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Pathological Studies of Fungi Associated with Pulse Seeds during Storage in Dammam Province, Kingdom of Saudi Arabia

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ABSTRACT

This study aimed to identify fungi associated with ten varieties of legumes seeds in an eastern region of the Kingdom of Saudi Arabia: Lupine (*Lupinus albus* L.), dry and kidney beans (*Phaseolus vulgaris*), cowpea and mung beans (*Vigna radiata* L.), faba and field beans (*Vicia faba* L.), brown and green lentil (*Lens culinaris*), chickpea (*Cicer judaicum*). The highest percentage of fungal infection was associated with mung bean (23.29% of infection) followed by faba beans (15.75%), field beans (13.87%), dry beans (11.99%), brown lentil (11.13%), lupine (6.69%), kidney beans (6.16%), green lentil (5.14%), cowpea (3.25%) and chickpea (2.74%). Several fungi associated with these were isolated at the following frequencies: *Rhizoctonia solani* (21.18%), *Pythium aphanidermatum* (17.8%), *Sclerotinia sclerotiorum* (13.04%), *Alternaria alternata* (12.18%), *Aspergillus flavus* (9.43%), *Penicillium* spp. (6.86%), *Aschocayta* spp. and *Phytophthora* spp. (4.12%). The potency of the first four most frequent fungi to infect all studied seeds was ranked, in decreasing order, as: *R. solani* (35.33%), *P. aphanidermatum* (28.67%), *A. alternate* (27.67%), *S. sclerotiorum* (18.67%). The weight of infected plants was significantly lower than that of healthy plants.

Keywords: beans, chickpea, lentil, lupine

INTRODUCTION

Seeds of legumes are valuable food resources, and are considered alternatives to meat as they contain proteins (20-30% of dry weight). There are more than 500 varieties of pulses that play a useful role in soil by an association with nitrogen-fixing bacteria, by increasing soil fertility. These seeds also have a low fat content (about 5%), carbohydrates, fibers, iron, zinc, calcium, and folic acid. Legume seeds contain ω-3 which helps to prevent cancer (Scarmeas *et al.* 2006; Scarafoni *et al.* 2007; Zhang 2007; Tang *et al.* 2008; Villegas *et al.* 2008). During storage, seeds are exposed to infection by microbes and insects.

The most important pathogenic genera of fungi which infect seeds are *Colletotrichum*, *Sclerotinia*, *Alternaria*, *Fusarium*, *Rhizoctonia*, *Pythium*, *Ascochyta* and *Botrytis* (Sumar *et al*. 1982; Sweetingham 1989; Mackie *et al*. 1999; Lardner *et al*. 1999; Zhang and Yang 2000; Elmer *et al*. 2001; Anastasios *et al.* 2005; Wen *et al.* 2005). Storage fungi of the genera *Aspergillus* and *Penicillium* may also appear with high moisture. These fungi produce aflatoxins which destroy the liver and induce carcino-, muta- and teratogenesis (Pereyra *et al.* 2008).

Identification of fungi associated with such seeds gives an idea of some of the problems faced by seeds in the cultivation of legumes, especially during the growth of a seedling, that occur in varying degrees among different species. Swanson *et al*. (1984) noted that the injury plants pulses in the field varied: 52, 15, 8 and 4% in pea (*Pisum sativum*), common beans (*Phaseolus vulgaris*), faba beans (*Vicia faba* L.) and lentil (*Lens culinaris*), respectively.

Many authors proved such infection ands its effect on whole plants leading to great economic loss: Burgess *et al*. (1997), Phan *et al*. (2002), Yang *et al*. (2002). Diseases caused by *Sclerotinia sclerotiorum* and *Sclerotinia minor* are responsible for economically important losses on several crops in eastern Canada, including canola, cabbage, carrots, celery, lettuce, snap beans, soybeans, and white beans (McDonald and Boland 2004; McLaren *et al*. 2004). Ascochyta blight (AB), caused by *Ascochyta rabiei*, is a major disease of chickpea (*Cicer arietinum* L.). Several epidemics of AB causing complete yield loss have been reported by Pande *et al*. (2005). Botrytis grey mould (BGM), caused by *Botrytis cinerea* Pers. ex. Fr., is an economically important disease of chickpea (*Cicer arietinum* L.), especially in areas where cool, cloudy, and humid weather persists. Several epidemics of BGM causing complete crop loss in the major chickpea-producing countries have been reported by Pande *et al*. (2006).

The aim of this study was to isolate fungi associated with the seeds of 10 varieties of pulses in markets in Dammam province in the eastern region of Saudi Arabia, namely: lupine, dry beans, kidney beans, cowpea, mung beans, brown lentil, green lentil, field beans, chickpea and faba beans. The percentage infection and potency of the more frequent fungi were studied.

MATERIALS AND METHODS

Sample collection

Samples of 10 types of seeds (Lupine (*Lupinus albus* L.), dry and kidney beans (*Phaseolus vulgaris*), cowpea and mung beans (*Vigna radiata* L.), faba and field beans (*Vicia faba* L.), brown and green lentil (*Lens culinaris*), and chickpea (*Cicer judaicum*) were collected from 5 different grocery stores for the following studies.

Percentage germination

One hundred seeds from each plant were sterilized in 2% sodium hypochlorite and kept at room temperature between moist filter papers in Petri dishes for about 7 days. Percentage germination was calculated according to El-Awadi (1993).

$$\% \text{ Germination} = \frac{\text{number of germinated seeds}}{\text{total number of seeds}} \times 100$$

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Estimation of relative humidity

The "two-stage air oven method" (Anonymous 1975) was used. A known seed weight was placed in an oven at 70°C. After 24 h its weight was recorded. Samples were redried and every hour the sample weight was recorded until a constant weight was obtained. The moisture content was calculated as:

$$\text{Moisture content} = \frac{\text{moist weig ht - dry weight}}{\text{moist weig ht}} \times 100$$

Percentage external infection

Five hundred seeds, chosen at random from each seed batch was used to calculate the percentage infection according to the Saber (1984) method using the equation:

$$\text{\% External infection} = \frac{\text{number of external infected seeds}}{\text{total number of seeds}} \times 100$$

External infection appeared as coloured, irregular or atrophic seeds (**Figs. 1-10**).

Isolation and identification of fungi

The coloured or irregular and atrophied seeds were chosen, disinfected by immersing in 10% household chlorine bleach ($NaClO_2$) for 3 min, rinsed in distilled water for 5 min and dried for almost 1 min (Elmer *et al*. 2001). Seeds were plated on Czabek's agar media (Onkar and Sinclair 1985). Five seeds were placed in each Petri dish with ten replicates and incubated at 28 ± 2°C for five days. The kind and number of fungi were recorded using the hyphal tip technique suggested by Dhingra and Sinclair (1985). The isolated fungi were identified according to their morphological characters. Identification of genus and species was confirmed with help of the Department of Mycology at the Agricultural Research Institute, A.R.E.

Frequency of fungal infection

The frequency of infection was calculated to determine the most susceptible seeds to fungal infection.

$$\text{\% Frequency} = \frac{\text{total number of fungal isolates/c rop}}{\text{total number of fungal isolates}} \times 100$$

Pathogenicity tests

Four fungal genera, representing the most frequently isolated seed pathogens, named *Rhizoctonia solani* (frequency 21.18%), *Pythium aphanidermatum* (17.8%), *Sclerotinia sclerotiorum* (13.04%), *Alternaria alternata* (12.18%) were grown on PDA media. Two hundred grams of barley seeds plus sixty grams of sand with 200 ml H_2O were autoclaved (Abada 1986) and used as media to cultivate fungi by incubating at 25 ± 2°C for 15 days. Sterilized pots with sand were used to mix soil with 5% of fungal isolate (Elian 1978) irrigated every two days for 15 days. Sterilized seeds were cultivated (15 seeds/pot) with three replicates in each infected soil with three control pots without fungal isolates. Fungal potency was estimated by calculating the percentage of pre-emergence damping off seedlings after 15 days post-cultivation or post-emergence damping-off seedlings with root rotting symptoms (at 30 days cultivation). The percentage normal seed growth was used for comparison. The wet and dry weights of the shoot system of one month-old plants were calculated.

Statistical analysis

Data obtained were statistically analyzed using SPSS Ver 6. Treatment averages were compared at the 0.05 level of probability using LSD (Marija 1990).

RESULTS AND DISCUSSION

Detection of fungal infection

Kidney beans had the highest percentage infection (26.1%) followed by faba beans (19.5%), dry beans (11.2%), field beans (10.8%), cowpea (9.9%), brown lentil (7.7%), green lentil (7.20%), mung bean (6.68%) and finally chickpea (3.7%) (**Table 1**). The percentage seed germination ranged from zero to 99.4%. The loss of seed viability may be due to exposure to high temperature for preservation, or by exposure to microbial infection (**Table 1**).

There was great variation between germination rates, external symptoms of infection and moisture content of seeds (**Table 1**). The coloration of seeds may be a reaction between nitrogenous compounds and reduced sugars and the loss of seed viability may be due to microbial infection, especially by fungi, or internal changes due to bad storage conditions such as oxidation of some compound fats (Christensen 1973; Bothast 1978; Osman 1982).

Table 1 Percentage of external symptoms, percentage germination and percentage moisture content of 10 varieties of dried pulse seeds from Dammam at the Eastern Province of Saudi Arabia (n = 5).

Pulse crop	% External symptoms	% Germination	% Moisture content
Lupine	7.56 ± 9.33	94.24 ± 2.61	5.42 ± 1.12
Dry beans	11.20 ± 7.32	32.24 ± 44.16	6.75 ± 1.66
Kidney beans	26.12 ± 13.66	45.20 ± 41.56	6.63 ± 0.99
Brown lentil	7.72 ± 3.50	93.11 ± 2.66	7.65 ± 2.19
Green lentil	7.34 ± 2.87	88.08 ± 16.99	8.27 ± 0.49
Faba beans	19.50 ± 10.15	81.56 ± 15.52	5.83 ± 1.61
Field beans	10.80 ± 4.45	57.88 ± 14.46	6.46 ± 1.24
Mung beans	6.68 ± 7.41	95.44 ± 5.91	6.73 ± 1.34
Chickpea	3.64 ± 2.33	84.69 ± 21.57	6.18 ± 0.80
Cowpea	9.88 ± 6.42	93.43 ± 5.74	8.43 ± 1.71

Table 2 Number of fungal isolate obtained from 10 varieties of dried pulses from Dammam at the Eastern Province of Saudi Arabia.

Pulse crop	Total № of fungal isolates/crop	% Frequency
Lupine	39	6.69
Dry beans	70	11.99
Kidney beans	39	6.69
Brown lentil	65	11.13
Green lentil	30	5.14
Faba beans	92	15.75
Field beans	81	13.87
Mung beans	136	23.29
Chickpea	19	3.25
Cowpea	19	3.25
Total № of fungal isolates	584	

Table 3 Number of isolates obtained from 10 varieties of dried pulses from Dammam at the Eastern Province of Saudi Arabia.

Isolated fungi	Total № of fungal isolates	% Frequency
Rhizoctonia solani	127	21.18
Pythium aphanidermatum	104	17.80
Sclerotinia sclerotiorum	76	13.04
Alternaria alternata	71	12.18
Aspergillus flavus	55	9.43
Penicillium	40	6.86
Aschocayt spp.	24	4.12
Phytophthora spp.	24	4.12
Aspergillus niger	23	3.95
Rhizopus stolonifer	16	2.74
Fusarium	10	1.72
Mucor sp.	9	1.50
Aspergillus spp.	4	0.70
Total № of fungal isolates	584	

Fig. 1 Healthy and infected seeds of 10 pulse crops examined in this study. (**A**) Lupine seeds. Left = infected seeds, discoloured and stained, caused by fungi. Right = healthy, well grown seeds. (**B**) Dry beans seeds. Left = infected seeds, discoloured, stained, wrinkled and small, caused by fungi. Right = healthy, well grown seeds. (**C**) Kidney bean seeds. Left = infected seeds, discoloured, stained, wrinkled and small, caused by fungi. Right = healthy, well grown seeds. (**D**) Mung bean seeds. Left = infected seeds, discoloured, stained, wrinkled and small, caused by fungi. Right = healthy, well grown seeds. (**E**) Cowpea seeds. Left = infected seeds, discoloured, stained, wrinkled and small, caused by fungi. Right = healthy, well grown seeds. (**F**) Faba bean seeds. Left = infected seeds, discoloured, stained, wrinkled and small, caused by fungi. Right = healthy, well grown seeds. (**G**) Field bean seeds. Left = infected seeds, discoloured, stained, wrinkled and small, caused by fungi. Right = healthy, well grown seeds. (**H**) Green lentil seeds. Left = infected seeds, discoloured, stained, wrinkled and small, caused by fungi. Right = healthy, well grown seeds. (**I**) Brown lentil seeds. Left = infected seeds, discoloured, stained, wrinkled and small, caused by fungi. Right = healthy, well grown seeds. (**J**) Chickpea seeds. Left = infected seeds, discoloured, stained, wrinkled and small, caused by fungi. Right = healthy, well grown seeds.

Fungal isolation

Fungi were frequently isolated from seeds (**Table 2**), most frequently in mung bean (23.3%) while 15.8% was isolated in faba bean, 13.9% infection in field beans followed by dry beans (12.0%), brown lentil (11.1%), lupine and kidney beans (6.7%), green lentil (5.1%) and the least percentage was recorded in chickpea and cowpea (3.3%).

The fungus *Rhizoctonia solani* was most frequently isolated (21.2%) from all isolates, followed by *Pythium aphanidermatum* (17.8%), *Sclerotinia sclerotiorum* (13.0%) and *Alternaria alternata* (12.18%) (**Table 3**). Differences in isolation frequency may be due to variation in the moisture content of seeds, varietal differences and seed susceptibility to infection besides specific environmental conditions in each seed store.

Different authors isolate related species of fungi from legumes seeds: Tu (1984) isolated *Fusarium oxysporum*, *Alternaria alternata*, and *Rhizoctonia solani* from dry beans; El-Toony (1987) isolated *R. solani* and *Pythium debaryanum* from pea seeds; Edema *et al*. (1995) mentioned that the most important fungi that infected cowpea in Uganda were *Colletotrichum*, *Ascochyta phaseolorum*, *Cladosporium vignae*, *Fusarium solani*, *Tracheiphilum lindemuthianum*, and *F. oxysporum f.* sp.

Our results concerning the differences in frequency of fungal isolation are in agreement with the results of several researchers (MacLeod and Sweetingham 1997; Abdel-Hafez 2004; El-Hussieni and Al-Shebel 2007).

Pathogenicity of isolated fungi

The obtained data are presented in **Tables 4-7**. The pathogenicity of the four frequent fungi in isolation, *Rhizoctonia solani*, isolated from field bean which has 24 from 127 isolates followed by *Pythium aphanidermatum*, from Mung beans (20 from total 104 isolates), *Sclerotinia sclerotiorum* from mung beans (23 from 76 isolates), and *Alternaria alternata* from lupine (9 from 71 isolates). The pathogenicity of the four fungi was measured against all tested cereals. Percentage of infection was estimated by measuring the number of infected seedlings before and after germination. Results proved the virulence of the four fungi to infect all tested seeds with different degrees.

Results in **Table 4** revealed that all (germinated seeds) were infected by *Sclerotinia sclerotiorum*. mung beans was the highest susceptible seeds with infection percentage (36.7%) followed by field beans (30.0%) green lentil

Table 4 Effect of the artificial inoculation with most frequent isolated fungi from the seeds of pulses tested.

Crops	*Sclerotinia sclerotiorum*				Control			
	% Infected plants			% Healthy plants	% Infected plants			% Healthy plants
	Pre-emergence	Post-emergence	Total		Pre-emergence	Post-emergence	Total	
Lupine	10.00	10.00	20.00	80.00	0	0	0	100.0
Dry beans	3.34	16.70	20.00	80.00	0	0	0	100.0
Kidney beans	3.34	3.34	6.67	93.30	3.34	0	3.34	96.7
Mung bean	13.30	23.30	36.70	63.30	0	3.34	3.34	96.7
Cow pea	10.00	3.34	13.30	86.70	0	0	0	100.0
Faba beans	10.00	0	10.00	90.00	0	0	0	100.0
Field beans	26.70	3.34	30.00	70.00	0	0	0	100.0
Brown lentil	0	16.70	16.70	83.30	3.34	0	3.34	96.7
Green lentil	0	23.30	23.30	76.70	0	0	0	100.0
Chickpea	6.67	3.34	10.00	90.00	0	0	0	100.0
L.S.D. at 5%	3.42	4.38	5.49	5.49	1.57	1.29	1.57	1.57

Table 5 Effect of artificial inoculation with most frequent isolated fungi from the seeds of pulses tested.

crops	*Pythium aphanidermatum*				Control			
	% Infected plants			% Healthy plants	% Infected plants			% Healthy plants
	Pre-emergence	Post-emergence	Total		Pre-emergence	Post-emergence	Total	
Lupine	30.00	20.00	50.00	50.00	0	0	0	100.0
Dry beans	13.30	13.30	26.70	73.30	0	0	0	100.0
Kidney beans	30.00	10.00	40.00	60.00	3.34	0	3.34	96.7
Mung bean	33.30	46.70	80.00	20.00	0	3.34	3.34	96.7
Cowpea	3.34	30.00	3.34	66.70	0	0	0	100.0
Faba beans	20.00	6.67	26.70	73.30	0	0	0	100.0
Field beans	3.34	13.30	16.70	83.30	0	0	0	100.0
Brown lentil	0	10.00	10.00	90.00	3.34	0	3.34	96.7
Green lentil	0	23.30	23.30	76.70	0	0	0	100.0
Chickpea	20.00	0	20.00	80.00	0	0	0	100.0
L.S.D. at 5%	7.96	7.45	11.78	7.80	1.57	1.29	1.57	1.57

Table 6 Effect of the artificial inoculation with most frequent isolated fungi from the seeds of pulses tested.

Crops	*Rhizoctonia solani*				Control			
	% Infected plants			% Healthy plants	% Infected plants			% Healthy plants
	Pre- emergence	Post- emergence	Total		Pre- emergence	Post- emergence	Total	
Lupine	50.00	26.70	76.70	23.30	0	0	0	100.0
Dry beans	10.00	10.00	20.00	80.00	0	0	0	100.0
Kidney beans	26.70	26.70	53.30	46.70	3.34	0	3.34	96.7
Mung bean	16.70	46.70	63.30	36.70	0	3.34	3.34	96.7
Cowpea	20.00	50.00	70.00	30.00	0	0	0	100.0
Faba beans	13.30	0	13.30	86.70	0	0	0	100.0
Field beans	10.00	6.67	16.70	83.30	0	0	0	100.0
Brown lentil	6.67	3.34	10.00	90.00	3.34	0	3.34	96.7
Green lentil	0	6.67	6.67	93.30	0	0	0	100.0
Chickpea	10.00	13.30	23.30	76.70	0	0	0	100.0
L.S.D. at 5%	10.54	11.15	18.33	18.33	1.57	1.29	1.57	1.57

Table 7 Effect of the artificial inoculation with most frequent isolated fungi from the seeds of pulses tested.

Crops	*Alternaria alternata*				Control			
	% Infected plants			% Healthy plants	% Infected plants			% Healthy plants
	Pre-emergence	Post-emergence	Total		Pre-emergence	Post-emergence	Total	
Lupine	20.00	36.70	56.70	43.30	0	0	0	100.0
Dry beans	0	13.30	13.30	86.70	0	0	0	100.0
Kidney beans	10.00	16.70	26.70	73.30	3.34	0	3.34	96.7
Mung bean	20.00	36.70	56.70	43.30	0	3.34	3.34	96.7
Cowpea	40.00	40.00	80.00	20.00	0	0	0	100.0
Faba beans	0	3.34	3.34	93.30	0	0	0	100.0
Field beans	10.00	0	10.00	90.00	0	0	0	100.0
Brown lentil	6.67	3.34	10.00	89.30	3.34	0	3.34	96.7
Green lentil	3.34	6.67	10.00	89.30	0	0	0	100.0
Chickpea	3.34	6.67	10.00	90.00	0	0	0	100.0
L.S.D. at 5%	6.67	18.63	9.62	9.62	1.57	1.29	1.57	1.57

Table 8A Effect of artificial inoculation with most frequently isolated fungi on the wet weight of shoots of pulses after one month of agriculture.

Fungi	Legume variety										% Loss of shoot system
	Lupine	Dry beans	Kidney beans	Mung bean	Cowpea	Faba beans	Field beans	Brown lentil	Green lentil	Chickpea	
Sclerotinia sclerotiorum	1.55	1.39	1.39	0.34	0.61	1.59	8.53	0.5	0.07	0.74	
% -	63.36	61.81	42.80	37.04	75.79	55.59	41.38	73.68	66.67	71.65	58.68
Pythium aphanidermatum	2.03	1.195	1.09	0.24	0.81	2.78	5.93	0.04	0.05	0.18	
% -	52.01	67.17	55.14	55.55	67.86	22.35	59.24	78.95	76.19	93.10	62.76
Rhizoctonia solani	2.1	2.01	1.7	0.23	0.31	3.23	6.73	0.05	0.09	0.54	
% -	50.36	44.78	30.04	57.41	87.7	9.78	53.75	73.68	57.14	79.31	54.40
Alternaria alternata	1.81	1.37	1.94	0.24	0.84	1.99	7.04	0.06	0.05	1.38	
% -	57.21	62.36	20.17	55.55	66.67	44.41	51.16	68.42	76.19	47.13	54.93
Control	4.23	3.64	2.43	0.54	2.52	3.58	14.55	0.19	0.21	2.61	
L.S.D. at 5%	1.04	0.96	0.77	0.36	0.9	0.96	1.9	0.25	0.26	0.9	

Table 8B Effect of artificial inoculation with most frequently isolated fungi on the dry weight of shoots of pulses after one month of agriculture.

Fungi	Legumes variety										% Loss of shoot system
	Lupine	Dry beans	Kidney beans	Mung bean	Cowpea	Faba beans	Field beans	Brown lentil	Green lentil	Chickpea	
Sclerotinia sclerotiorum	0.18	0.15	0.15	0.04	0.07	0.195	0.85	0.03	0.04	0.18	
% -	51.35	62.5	70	33.33	61.11	40.91	27.35	80	75.61	47.06	54.92
Pythium aphanidermatum	0.195	0.13	0.12	0.05	0.07	0.23	0.59	0.02	0.04	0.17	
% -	47.30	67.5	76	16.67	61.11	30.30	49.57	86.67	75.61	50	56.07
Rhizoctonia solani	0.22	0.34	0.35	0.045	0.04	0.25	0.67	0.015	0.04	0.2	
% -	40.54	15	30	25	77.78	24.24	42.74	90	75.61	41.18	46.02
Alternaria alternata	0.2	0.12	0.15	0.04	0.09	0.18	0.55	0.03	0.04	0.2	
% -	45.95	70	70	33.33	50	45.46	52.99	80	75.61	41.18	56.45
Control	0.37	0.4	0.5	0.06	0.18	0.33	1.17	0.15	0.164	0.34	
L.S.D. at 5%	0.28	0.39	0.4	0.099	0.24	0.26	0.5	0.08	0.085	0.27	

(23.3%) dry beans (20.0%), lupine (20.0%), Brown lentil (16.7%), cowpea (13.3%), faba beans (10.0%), chickpea (10.0%) and kidney beans (6.7%).

The fungus *Pythium aphanidermatum* infected mung beans greatly (80.0%) (**Table 5**), followed by lupine (50%), kidney beans (40.0%), dry beans and faba beans (26.7%), green lentil (23.3%), chickpea (20%), field beans (16.7%), brown lentil (10.0%) and cowpea (3.34%).

Results in **Table 6** proved also that the fungus *Rhizoctonia solani* infected to different degrees, in decreasing order: lupine (76.7%), cowpea (70.0%), mung bean (63.3%), kidney beans (53.3%), chickpea (23.3%), dry beans (20%), field beans (16.7%), faba beans (13.3%), brown lentil (10.0%) and green lentil (6.7%). While the fungus *Alternaria alternata* (**Table 7**) affected the seeds of cowpea most (80.0%), followed by mung beans and lupine (56.7%), kidney beans (26.7%), dry beans (13.3%), chickpea has (10.0%), field beans (10%), faba beans (3.3%), and green and brown lentil (6.7 and 3.3% respectively)

From results it appeared that the most frequently isolated fungal species are not the most pathogenic. Some fungi can infect seeds before harvest but not at the storage stage while others have reverse effect. Similar results were observed by several authors (Vock *et al.* 1980; Khan *et al.* 1999; Pande *et al.* 2005).

Differences in susceptibility to infection could also be attributed to the difference in the genetic structure of each seed, as also reported by several groups (Kohpina *et al.* 2000; Nguyen *et al.* 2005; Al-Shebel and El-Hussieni 2007).

Different secretions from the roots of these varieties may also play some role in protecting plants from fungal infection (Youssef *et al.* 1975; McLaren *et al.* 2004; McDonald and Boland 2004; Ralph *et al.* 2006). Differences in isolation frequency may be due to environmental factors and variation in each cultivated land, especially relative humidity. The type of soil and fertilization system plays a role in fungal infectivity (Kimati and Mascarenhas 1967; Baker and Martinson 1970). Chongo *et al.* (2001) found that the virulence of *Fusarium graminearum* against faba beans, peas, lentil and chickpea increased with increasing temperature from 10 to 30°C.

Results in **Table 8A** proved that fungal infection affects seedlings in a significant way. Signs of infection did not appear in growing seedlings of brown lentil, but shoots showed 80% growth reduction, followed by green lentil (75.9%), kidney beans (70%), dry beans (62.5%), cowpea (61.1%), lupine (51.5%), chickpea (47.1%), faba beans (27.4%) mung beans (33.33%) and finally field beans (27.4%), when infected by *Sclerotinia sclerotiorum*.

In the case of artificial inculation by *Pythium aphanidermatum* (**Table 8B**), the most affected seedlings were those of brown lentil (86.7%), kidney beans (76.0%), green lentil (75.6%), dry beans (67.5%), cowpea (61.1%), chickpea (50.0%), field and faba beans (49.6%), lupine (47.3%) and mung beans (16.7%).

The lowest rate of growth, after infection by *Rhizoctonia solani* was recorded on seedlings of brown lentil (90.0%), followed by cowpea (77.8%), green lentil (75.6%), field beans (42.7%), chickpea (41.2%), lupine (40.5%), kidney beans (30.0%), mung beans (25.0%), faba beans (24.2%) and dry beans (15.0%).

Artificial inculation by *Alternaria alternata* decreased the rate of seedling emergence in brown lentil (80.0%) followed by green lentil (75.6%), dry and kidney beans (70.0%), field beans (53.0%), cowpea (50.0%), lupine (46.0%), faba beans (45.5%), chickpea (41.2%) and mung beans (33.3%).

Results showed that brown lentil was the most susceptible plant to all fungal infections, followed by green lentil; the other seeds showed variable susceptibility to fungi.

Our results agree with those of Pakhomova (1969), Chul-kina (1971), Sidorov (1990), Wildermuth *et al.* (1992), Al-Abdalall (1998), Paulitz *et al.* (2002), Smith *et al.* (2003a), and Smiley *et al.* (2005a, 2005b).

Fungi use plant sugars for growth and respiration which affects the carbohydrate content of seeds leaving them irregular and abnormal beside the weakness of the growing seedlings (Younes 1977; Shalaby 1988; Wildermuth *et al.* 1992; Al-Abdalall 1998). The disorder in food changes in infected plants explains the fungus consumption of lipids and proteins in seeds.

Generally, rotten roots lead to loss of loss of shoot numbers, shrinkage and atrophy of seeds with loss of its food value which lead to loss of the crops (Sidorov 1990; Wildermuth *et al.* 1992; Lucas *et al.* 1993; Smith *et al.* 2003b).

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