



ISOLATION LEGUMES SEED BORNE FUNGI AND THEIR EFFECT IN SEED GERMINATION

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ABSTRACT 255 fungal isolates were obtained and identified, these isolates belong to different species as well as 89 yeast isolates from 15 samples of legume seeds, they were 96 isolates from seeds of pea (*Pisum sativum* L.) cultivar English produced in season (2010-2011), and 93 isolates from seeds of cultivar Jof produced in season (2011-2012) and one isolate from lentil seeds (*Lens culinaris*), the fungus *Aspergillus terreus* was the most frequently in legume seeds with rate (19.18%) followed by *Aspergillus niger* (18.31%), A preliminary test of pathogenicity by cabbage seeds which included three separated experiments, showed that all the 71 isolates caused a significant reduction in the rate of cabbage seeds germination but only ten isolates were highly pathogenic, the percentage of germination in their treatments were 0-15% compared to 97% in control treatments, the isolates of *Fusarium spp.* which include Fs-1, Fs-2 and Fs-4 gave the highest rate of decrease in seed germination (0%). Under greenhouse conditions the pathogenicity results indicated that the isolate Fs-1 caused the highest infection rate on Koçbaşı cultivar of chickpea plants (66.6%) followed by *Rhizctonia solani* -15 (Rs-15) and Fs-4 (56.6%), while the infection rate was 0% in FLIPO9-143C cultivar of chickpea plants by the isolate Rs-5.

KEYWORDS : Seed born fungi, Leguminous and Germination

INTRODUCTION

In a world facing problem of malnutrition, protein rich crops assume special significance. Obtaining maximum production through all available avenues and protecting adequately what is produced would certainly alleviate the problem, Legume seeds have comparatively higher protein content than non-legume plant. The high protein content makes them desirable crops in agriculture. The seeds of legumes are second only to cereals as the most important source of food for humans and animals (Rathod et al, 2012; Ghangaokar and Kshirsagar, 2013). Several leguminous crops such as *Cajanus cajan*, *Cicer arietinum*, *Dolichos lablab*, *Medicago sativa*, *Phaseolus vulgaris*, and *Vigna unguiculata* are cultivated as green manure for improving the soil fertility and reduced the amount of the expensive nitrogen fertilizer (Abdulwehab, et al, 2015).

All varieties of pulses are excellent source of easily digestible protein. But there are several factors which are responsible for their low production. Among them, diseases play an important role, Seed-borne diseases have been found to affect the growth and productivity of crop plants. Presence or absence of seed borne fungi on seed surface is one of the important aspects that determine the quality of seed. (Ghangaokar and Kshirsagar, 2013).

Many fungal pathogens, some of which are seed transmitted, often reduce the germination ability or kill the infected plants or substantially reduce the productive capacity. Some of these fungi produce aflatoxins which damage the liver and induce carcinogenic, mutagenic and teratogenesis. Therefore, control of seed-borne fungi is extremely important and the damaging effects can be relieved through integrated approaches. (Agarwal, et al, 2011).

The seeds are locally produced or imported and stored by the farmers under poor quarantine regulations and legislations. Therefore, seed contamination can occur by seed borne fungi which adversely affect the production and productivity of these crops, which may be disastrous if introduced into disease free areas (Ghangaokar and Kshirsagar, 2013; Abdulwehab, et al., 2015). Seeds play a vital role in the production of healthy crops. Healthy seed is the foundation of healthy plant; a necessary condition for good yields (Diaz, et al, 1998). Hence this study has been undertaken to investigate percentage incidence of seed-borne fungi associated with pulses of some varieties of legumes. The pathogenicity of isolated fungi was studied in the laboratory (on cabbage seeds) and greenhouse conditions (on legume seeds and seedlings).

MATERIALS AND METHODS

Collection of the seed samples

15 seed samples from 5 leguminous crop namely *Pisum sativum* (Pea) Little marvel, Jof and English cultivar produced in (2010-2011 and 2011-2012) seasons, *Vicia faba* (Faba bean) Iraqi cultivars with small and large size of grains, in addition to Egyptian and Spanish cultivars, *Cicer arietinum* (chickpea) FLIPO9-186C, FLIPO9-143C and Turkish

kokbasi cultivars, *Phaseolus vulgaris* (White bean), and *Lens culinaris* (Lentile).

All legume seed samples were collected from different localities of (Kirkuk, Mosul and Erbil) / Iraq. The samples were collected and tested as recommended by the rules of the International Seed Testing Association (ISTA 1976).

Seed Borne fungi analysis:

Detection of seed borne fungi from selected legume seeds was done by agar plate method as recommended by International Seed Testing Association ISTA (1976). The fungi which appeared on seed were isolated in pure culture for identification and for further study.

Agar plate method:

Pre sterilized petri plates were poured with 15 mL of autoclaved Potato Dextrose Agar (PDA). On cooling the medium, ten seeds per plate of the sample to be studied were equidistantly placed aseptically. 100 seeds were used in each experiment. The plates were incubated at $25 \pm 2^\circ\text{C}$ under diurnal conditions. On seventh day of incubation, seeds were first examined under stereoscopic microscope for determining the various fungal growth. The identification and further confirmation of seed borne fungi was made by preparing slides of the fungi.

Identification of isolated fungi:

The isolated fungi were identified using macroscopic features based upon colony morphology and microscopic observations of mycelia and asexual/sexual spores (Ellis, 1971; Barnett and Hunter, 1972; Pitt and Hocking, 2009). For best microscopic observation for conidia and conidiophores, discs of mycelial agar plugs (0.7 cm – diameter) from each fungus was inoculated on the center of water agar 1.5%, the inoculated petri dishes were incubated at 25°C for 7 days. Where the growth of the colonies was slow and weak and formed clear conidia, a small square piece of agar was taken with the developing fungal colony, placed on a glass slide with a few drops of lactophenol cotton blue dye. Then the cover has been put on the slide with light pressure and exposed the slide to simple heat to partially liquefy the agar with light pressure on the cover slide to get the clearest microscopic image.

Identification of *Aspergillus* species :

Three differential media: Czapek Yeast Agar (CYA), Malt Extract Agar (MEA), and Glycerol Nitrate Agar (G25%N) in three different temperature $5, 25, 37^\circ\text{C}$. Morphological features of colonies on above culture media as well as microscopically characteristics were studied (Pitt and Hocking, 2009)

Frequency of fungal isolates :

The frequency of fungal isolates was calculated according to the followed equation (Al-Abdallah, 2008):

$$\% \text{ Frequency} = \frac{\text{Total number of fungal isolates/crop}}{\text{Total number of fungal isolates}} \times 100$$

Pathogenicity tests :

Pathogenicity test on cabbage seeds in laboratory :

Three separated experiment was performed to detect the pathogenicity of isolated seed borne fungi and to select the most pathogenic fungi ,24 *Alternaria alternate* isolates and one isolate of *Rhizopus spp.* were evaluated in first experiment ,the second experiment included 29 isolates of *Rhizoctonia solani* and one isolate of *Drechslera spp.*, in addition to 9 isolates of *Fusarium spp.*,4 isolates of *Stemphylium spp.* , 2 isolates of *Cladosporium spp.* and one isolate of *Macrophomina phasiolina* . Twenty-five cabbage seeds , treated with 1% sodium hypochlorite for 1 min and rinsed with sterile distilled water , were distributed 1 cm a part on top of a 4-5 day old colony on 1.5% water agar . After the seed germinated , a sterile petri dish bottom was substituted for the original lid and taped in place to make room for the seedling . Results were recorded 6 days after the seeds placed on the colony of tested fungi . Each experiment was replicated 4 times . The inoculum for all the tests of pathogenicity was grown on Potato Dextrose Agar (PDA) medium for 5 days at 25 ± 2 °C (Bolkan and Butler,1974).

Pathogenicity test of legumes seeds fungi under greenhouse conditions :

Ten fungal pathogens *Alternaria alternate*-9 (Aa-9) , *Alternaria alternate*-14 (Aa-14), *Rhizoctonia solani*-4 (Rs-4) , *Rhizoctonia solani*-5 (Rs-5) , *Rhizoctonia solani*-11(Rs-11) , *Rhizoctonia solani*-15 (Rs-15) , *Fusarium spp.*-1(Fs-1) , *Fusarium spp.*-2(Fs-2) , *Fusarium spp.*-4 (Fs-4) , *Stemphylium spp.*-4 (S-4) were selected from preliminary test of pathogenicity by cabbage seeds which were highly pathogenic and evaluated for their pathogenicity on legumes seeds and seedlings under greenhouse conditions .

Inoculum from each of the above cultures was colonized separately on PDA. Sandy clay soil was autoclaved and transferred in to sterilized pots each containing 1 kg soil , pots were inoculated with the selected fungi growing on PDA for 7 days separately at the rate of ½ petri dish/ pot which mixed with the surface of the soil at depth 1-3 cm (Saydam, et.al.1973). Then the pots kept in greenhouse for 3 days before sowing the seeds, Pots containing non-inoculated soil were used as control. Three replicates were used per treatments .Pathogen free-seeds were surface sterilized and planted (10 seeds / pot) in both inoculated and non-inoculated soils . All pots maintained in greenhouse under natural condition during the winter season and watered as need , fifteen days after sowing the diseases ratio were determined by recording the number of non emerged seeds (pre-emergence damping-off) while post emergence damping-off and surviving plants were recording 30 days after sowing .The equation described by Khalifa (1987) were followed :

$$\text{Pre-emergence (\%) damping off} = \frac{\text{No. of non emerged seeds}}{\text{No. of sown seeds}} \times 100$$

$$\text{Post-emergence (\%) damping off} = \frac{\text{No. of killed seedling}}{\text{No. of sown seeds}} \times 100$$

$$\text{Surviving plants (\%)} = \frac{\text{No. of surviving plants}}{\text{No. of sown seeds}} \times 100$$

Re-isolation of the tested fungi were made from diseased seeds and seedlings manifesting symptoms .

RESULTS AND DISCUSSION

Fungi recovered from legume seeds

Agar plate method :

Agar plate method recommended by ITS , 1976 was used . Surface sterilized legume seeds in 1% sodium hypochlorite solution was examined to detect seed borne fungi . 15 samples of seeds were selected for use in the present investigation . 100 seeds from each sample were tested .Many fungal species belonging to 10 genera (Table, 1) were isolated and identified from the collected seed samples using Agar plate method .

Frequency of fungal isolates :

Fungi were frequently isolated from seeds (Table, 2), most frequently in Pea/English cultivar (2010-2011) 27.90% while 27.03% was isolated in Pea /Jof (2011-2012),11.04% infection in Faba bean/Spanish cultivar followed by Faba bean/Egyptian (10.17%), Pea

/little marvel(2011-2012) (7.55%),Turkish chickpea/ kocbasi (6.10%) ,Faba bean/Iraqi(large size grain) and Pea/English(2011-2012) (2.03%), Faba bean/Iraqi(small size grain) (1.74%),Chickpea / FLIPO9-143C and Chickpea/ FLIPO9-186 C (1.16%) , White bean ,Pea /Jof (2010-2011) and Pea/little marvel (2010-2011) (0.58%) and the least percentage was recorded in lentil (0.29%). The yeast was most frequently isolated (25.87%) from all crops, followed by *Aspergillus terrus* (19.18%),*A.niger* (18.31%),*A.flavus* (11.91%), *Rhizoctonia solani* (8.72%) , *Alternaria alternate* (7.55%),*Penicillium spp.* (3.19%), *Fusarium spp.* (2.61%) , *Stemphylium spp.* (1.16%), *Cladosporium spp.*(0.58%), *Rhizopus stolonifer*; *Macrophomina phasiolina* and *Drechslera spp.* (0.29%). (Table 3).

Table 1 : Seed borne fungi isolated from some legume seeds

Seed samples/cultivar	Isolated fungal species	Isolation%
Pea /Jof (2011-2012)	<i>Aspergillus niger</i>	26
	<i>A.terrus</i>	52
	<i>A.flavus</i>	6
	<i>Penicillium spp.</i>	2
	<i>Rhizoctonia solani</i>	4
	<i>Alternaria alternata</i>	3
Pea /Jof (2010-2011)	<i>Alternaria alternata</i>	1
	<i>A.terrus</i>	1
Pea /little marvel(2011-2012)	<i>Aspergillus terrus</i>	4
	<i>Rhizoctonia solani</i>	3
	<i>Alternaria alternata</i>	9
	<i>Fusarium spp.</i>	5
	<i>Stemphylium spp.</i>	3
	<i>Rhizopus stolonifer</i>	1
	<i>Drechslera spp.</i>	1
Pea /little marvel (2010-2011)	<i>Alternaria alternata</i>	1
	<i>Penicillium spp.</i>	1
Pea/English(2011-2012)	<i>Alternaria alternata</i>	3
	<i>Cladosporium spp.</i>	2
	<i>Rhizoctonia solan</i>	2
Pea/English(2010-2011)	<i>Aspergillus flavus</i>	2
	<i>A. terrus</i>	8
	Yeasts	86
Faba bean/Iraqi(large size grain)	<i>Aspergillus niger</i>	4
	<i>Rhizoctonia solani</i>	2
	<i>Stemphylium spp.</i>	1
Faba bean/Iraqi(small size grain)	<i>Aspergillus flavus</i>	1
	<i>A. niger</i>	2
	<i>Penicillium spp.</i>	3
Faba bean/Egyptian	<i>Rhizoctonia solani</i>	6
	<i>Aspergillus flavus</i>	21
	<i>A. niger</i>	6
	<i>Alternaria alternata</i>	2
Faba bean/Spanish	<i>Alternaria alternata</i>	4
	<i>Aspergillus flavus</i>	11
	<i>A. niger</i>	21
	<i>A. terrus</i>	1
	<i>Fusarium spp</i>	2
Chickpea / FLIPO9-143C	<i>Aspergillus niger</i>	1
	<i>Rhizoctonia solani</i>	1
	<i>Alternaria alternata</i>	2
Chickpea/ FLIPO9-186 C	<i>Fusarium spp.</i>	2
	<i>Penicillium spp.</i>	2
Chickpea/Koçbaşı	<i>Penicillium spp.</i>	2
	<i>Aspergillus niger</i>	3
	<i>Rhizoctonia solani</i>	12
	<i>Macrophomina phasiolina</i>	1
	Yeast	3
White bean	<i>Alternaria alternata</i>	1
	<i>Aspergillus niger</i>	1
Lenitle	<i>Penicillium spp.</i>	1

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.(Al-Abdalla,2008).

Table 3 : Total number and frequency of fungi associated with

some legume pulses

Crops	No. of fungal isolate/crop	% Frequency
Pea /Jof (2011-2012)	93	27.03
Pea /Jof (2010-2011)	2	0.58
Pea /little marvel(2011-2012)	26	7.55
Pea /little marvel (2010-2011)	2	0.58
Pea/English(2011-2012)	7	2.03
Pea/English(2010-2011)	96	27.9
Faba bean/Iraqi(large size grain)	7	2.03
Faba bean/Iraqi(small size grain)	6	1.74
Faba bean/Egyptian	35	10.17
Faba bean/Spanish	38	11.04
Chickpea / FLIPO9-143C	4	1.16
Chickpea/ FLIPO9-186 C	4	1.16
Chickpea/Koçbaşı	21	6.10
White bean	2	0.58
Lenitle	1	0.29

Table 3 : Total number and frequency of fungi isolated from some legume seeds on PDA.

Isolated fungi	Total NO. of fungal isolates	% Frequency
<i>Aspergillus niger</i>	63	18.31
<i>A. terrus</i>	66	19.18
<i>Penicillium spp.</i>	11	3.19
<i>Rhizoctonia solani</i>	30	8.72
<i>Rhizopus stolonifer</i>	1	0.29
<i>Alternaria alternata</i>	26	7.55
<i>Aspergillus flavus</i>	41	11.91
<i>Drechslera spp.</i>	1	0.29
<i>Stemphylium spp.</i>	4	1.16
<i>Fusarium spp.</i>	9	2.61
<i>Cladsporium spp.</i>	2	0.58
<i>Macrophomina phasiolina</i>	1	0.29
Yeasts	89	25.87

Different authors isolate related species of fungi from legumes seeds :

Nik and Yap (1979) isolated *Aspergillus spp.*, *Fusarium spp.*, *Rhizoctonia solani*, *Rhizopus stolonifer* and *Penicillium spp.* from French bean seeds ; Al-Abdallal (2008) isolated *Aspergillus niger*, *A.flavus*, *Rhizoctonia solani*, *Alternaria alternata*, *Penicillium spp.*, *Rhizopus stolonifer* and *Fusarium spp.* from Lupine, Cowpea, Mung beans, Faba beans, Brown and Green lentil ; Sarhan (2009) isolated *Alternaria spp.*, *Aspergillus spp.*, *Fusarium spp.*, *Penicillium spp.*, *Rhizoctonia spp.*, *Stemphylium spp.* from white bean, faba bean, chickpea, mung bean and pea seeds ; Al-Abdallal(2010) isolated *Rhizoctonia solani* from Mung beans, Faba beans, Brown and Green lentil, Lupine and chickpea seeds ; Agarwal *et al* (2011) isolated *Alternaria alternata*, *Aspergillus niger*, *A.flavus*, *Fusarium oxysporum*, *Penicillium spp.*, *Rhizopus spp.* from chickpea and black gram seeds ; Rathod *et al.* (2012) isolated *Aspergillus niger*, *A.flavus*, *Fusarium oxysporum*, *Penicillium spp.*, *Macrophomina phasiolina*, *Rhizoctonia solani* from groundnut seeds; Ghangaokar and Kshirsagar(2013) isolated *Aspergillus niger*, *A.flavus*, *Fusarium solani*, *Penicillium spp.*, *Macrophomina phasiolina*, *Alternaria alternata*, *Cladsporium*, *Drechslera spp* ; from pea, lentil, French bean, chickpea, Cowpea and pigeon peas seeds ; Ivic (2014) isolated *Fusarium solani* from soybean seeds ; Wilman,*et.al.*(2014) isolated *Alternaria*, *Fusarium*, *Stemphylium* from pea seeds ; Trivedi and Rathi (2015) isolated *Fusarium oxysporum*, *Penicillium spp.*, *Rhizopus spp.*, *Aspergillus niger* from chickpea seeds .

Identification of Aspergillus species :

Aspergillus isolate was identified as *Aspergillus terrus* according to Morphological features of colonies on CYA, MEA, and G25%N in three different temperature 5, 25, 37°C which described by Pitt and Hocking (2009).

Pathogenicity tests :

Pathogenicity test on cabbage seeds in laboratory conditions

Three separated experiments were performed to evaluate the preliminary pathogenicity of 71 fungal isolates at laboratory conditions on 1.5% water agar petri-dishes by using cabbage seeds, the results indicated that all isolates caused a significant reduction in

the rate of cabbage seeds germination but only ten isolates were highly pathogenic, the percentage of germination in their treatments were 0-15% compared to 97% in control treatments, the isolates of *Fusarium spp.* which include Fs-1, Fs-2 and Fs-4 gave the highest rate of decrease in seed germination (0%), followed by Aa-9, Aa-14, Rs-4, Rs-5, Rs-11, Rs-15 and S1 isolates which reduce the cabbage seeds germination significantly at rate 5-15 %, also the other isolates affected significantly at reduction rate 41-95% (Table.4). Cabbage seeds are highly sensitive to pathogens and for this reason used in this test .The variation in virulence among fungal isolates may be due to genetic variation specially they are isolated from different seeds and seasons, as well as the specific environmental conditions in each seed store, pectinase and cellulose production result in seed rot and prevent the germination (Hussein and Juber, 2015). This results are in agreement with the results of several researchers (Hussein and Juber, 2015) which referred that the *F.solani* isolates (D5FS-2 and T4FS-15) the causes of root rot diseases in watermelon prevent the cabbage seed germination completely ; Matlob and Juber (2012) finding that the isolates of *Macrophomina. phaseolina* were differed in their effect on cabbage seed germination. The isolates MP-2B, MP-3, MP-5 prevented seeds germination completely, compared with control (93.33 %) ; Juber and Saeed (2015) studied the preliminary pathogenicity of *Fusarium spp.* species which associated with zinnia seeds and indicated that all the 22 isolates caused a significant reduction in the rate of cabbage seeds germination but only seven isolates were highly pathogenic, the percentage of germination in their treatments were 0-20% compared to 97 and 98% in control treatments. *F.culmorum* isolate (A33) gave the highest rate of decrease in seed germination (0%).

Table4: Pathogenicity testing of some legume seeds-borne fungi on cabbage seeds under laboratory conditions

% Germination	Seed samples	Fungal isolates	% Germination	Seed samples	Fungal isolates
47	Pea /Jof	Aa-15	97		Control
46	Pea /Jof	Aa-16	0	Chickpea/kocbasi	Fs-1
71	Pea /Jof	Aa-17	0	Faba bean/Spanish	Fs-2
68	Pea/English	Aa-18	45	Pea /little marvel	Fs-3
77	Pea/English	Aa-19	0	Pea /little marvel	Fs-4
73	Pea/English	Aa-20	55	Pea /little marvel	Fs-5
88	Faba bean/Spanish	Aa-21	72	Pea /little marvel	Fs-6
95	Faba bean/Spanish	Aa-22	50	Pea /little marvel	Fs-7
92	Faba bean/Spanish	Aa-23	52	Faba bean/Spanish	Fs-8
85	Faba bean/Spanish	Aa-24	68	Chickpea/ FLIPO9-186 C	Fs-9
88	Chickpea/ FLIPO9-186 C	Aa-25	62	Chickpea/ FLIPO9-186 C	Fs-10
92	Chickpea/ FLIPO9-186 C	Aa-26	66	Chickpea/kocbasi	Fs-11
59	Pea /Jof	Rs-1	75	Chickpea/Kocbasi	Fs-12
81	Pea /Jof	Rs-2	50	Pea /little marvel	Aa-1
70	Pea /Jof	Rs-3	61	Pea /little marvel	Aa-2
7	Pea /Jof	Rs-4	57	Pea /little marvel	Aa-3
13	Chickpea/ FLIPO9-186 C	Rs-5	59	Pea /little marvel	Aa-4
70	Faba bean/Iraqi(large size grain)	Rs-6	54	Pea /little marvel	Aa-5
75	Faba bean/Iraqi(large size grain)	Rs-7	71	Pea /little marvel	Aa-6
42	Chickpea/Kocbasi	Rs-8	44	Pea /little marvel	Aa-7
53	Chickpea/Kocbasi	Rs-9	48	Pea /little marvel	Aa-8
67	Chickpea/kocbasi	Rs-10	13	Faba bean/Egyptian	Aa-9
5	Pea/English	Rs-11	41	Faba bean/Egyptian	Aa-10
74	Pea/English	Rs-12	75	Pea /little marvel	Aa-11
77	Pea /little marvel	Rs-13	66	Pea /little marvel	Aa-12
76	Pea /little marvel	Rs-14	60	White bean	Aa-13
10	Pea /little marvel	Rs-15	11	Pea /Jof	Aa-14

Table 5 : Pathogenicity testing of some legume seeds-borne fungi on cabbage seeds under laboratory conditions

%Germination	Seed samples	Fungal isolates	%Germination	Seed samples	Fungal isolates
62	Chickpea/Kocbasi	Rs-27	72	Faba bean/Egyptian	Rs-16
55	Chickpea/Kocbasi	Rs-28	61	Faba bean/Egyptian	Rs-17
75	Chickpea/Kocbasi	Rs-29	74	Faba bean/Egyptian	Rs-18
47	Faba bean/Iraqi(large size grain)	S1	53	Faba bean/Egyptian	Rs-19
67	Pea /little marvel	S2	56	Faba bean/Egyptian	Rs-20
58	Pea /little marvel	S3	50	Faba bean/Egyptian	Rs-21
15	Pea /little marvel	S4	71	Chickpea/Kocbasi	Rs-22
64	Pea /little marvel	Ds1	55	Chickpea/Kocbasi	Rs-23
90	Pea/English	Cld1	62	Chickpea/Kocbasi	Rs-24
95	Pea/English	Cld2	58	Chickpea/Kocbasi	Rs-25
67	Chickpea/kocbasi	Mac	60	Chickpea/Kocbasi	Rs-26

Pathogenicity testing of some pathogenic seed borne fungi of some legume seeds under greenhouse conditions

Results of the pathogenicity tests of Aa-9, Aa-14, Rs-4, Rs-5, Rs-11, Rs-15, Fs-1, Fs-2, Fs-4, S-4 are shown in Table (5). Diseases severity reading of the pathogens on seeds and seedlings were made after 15 and 30 days after planting for pre-emergence damping off , post-emergence damping off and surviving seedlings .Data in Table 5 . shows that Rs-11 had the highest percentage of pre-emergence damping off among all isolated fungi . The percentage of infection was 36.6% on seed compared with the control treatment , which had 10% infection . Fs-1 caused the highest percentage of post emergence damping off among all the pathogens . The recorded percentage was 33.3 % on seedlings .

The pathogenicity tests showed that the most pathogenic fungi isolated from legume seeds which showed seed decay , pre-emergence damping off , post-emergence damping off but in various degree .

Fungi used plant sugars for growth and respiration which affect carbohydrate content of seeds leaving them irregular and abnormal beside the weakness of the growing seedlings (Wildermuth *et al.* ,1992) .

Rhizoctonia solani produce pectinase and cellulose Which lead to seed rot and seedling damping off , as well as Some plant toxic compounds, such as Phenyl Acetic Acid which cause the killing of seed embryos (Rush *et al.* , 1994; Weinholt and Sinsclair, 1996).

Seed borne fungi produce toxic chemicals that affect seeds and enzymes , which cause significant damage to the vitality of seeds, especially the fungi found within the seed, which reduces the germination rates , because it leads to the weakening or killing of the embryo and rotting seeds when suitable conditions are available (Sarhan, 2009).

Difference in susceptibility to infection could also be attributed to be difference in the genetic structure of each seed, different secretion from the roots of these varieties may also play some role in protecting plants from fungal infection (Abdallal 2008) .

Table 5 : Pathogenicity testing of some pathogenic seed borne fungi of some legume seeds under greenhouse conditions.

Legumes cultivars	Treated fungi	%Infected plants			% Surviving plants
		Pre - emergence	Post - emergence	Total	
Pea /little marvel	Rs-15	26.6	30	56.6	43.3
Pea /Jof	Rs-4	23.3	26.6	50	50
Chickpea / FLIPO9-143C	Rs-5	0	0	0	100
Pea/English	Rs-11	36.6	13.3	50	50
Faba bean/Spanish	Fs-2	3.3	6.6	10	90
Pea /little marvel	Fs-2	33.3	23.3	56.6	43.3
Chickpea/Koçbaşı	Fs-1	33.3	33.3	66.6	33.3
Pea /Jof	Aa-14	20	10	30	70
Faba bean/Egyptian	Aa-9	6.66	13.3	20	80

Faba bean/Iraqi(large size grain)	S-4	13.3	0	13.3	86.6
Pea /little marvel	Rs-15	0	0	0	100
Pea /Jof	Rs-4	10	0	10	90
Chickpea / FLIPO9-143C	Rs-5	0	0	0	100
Pea/English	Rs-11	10	0	10	90
Faba bean/Spanish	Fs-2	0	0	0	100
Pea /little marvel	Fs-2	0	0	0	100
Chickpea/Koçbaşı	Fs-1	3.3	0	3.3	96.6
Pea /Jof	Aa-14	10	0	10	90

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