Although *A. lentis* was the primary pathogen isolated from original seeds of foreign lentil PI accessions, other fungi pathogenic to lentil occasionally were isolated. These included: *B. cinerea, F. avenaceum, Macrophomina phaseolina* (Tassi) Goid., *P. medicaginis* var. *pinodella*, and *Rhizoctonia solani* Kühn. Nonpathogenic fungi also isolated from original seeds included species of *Alternaria, Aspergillus, Chaetomium, Cladosporium, Mucor, Penicillium, Rhizopus*, and *Stemphylium*.

The time of planting (spring versus fall) markedly affects the incidence of seedborne *A. lentis* in the Palouse region. There was a drastic reduction in the incidence of infection by *A. lentis* in original seeds of 20 lentil accessions to seeds of the same accessions increased subsequently in spring plantings at Pullman (Table 5). Although *A. lentis* was present in 5.0–61.7% of the original seeds, the fungus was seedborne in increase seeds (0.5–2.5%) of only five accessions. There did not appear to be any relationship between a high level of seedborne *A. lentis* in the original seed and its transmission to seeds increased subsequently at Pullman in spring plantings.

TABLE 2. Incidence of seedborne *Ascochyta lentis* in original and increase seeds of five lentil accessions from Turkey[a]

	Infection incidence (%)		
		Increase seed[b]	
Lentil line	Original seed[b]	Pullman	Central Ferry
PI 438515	2.0	37.0	10.0
PI 438516	3.5	58.5	26.5
PI 438517	1.5	68.5	42.5
PI 438518	0	0.5	16.5
PI 438519	0	0	13.0
Benewah[c]	0	0	0

[a] Cold-tolerance trials planted at Pullman and Central Ferry, WA, in the fall of 1980 and harvested in the summer of 1981.
[b] Tested 200 original and 200 increase seeds per PI accession for seedborne *A. lentis*. Seeds surface sterilized in 0.25% NaOCl for 5 min and plated on 2% water agar.
[c] Benewah lentil was included as a local control.

TABLE 3. Relationship between seed size and percentage of seed infected with *Ascochyta lentis* in two lentil accessions from Turkey[a]

Lentil accession	Seed size (mm)[b]	Total seed wt. (g)	Percent of total sample	Seed infected (%)[c]
PI 438516	3.2	234	7.5	65.5[d]
	4.0	1,832	58.6	60.5
	4.8	1,029	32.9	48.0
	5.6	30	1.0	33.0
PI 438517	3.2	217	12.7	67.5[e]
	4.0	1,005	58.6	65.5
	4.8	474	27.7	51.0
	5.6	18	1.0	43.5

[a] Planted in a cold-tolerance trial at Pullman, WA, in the fall of 1980 and harvested in the summer of 1981. U.S. Department of Agriculture plant inventory (PI) maintained at the Western Regional Plant Introduction Station, Pullman, WA.
[b] Seeds were sized through metal sieves with four different-sized round holes.
[c] Seeds surface sterilized in 0.25% NaOCl for 5 min and plated on 2% water agar. Tested 200 seeds per size for seedborne *A. lentis*.
[d] Linear contrast over total seed size range is significant at $P = 0.05$. Correlation value $r = -0.98$ at $P = 0.05$.
[e] Linear contrast over total seed size range is significant at $P = 0.05$. Correlation value $r = -0.97$ at $P = 0.05$.

Survival of the fungus. The survival of *A. lentis* in naturally infected lentil pods and seeds from the Pullman cold-tolerance trial was observed over a 3-year period (Fig. 5). The fungus lost its viability after 21 wk in seeds and 29 wk in pods at a soil depth of 16 cm, but remained alive on the soil surface for more than 1.5 yr on pods and 1.7 yr on seeds. The pathogen was still viable in infected tissues after 3 yr at 4–6 C and outdoors in weather station shelters at Pullman and Central Ferry. The pathogenicity of cultures of *A. lentis* from infected tissues incubated in soil, in the weather station shelters, and stored at 4–6 C was tested periodically. All isolates of the fungus tested were still pathogenic to lentils.

DISCUSSION

The USDA lentil germ plasm collection at Pullman was found to harbor several seedborne fungal pathogens. *A. lentis* was the most prevalent and potentially important pathogen detected in original introduced seed from 16 of 30 countries. The fungus was first reported as a pathogen of lentil in Russia in 1938 (3). Subsequently, it has been reported to infect lentil in Argentina (15), Brazil (22), Canada (16), Greece (4), India (13,20), Pakistan (12), and the United States (9,10). In the present study, we have detected *A. lentis* in original seeds of lentil PI accessions from all the countries listed above, except Argentina, and from the following countries in which it appears to be unrecorded: Australia, Ethiopia, Hungary, Italy, Morocco, Spain, Syria, Turkey, and Yugoslavia. Other *Ascochyta* sp., particularly *A. pisi* Lib., have been reported to infect lentil (1,2,14,17). The present study has demonstrated the importance of lentil germ plasm in the introduction, spread, and survival of *A.*

TABLE 4. Incidence of *Ascochyta lentis* in original seeds of lentil (*Lens culinaris*) introduced into the United States from different countries[a]

	Tested (no.)		Infected (no.) with *A. lentis*[b]	
Country	Accessions	Seeds[c]	Accessions	Seeds
Afghanistan	12	720	0	0
Argentina	5	240	0	0
Australia	4	200	2	11
Belgium	1	60	0	0
Brazil	9	540	1	2
Canada	2	200	1	2
Chile	46	2,760	1	1
Costa Rica	1	60	0	0
Ecuador	2	40	0	0
Egypt	3	120	0	0
Ethiopia	3	240	1	5
Greece	7	380	1	8
Guatemala	1	60	0	0
Hungary	7	220	2	3
India	4	240	1	19
Iran	33	1,800	0	0
Italy	4	260	4	17
Jordan	8	480	0	0
Lebanon	9	540	0	0
Mexico	8	480	0	0
Morocco	6	320	2	8
Pakistan	15	1,060	1	38
Peru	1	60	0	0
Spain	2	120	2	28
Syria	18	940	8	25
Turkey	29	2,420	15	157
United States	2	120	0	0
USSR	25	1,300	2	5
Yemen	1	60	0	0
Yugoslavia	16	1,020	2	15
TOTAL:	284	17,060	46	344

[a] Seeds are of lentil plant inventory (PI) accessions in the USDA plant germ plasm collection maintained at the Western Regional Plant Introduction Station, Pullman, WA.
[b] Seeds were surface sterilized in 0.25% NaOCl for 5 min and plated on 2% water agar.
[c] Sixty seeds per accession were tested, except in a few cases where original seed supplies were limiting.

TABLE 5. Incidence of seedborne *Ascochyta lentis* in original and increase seeds of imported lentil PI accessions[a]

Lentil accession	Country of origin	Infection incidence (%)[b]	
		Original seeds[c]	Increase seeds[d]
PI 167386	Turkey	8.3	0.5
PI 169552	Turkey	23.3	0
PI 172993	Turkey	13.3	0
PI 174873	India	31.7	0.5
PI 179324	Syria	6.7	0.5
PI 182220	Turkey	13.3	0
PI 254554	Syria	10.0	0
PI 283608	Australia	40.0	0
PI 297779	Greece	13.3	0
PI 298644	Spain	45.0	0
PI 298923	Italy	8.3	0
PI 298924	Italy	10.0	0
PI 300253	Syria	8.3	0
PI 344077	Turkey	10.0	0
PI 345631	Russia	5.0	0
PI 368645	Yugoslavia	15.0	0
PI 368651	Yugoslavia	10.0	0
PI 370632	Turkey	33.3	2.5
PI 370634	Turkey	61.7	0.5
PI 374120	Morocco	35.0	0

[a] U.S. Department of Agriculture, plant inventory (PI) collection maintained at the Western Regional Plant Introduction Station, Pullman, WA. Increase plots which were grown under dryland conditions at Pullman were planted in April–May and harvested in July–August of different years.

[b] Seeds surface sterilized in 0.25% NaOCl for 5 min and plated on 2% water agar.

[c] Tested 60 seeds per accession, except PI's 283608, 374120, and 298924 where 20, 20, and 80 seeds were tested, respectively.

[d] Tested 200 seeds per accession.

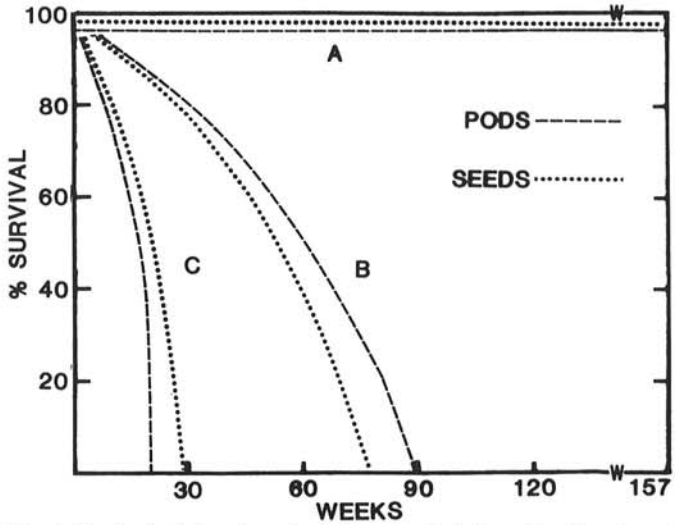


Fig. 5. Survival of *Ascochyta lentis* in naturally infected lentil pods and seeds. **A,** Incubated at 4–6 C or outdoors in weather station shelters. **B,** Placed outdoors on soil surface. **C,** Buried 16 cm in soil.

lentis, and has provided new and pertinent information on the geographic distribution of the pathogen. Also indicated is that plant introduction centers in other countries that maintain *Lens* germ plasm may be harboring seedborne pathogens, like *A. lentis* and pea seedborne mosaic virus. Special procedures are needed to detect and eradicate these pathogens before seeds are distributed to cooperators worldwide.

Ascochyta blight is a potentially important foliar disease of lentil in the Palouse region of northern Idaho and eastern Washington which is the major center of lentil production in the United States, particularly if the crop is grown as a winter annual. This was vividly demonstrated in the lentil cold-tolerance trials at Pullman and Central Ferry in 1980–1981. Cool, wet weather in the spring of 1981 appeared to be favorable for spread and infection of the pathogen, and these conditions have been shown to enhance development of the blight disease (16, and *unpublished*). Rainfall in April–June 1981 was 31–51 mm higher at Central Ferry and Pullman (21), respectively, than the 30-year average. Although the fungus caused necrosis of the foliage of many plants in the trial, the major adverse effect of blight was on reduction in seed quality. In heavily infected accessions, there was extensive discoloration and shriveling of seeds. In Canada, where Ascochyta blight is a major lentil disease (16), Gossen and Morrall (6,7) demonstrated that even slight discoloration of seeds by *A. lentis* resulted in lower seed grades. In both cold-tolerance trials, we observed a dramatic increase (greater than 500%) in the percentage of seeds infected with *A. lentis*, e.g., in lentil PI 438517 seed infection increased from 1.5% in the original seeds to 42.5 and 68.5% in the increase seeds at Central Ferry and Pullman, respectively.

Although *A. lentis* was the most prevalent and potentially important pathogen isolated from seeds of 284 lentil PI accessions tested, other seedborne fungal pathogens were isolated in this study. Several pathogenic fungi isolated occasionally from seeds of the lentil PI accessions included *B. cinerea, F. avenaceum, M. phaseolina, P. medicaginis* var. *pinodella*, and *R. solani*. The authors (*unpublished*) have isolated all of these fungi from seeds of commercial lentils grown in the Palouse region, and some of these fungi have been reported to cause root and foliar diseases of commercial lentils in the Pacific Northwest (23). Even though there was a low incidence of infected seeds in the PI accessions tested, there is the potential danger of introducing new and/or more virulent strains of these seedborne pathogens with imported lentil germ plasm.

At Pullman, *A. lentis* survived for 1.5 yr in infected pods and seeds on the soil surface, but lost viability in 29 wk when buried in the soil. Deep plowing of infected debris is a potentially useful cultural control measure which should result in fairly rapid inactivation of the pathogen. Combined with crop rotation and use of a seed treatment fungicide, the likelihood of reinfection of subsequent lentil plantings should be greatly reduced. However, care must be taken to avoid introduction of the pathogen on contaminated seeds. Seed transmission is an important factor in spread and survival of the fungus (4,16). *A. lentis* survived long periods in infected lentil seeds, even under adverse environmental conditions. Controlling the seedborne phase of the disease with seed treatment fungicides would help to prevent introduction of the blight pathogen with infected seeds. Thiabendazole, a systemic fungicide, controls the seedborne phase of the disease (11), but this chemical has not been registered for use on lentil in the Pacific Northwest. Further research on seed treatments for control of *A. lentis* is needed.

LITERATURE CITED

1. Anonymous. 1982. Plant Disease Survey, 1980–1981. 50th Annual Report, N.S.W. Dep. Agric., Rydalmere, N.S.W., Australia. 47 pp.
2. Bellar, M., and Kebabeh, S. 1983. A list of diseases, injuries, and parasitic weeds of lentils in Syria (Survey 1979–1980). Lens Newslett. 10:30-31.
3. Bondartzeva-Monteverde, V. N., and Vassilievsky, N. I. 1940. A contribution to the biology and morphology of some species of *Ascochyta* on Leguminosae. Pages 345-376 in: Acta Inst. Bot., Acad. Sci. U.R.S.S. 1938, Ser. II (Pl. Crypt.) (In Russian) (In: Rev. Appl. Mycol. 20:232-233, 1941).
4. Davatzi-Helena, K. 1980. A disease of lentil caused by *Ascochyta lentis* Vassiljevsky. Ann. Inst. Phytopathol. Benaki (N.S.) 12:256-260.
5. Erskine, W. 1984. Evaluation and utilization of lentil germplasm in an international breeding program. Pages 225-237 in: Genetic Resources and Their Exploitation—Chickpeas, Faba Beans and Lentils. J. R. Witcombe and W. Erskine, eds. Martinus Nijhoff/Dr. W. Junk, Publishers, The Hague/Boston/Lancaster.
6. Gossen, B. D., and Morrall, R. A. A. 1983. Effect of ascochyta blight on seed yield and quality of lentils. Can. J. Plant Pathol. 5:168-173.
7. Gossen, B. D., and Morrall, R. A. A. 1984. Seed quality loss at harvest due to ascochyta blight of lentil. Can. J. Plant Pathol. 6:233-237.

8. Hampton, R. O. 1982. Incidence of the lentil strain of pea seedborne mosaic virus as a contaminant of *Lens culinaris* germ plasm. Phytopathology 72:695-698.
9. Kaiser, W. J. 1983. Plant introduction and related seed pathology research in the United States. Seed Sci. Technol. 11:1197-1212.
10. Kaiser, W. J., and Hannan, R. M. 1982. *Ascochyta lentis*: Incidence and transmission in imported lentil seed. (Abstr.) Phytopathology 72:944.
11. Kaiser, W. J., and Hannan, R. M. 1984. Chemical control of *Ascochyta lentis* in infected lentil seeds. (Abstr.) Phytopathology 74:1139.
12. Khan, B. A., Haq, I. U., Rehman, F. U., and Aslam, M. 1983. Ascochyta blight of lentil—A new disease in Pakistan. Pak. J. Bot. 15:121.
13. Khare, M. N., Agrawal, S. C., and Jain, A. C. 1979. Diseases of lentil and their control. Tech. Bull., Jawaharlal Nehru Krishi Vishwa Vidyalaya, Jabalpur, India. 29 pp.
14. Khatri, H. L., and Singh, K. 1975. Lentil blight—A new record for India. Labdev J. Sci. Tech. 13-B:73-74.
15. Mitidieri, I. Z. M. de. 1975. La mancha de la lenteja (*Lens culinaris* Medik.) causada por *Ascochyta lentis* Bond. y Vassil., en la Argentina. Rev. Invest. Agropecu., Ser. Cinco, Patol. Veg. Buenos Aires 11:43-55.
16. Morrall, R. A. A., and Sheppard, J. W. 1981. Ascochyta blight of lentils in western Canada: 1978 to 1980. Can. Plant Dis. Surv. 61:7-13.
17. Mujica, F. 1943. Hongos chilenos no mencionados anteriormente en la literatura. Bol. Sanidad Veg. Chile 3:33-35. (In: Rev. Appl. Mycol. 23:475-476, 1944).
18. Nelson, P. E., Toussoun, T. A., and Marasas, W. F. O. 1983. *Fusarium* species, an illustrated manual for identification. The Pennsylvania State University Press, University Park. 193 pp.
19. Sepúlveda, P., and Alvarez, M. 1982. Determinacion de *Ascochyta lentis* Bond. y Vassil. en lenteja (*Lens culinaris* M.). Agric. Tec. (Santiago) 42:351-353.
20. Singh, G., Singh, K., Gill, A. S., and Brar, J. S. 1982. Screening of lentil varieties/lines for blight resistance. Indian Phytopathol. 35:678-679.
21. United States Department of Commerce. 1981. Climatological data for the State of Washington. Annual Summary, Vol. 85 (13). National Oceanic and Atmospheric Administration, National Climate Center, Asheville, NC.
22. Veiga, P., Manara, W., Manara, N. T. F., and Tarragó, M. F. S. 1974. Ocorrência de *Ascochyta lentis* Bond. et Vassil. em lentilha (*Lens culinaris* Medik) no Rio Grande do Sul. Rev. Cent. Cienc. Rurais 4:239-242.
23. Wilson, V. E., and Brandsberg, J. 1965. Fungi isolated from diseased lentil seedlings in 1963-64. Plant Dis. Rep. 49:660-662.