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Pathological Studies on Root Rots of Wheat and Barley Induced by *Rhizoctonia solani* and *Fusarium culmorum* and their Effects on the Crop

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ABSTRACT

The objective of this study was to investigate the effect of *Rhizoctonia solani* and *Fusarium culmorum*, which caused root rot, on the growth and yield of wheat and barley plants. Wheat cv. 'Yecorarogo' and barley cv. 'Gustuo' were planted in soil artificially infected with *R. solani* and *F. culmorum*. There was a significant decrease in the growth and yield of infected plants compared with healthy plants, in particular the length, fresh and dry weight of roots and shoots, and the number of grains for each spike. Both fungi damaged wheat plants more than barley plants. *R. solani* had the greatest effect on the growth and yield of wheat and barley. The content of biochemical compounds also decreased in all cases of infection. Total carbohydrate, proteins and fats decreased in the range between 23.28-45.68, 0.55-26.22 and 28.45-59.94%, respectively.

Keywords: *Hordeum vulgare*, *Triticum aestivum*

INTRODUCTION

Wheat (*Triticum aestivum*) is one of the most important cereal crops as most people consider it as a principle food due to its high carbohydrate, protein, lipid, vitamin and mineral contents. Humans use every part of wheat plants, even the seed coat. Barley (*Hordeum vulgare*) is also used as a food rich in fibers and minerals; it is frequently used as food for domestic animals.

Great economic losses are recorded as a result of fungal infection of the crop, in particular those causing root rots; wheat and barley are the most affected. Field infection results in the complete eradication of the plant in many areas due to necrosis, the result of rotten roots.

Wheat may be infected by more than one species of *Fusarium* (Smiley and Patterson 1996). Smiley *et al*. (2005a) recorded a great loss in a wheat crop after infection by *F. culmorum* and *F. pseudograminearum* in the North West Pacific.

Al-Abdalall-Amira (1998) showed that the fungi *Rhizoctonia solani* and *F. culmorum* had the highest infection rate in wheat cv. 'Yecorarogo' and barley cv. 'Gustuo', causing rotten roots, and hence their pathogenicity. This study takes those earlier results further by examining the effects of this infection on the plant vegetative parts, root growth, plant weight, grain yield, and carbohydrate, protein and lipid contents.

MATERIALS AND METHODS

Source of isolates

R. solani and *F. culmorum* isolates had the highest infection rate in wheat 'Yecorarogo' and barley 'Gustuo', respectively, causing rotten roots, a result also found in a previous study by Al-Abdalall-Amira (1998). The isolates were identified at the Fungal Taxonomy Department, Plant Pathology Research Institute, Agricultural Research Center, Giza, Egypt and isolated from the rotting roots of these two cultivars.

Soil inoculation was prepared by using barley medium (200 g of barley kernels, 200 ml water and 60 g sand) in 0.5 L glass bottles. Bottles with medium were autoclaved at 121°C at 1.5 kg/cm^2 for 60 min (Abada 1986). Autoclaved barley medium was inoculated using PDA slants from each of the isolated fungi, and incubated at 25°C for 15 days. Sandy soil was autoclaved at 121°C at 1.5 kg/cm^2 for 3 h. Barley inoculum was added to the sterilized soil at a rate of 1% (w/w), mixed and rubbed together to release mycelium and spores. Pots (ϕ = 25 cm) were immersed in a 5% formalin solution for 15 min and left for 2 days to dry and for formalin to evaporate. Pots were then partially filled with soil (6 kg/pot) and infested by each tested fungi separately. A set of pots containing sterilized soil only served as the control (Elian 1978).

Wheat and barley grains obtained from a farm of Eastern Co. in Fadly (Saudi Arabia) were surface sterilized by immersing them in 2% sodium hypochlorite for 2 min, then washing 3 times with sterilized distilled water. The grains were sown on the surface at a rate of 15 grains/pot and had sufficient space between them (1 Qatar = 25 cm). Three replicates were used for each treatment (El-Awadi 1993). The percentage of pre-, post-emergence damping-off, root rot and healthy seedlings was recorded.

The experiment was repeated 4 times for each fungal species, with the control experiment free from fungal contamination (Elian 1978).

Measurements included root length, dry and wet weight of roots and shoots, number of leaves, grain yield (number, dry and wet weight) of infected and control experiments. Ground grains of wheat and barley were used to estimate carbohydrate, total protein and total lipid contents.

Determination total soluble sugar content

Sample preparation: 5 g of wheat or barley grains (infected and healthy) were ground in 25 ml of 70% boiling ethanol for about 10 min, then filtered twice. The filtrate was diluted to 100 ml with 70% ethanol, then the volume was adjusted to 100 ml with 70% ethanol. The filtrate was used to estimate total carbohydrates according to the method of AOAC (Anonymous 1965). A standard curve for glucose was designed according to Aly (1972).

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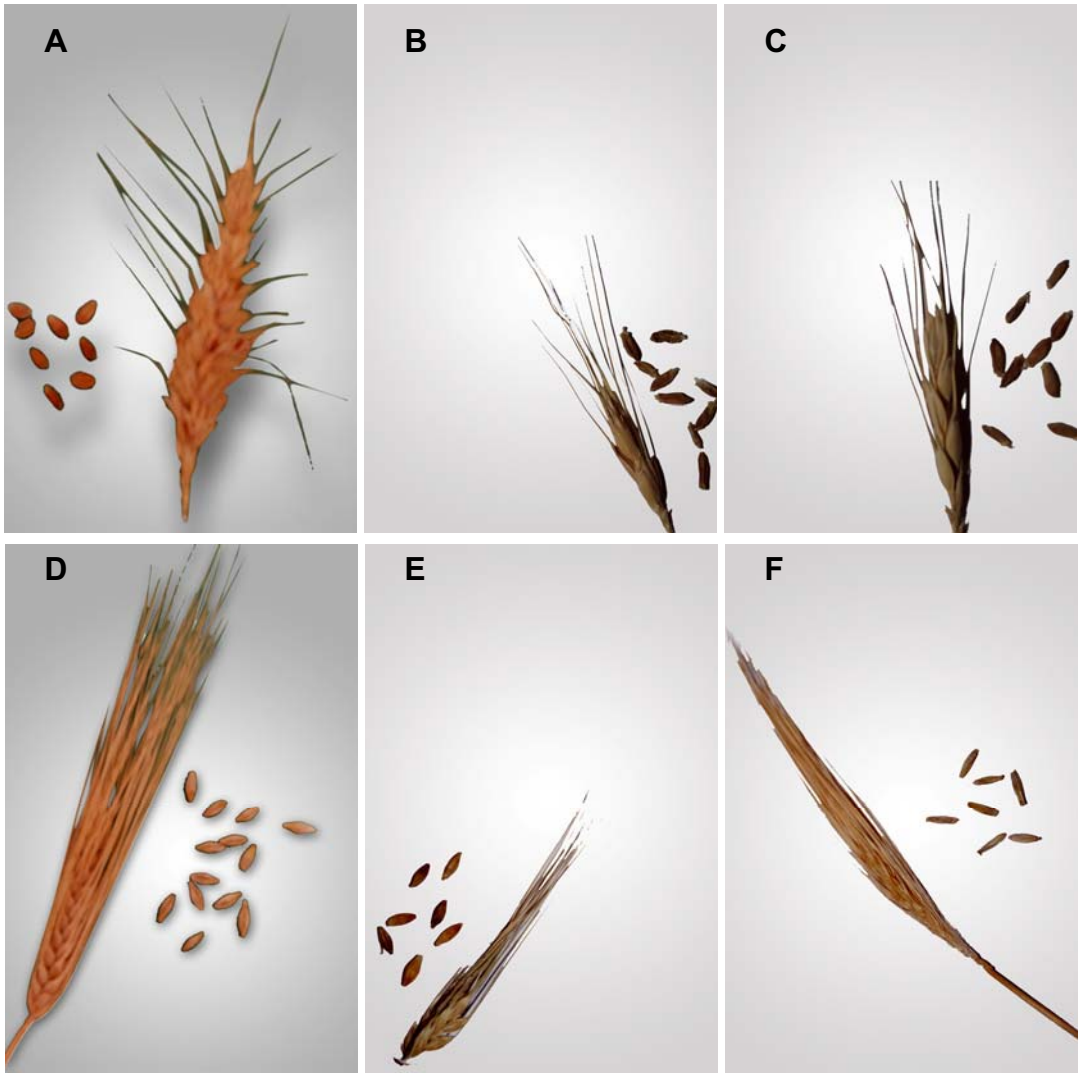

Fig. 1 Effect of artificial inoculation with *R. solani* and *F. culmorum* on wheat (A-C) and barley (D-F) crops. (**A**) Healthy wheat with well grown grains. (**B**) Spike wheat infected with *R. solani* wrinkled and small grains. (**C**) Spike wheat infected with *F. culmorum* small and wrinkled grains. Effect of artificial inoculation with *R. solani* and *F. culmorum* on barley crops. (**D**) Healthy barley with well grown grains. (**E**) Spike barley infected with *R. solani* wrinkled and small grains. (**F**) Spike barley infected with *F. culmorum* small and wrinkled grains.

Determination of crude protein

The methods of Meidner (1984) and Lowry *et al*. (1951) were used.

Determination of total lipids

The method of Bligh and Dyer (1959) was used to estimate the total lipid in seeds of both healthy and infected plants.

Statistical analyses

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

Effect of artificial inoculation with two tested fungi on wheat and barley crop

Laboratory infection of both cereal crops by fungi reduced root length, dry root and shoot weight, shoot length, the average number of leaves, the number of grains/spike, i.e. yield and grain dry weight significantly (**Table 1, Fig. 1A-F**). The root length of control barley plant was 44.66 which was reduced to 12.89 and 15.68 cm after infection by *R. solani* and *F. culmorum*, respectively.

The weight of the dry root content was greatly affected and reduced from 0.25 g in untreated wheat plants to 0.05 and 0.06 g after infection by *R. solani* and *F. culmorum*, respectively. The same result was recorded for barley plants whose root dry weight was 0.26 g which became reduced to 0.05 and 0.08 g after infection by the same two fungi.

The length of shoot system was also affected. Untreated wheat plants were 55.3 cm long which became reduced to 28.52 cm after infection by *F. culmorum* and 26.85 cm after infection by *R. solani*. Infection also affected the height of barley plants, reducing them from 50.65 cm (in healthy plants) to 34.95 and 33.35 cm after infection by *R. solani* and *F. culmorum*, respectively. The dry weight of the shoot system of wheat plants showed a similar reduction from 1.3 g in healthy plants to 0.37 g in *F. culmorum*-infected and 0.23 g in *R. solani*-infected plants; in barley these three equivalent values were 1.09, 0.68 and 0.81 g, respectively.

The average number of leaves was also reduced by fungal infection. Normal wheat and barley plants had 9 leaves but after infection with *F. culmorum* this was reduced to 6.35 and 7 for wheat and barley and after infection with *R. solani* this was reduced to 6.96 and 7.6.

Fungal infection also reduced grain yield from 56 grains/spike to 15.17 and 21.1 grains/spike after *R. solani* and *F. culmorum* infection. Infection also affected the grain yield of the barley crop, with 27.5 and 27.7 grains/spike following the infection by these two fungi compared to 53 grains/spike in healthy plants. Grains of pre-infected plants

Table 1 Effect of artificial inoculation with *R. solani* and *F. culmorum* on wheat and barley crops.

Crop	Treatment	Part of plant									
		Root system			Shoot system			№ leaves/ plant	Grains		
		Length (cm)	Fresh weight (g)	Dry weight (g)	Length (cm)	Fresh weight (g)	Dry weight (g)		№ spikes	Fresh weight (g)	Dry weight (g)
Wheat	Control	36.24	28	0.25	55.3	1.38	1.3	9	56	1.65	1.86
	R. solani	4.75	0.05	0.05	26.85	0.26	023	6.92	15.17	0.31	0.28
	% + or -	86.89	99.82	80	51.45	81.16	82.31	23.11	72.91	81.21	84.95
	F. culmorum	16.12	0.07	0.06	28.52	0.38	0.37	6.35	21.1	0.37	0.35
	% + or -	55.52	99.75	76	48.43	72.46	71.54	29.44	62.32	77.58	81.18
Barley	Control	44.06	0.29	0.26	50.65	1.14	1.09	9	53	1.62	1.55
	R. solani	12.89	0.065	0.05	34.95	0.73	0.68	7.9	27.5	0.395	0.375
	% + or -	70.74	77.59	80.77	30.99	35.97	37.61	15.56	48.11	75.62	75.81
	F. culmorum	15.68	0.08	0.08	33.35	0.91	0.81	7	27.5	0.43	0.42
	% + or -	64.41	73.41	69.23	34.16	20.18	25.69	22.22	48.11	73.46	72.9
L.S.D. at (0.0 5)		1.779	0.729	0.033	1.78	1.044	1.037	1.779	1.78	1.04	1.04

% + or - = percentage increase or decrease

were less volumous than healthy ones (**Fig. 1**).

Comparing the grain dry weight, healthy wheat grains had 1.86 g, which was reduced to 0.35 and 0.28 g after *R. solani* and *F. culmorum* infection, respectively. Barley grain dry weight was 1.55 g which became 0.42 g after *F. culmorum* infection and 0.375 g after *R. solani* infection.

Similar results (i.e. a decrease in crop-related parameters) were obtained by Pakhomova (1969), Chulkina (1971), Sidorov (1990), Wildermuth *et al*. (1992), Al-Abdalall-Amira (1998), Paulitz *et al*. (2002), Smith *et al*. (2003a) and Smiley *et al*. (2005a).

Effect of artificial inoculation with tested fungi on biochemical compounds of wheat and barley grains

There were significant differences in the quantity of carbohydrates, soluble sugars, proteins and lipids between grains of healthy and fungal-infected plants (**Table 2**).

Soluble sugars of healthy wheat grains were 59.83 mg/ 10 g flour, but greatly reduced to 45.67 and 43.17 mg/10 g flour after infection by *R. solani* and *F. culmorum*, respectively. A similar reduction was recorded for barley grains: from 68 mg/10 g flour in healthy plants to 51.78 and 52.17 mg/10 g flour after infection by the two fungi.

Total protein content in healthy wheat grains was estimated to be 13.26%. After infection by *R. solani*, it was reduced to 10.20% and to 12.21% after infection by *F. culmorum*, a reduction percentage of 23.10 and 7.92, respectively. Protein content of barley grains was estimated to be 13.31%, greatly affected by infection with *R. solani* (9.82%) but only slightly so (13.24%) after *F. culmorum* infection, equivalent to a 26.22 and 0.55% reduction.

The total lipids were also reduced following infection, from 3.18 to 2.27 and 1.27% in wheat after the infection with *R. solani* and *F. culmorum*, respectively. This was equivalent to a 28.7 and 60.01% reduction. Barley grains were similarly affected with the lipid content changing from 2.80 to 1.80 and 1.74%, respectively. This was equivalent to a 35.13 and 37.18% reduction.

Fungi consume sugar during growth and activity. Plants use also sugar to build their component parts and to create energy which leads to a consumption of sugar, shrinking grains and leaving them wrinkled. Moreover this causes a reduction in crop product and weaker vegetative parts, as attested by Younes (1977), Shalaby (1988), Wildermuth *et al*. (1992) and Al-Abdalall-Amira (1998).

Infected plants had significantly reduced total protein and lipid contents, which were consumed by fungi. Similar results were obtained by Pukhal'skii *et al*. (1986), Gubanova and Grigorev (1988), Troshina and Yamaleev (1990, 1991), Smiley and Uddin (1993), Al-Abdalall-Amira (1998), Smith *et al*. (2003) and Smiley *et al*. (2005b).

Generally, fungal infection leads to rotting roots which leads to the inability of a plant to grow, especially at early stages of growth. Shrinkage of grains is an important sign, with reduced food content leading to crop loss. These decreasing trends and conclusions demonstrated by this study were similar to those by Sidorov (1990), Wildermuth *et al*. (1992), Lucas *et al*. (1993), Smith *et al*. (2003b).

Table 2 Effect of artificial inoculation with tested fungi on biochemical compounds of wheat and barley grains.

Crop	Treatment	Total soluble sugars	Crude protein %	Total lipids %
Wheat	Control	59.83	13.26	3.18
	R. solani	45.67	10.20	2.27
	% + or -	-23.67	-23.10	-28.45
	F. culmorum	43.17	12.21	1.27
	% + or -	-27.85	-7.92	-59.94
Barley	Control	68	13.31	2.80
	R. solani	52.17	9.82	1.797
	% + or -	-23.28	-26.22	-35.13
	F. culmorum	51.78	13.24	1.74
	% + or -	-23.85	-0.55	-37.18
L.S.D. at (0.05)		1.72	0.049	0.734

% + or - = percentage increase or decrease

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