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WATER USE AND PHOSPHORUS AND POTASSIUM STATUS OF WHEAT SEEDLINGS COLONIZED BY *GAEUMANNOMYCES GRAMINIS* OR *PHIALOPHORA RADICICOLA*

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KEY WORDS

Gaeumannomyces Phialophora Phosphorus Potassium Water use Wheat

SUMMARY

The water consumption and levels of phosphorus, potassium, and total minerals were measured for wheat seedlings colonized by *Gaeumannomyces graminis* var. *tritici*, *Phialophora radicicola* var. *radicicola*, or *Phialophora radicicola* var. *graminicola*. Infection by *G. graminis* resulted in a considerable reduction in water consumption, and reduced level of phosphorus when the supply of phosphorus to the seedlings was plentiful. Colonization by *P. radicicola* var. *radicicola* increased levels of phosphorus and potassium, but these increases varied according to the isolate of the fungus and the supply of phosphorus and potassium available to the seedlings. Colonization by *P. radicicola* var. *graminicola* resulted in reduced water consumption by the seedlings.

The results are discussed in relation to stelar cell wall thickening in wheat roots colonized by *P. radicicola*, and the effects on nutrient uptake of mycorrhizal root systems.

INTRODUCTION

Phialophora radicicola (Cain) var. *radicicola* (Deacon), and *Phialophora radicicola* (Cain) var. *graminicola* (Deacon), are known to be ectotrophic parasites of wheat roots[8]. *P. radicicola* has an infection habit closely resembling that of *Gaeumannomyces graminis* (Sacc.) Arx and Olivier var. *tritici* (Walker), and spreads over the root surface by means of dark-coloured runner hyphae[4], branches of which penetrate the root cortex without causing necrosis or any reduction in the growth of the host plant[1]. It has recently been found[9] that wheat seedling roots colonized by *P. radicicola* var. *radicicola* or by *P. radicicola* var. *graminicola* show a characteristic thickening of the inner tangential walls of the endodermis and the walls of the xylem vessels. There was evidence that the endodermal wall thickening was of extra suberin deposition and that the xylem wall thickening was of extra lignin deposition. Since the endodermis must be traversed by the flow of water and nutrients from the cortex into the xylem, it is possible that extra suberization at this site, and lignification of the xylem cell walls, may restrict the flow to some degree.

Contrary to this effect, it is also possible that the presence of a rhizosphere fungus which penetrates the root cortex may enhance the uptake of nutrients, especially phosphorus, in the same way as mycorrhizal systems[5,6,7]. The resultant effect, an increase or a decrease in water or nutrient uptake, will depend on which of the two primary effects is the greater. The purpose of the present study was to discover whether wheat seedlings colonized by *P. radicicola* take up greater or lesser amounts of water, phosphorus and potassium, compared with seedlings infected by *G. graminis* or uninfected plants.

EXPERIMENTAL

Wheat seedlings were grown in plastic pots 75 mm in diameter and 75 mm deep, with glass-fibre papers covering the drainage holes. Each pot contained 200 g acid-washed white silica sand, 25 g peat (air-dried and passed through a 1 mm mesh sieve), and 25 ml nutrient solution containing $NH_4^+NO_3^-$ (25 m*M*), $Ca^{2+}(Cl^-)_2$ (25 m*M*) and $Mg^{2+}SO_4^{2-}$ (5 m*M*). Four nutrient treatments, (a) deficient in both phosphorus and potassium, (b) deficient in phosphorus only, (c) deficient in potassium only and (d) without these deficiencies, were imposed by adding Na^+Cl^- (50 m*M*) to (a), K^+Cl^- (50 m*M*) to (b), $(Na^+)_2H^+PO_4^{3-}$ (25 m*M*) to (c), and $(K^+)_2H^+PO_4^{3-}$ (25 m*M*) to (d). Within each nutrient treatment, four replicate pots were inoculated with a 2 per cent admixture of a sand and 3 per cent maizemeal culture of one of the following: sterile culture (uninoculated controls); *G. graminis* var. *tritici*; *P. radicicola* var. *radicicola* isolate A; *P. radicicola* var. *radicicola* isolate B; *P. radicicola* var. *graminicola* isolate A; *P. radicicola* var. *graminicola* isolate B.

Wheat seed cv. Maris Freeman were pregerminated in shallow water for 48 hours at room temperature and then planted, five per pot, about 5 mm below the sand and peat surface. The seedlings were allowed to grow for four weeks in a growth room at 19 ± 1°C, with relative humidity varying from about 60 per cent to a controlled upper limit of 85 per cent. Lighting from 'Warm White' fluorescent tubes provided 70 $js^{-1}m^{-2}$ visible radiation with a 16 h light: 8 h dark cycle. The pots were weighed daily and the original weight was restored by adding the appropriate nutrient solution. Six pots with neither inoculum nor wheat seeds were included in the experiment to determine the daily weight loss due to the evaporation of water from the soil surface. The difference between this value (as the mean of six replicates) and the weight loss of the other pots containing the seedlings, was taken as a measure of transpiration.

The green shoots of the plants from each pot were harvested, dried at 80°C, weighed and ashed at 450°C overnight. Ash samples were then weighed, taken up in hydrochloric acid, and the phosphorus content determined by the method of Cavell[2]; the potassium content was measured by flame photometry. Water consumption, dry weight and ash weight data were analysed by a two-dimensional analysis of variance. The data representing levels of phosphorus and potassium in the plants were divided into two for the purpose of statistical analysis since there was a very considerable difference between results obtained in the deficient and non-deficient treatments; this difference might obscure the effects of the inoculation treatments if data from the deficient and non-deficient nutrient treatments were analysed together.

RESULTS AND DISCUSSION

Table 1 gives the quantities of water consumed by the wheat seedlings during four weeks' growth, per unit dry weight of the shoot.

P. radicicola, the organism found to cause stelar and endodermal cell wall thickening when it colonizes wheat roots[9], brought about changes in the water consumption, and also the levels of phosphorus and potassium (Tables 3 and 4). Colonization by isolate B of *P. radicicola* var. *radicicola* increased the phosphorus level by about 40 per cent when phosphorus was deficient, and by about 10 per cent when phosphorus was plentiful. Plants inoculated with this isolate also yielded a greater percentage of ash compared with uninoculated plants, but only when the growth medium was deficient in phosphorus.

Both isolates of *P. radicicola* var. *graminocola* had the effect of reducing water use by the seedlings. Colonization by isolate A of this fungus generally brought about increased shoot dry weights, but statistical significance was only reached in the low phosphorus treatment (Table 2).

Table 1. Water consumed (ml/g^{-1} shoot dry weight) during 4 weeks growth by wheat seedlings colonized by *G. graminis* or *P. radicicola*, and grown in media with or without added phosphorus (+P and −P), and with or without added potassium (+K and −K)

Inoculation treatment	−K treatments			+K treatments		Means
	−P	+P		−P	+P	
Uninoculated	420	519		258	435	408
G. graminis var. *tritici*	312	66		144	87	152
P. radicicola var. *radicola*						
Isolate A	327	363		243	354	322
Isolate B	372	345		255	363	334
P. radicicola var. *graminicola*						
Isolate A	333	357		231	336	314
Isolate B	312	363		210	294	295
Means	346	336		224	312	
LSD			75			90

Figures in columns 1, 2, 3, 4 are means of 4 replicates.
LSD = Least significant difference ($p = 0.05$).

Table 2. Dry weight (mg) of the shoots of wheat seedlings colonized by *G. graminis* or *P. radicicola*, and grown in media with or without added phosphorus (+P and −P), and with or without added potassium (+K and −K)

Inoculation treatment	−K treatments		+K treatments		LSD
	−P	+P	−P	+P	
Uninoculated	54.6	52.9	75.6	70.9	11.2
G. graminis var. *tritici*	38.3	30.8	44.0	50.3	9.8
P. radicicola var. *radicicola*					
Isolate A	56.3	50.2	84.0	64.9	11.2
Isolate B	56.8	52.8	71.6	73.1	12.6
P. radicicola var. *graminicola*					
Isolate A	60.4	57.3	92.3	73.4	13.3
Isolate B	57.2	51.1	72.2	69.9	6.0
LSD	6.2	9.1	12.0	12.7	

Figures are means of 4 replicates. LSD = Least significant difference ($p = 0.05$).

Infection by *G. graminis* var. *tritici* resulted in considerable reductions in water use, dry matter production, and phosphorus levels, especially when the medium was high in phosphorus. These findings were consistent with the well known ability of this fungus to cause necrosis and occlude the vascular tissues of the root[3]. There was however, an increase in the level of potassium of about 38 per cent in the potassium deficient treatments, in seedlings infected by *G. graminis* var. *tritici.*

No overall conclusion could be drawn about the effects of colonization by *P. radicicola.* The interactions that emerged from the analysis of the data suggested firstly, that different isolates of this fungus may be expected to bring about different effects on phosphorus and potassium levels in the seedlings and secondly, that these effects are likely to be further modified by the nutrient status of the growth medium. It has been shown that colonization by isolates of *P. radicicola* var. *radicicola* could bring about important increases in the levels of potassium and phosphorus and therefore colonization by fungi belonging to this group could be beneficial under conditions of low nutrient status.

Levels of total minerals, as represented by the percentage yield of ash from the

Table 3. Phosphorus content (per cent shoot dry weight) of the shoots of wheat seedlings colonized by *G. graminis* or *P. radicicola*, and grown in media with or without added phosphorus (+P and −P), and with or without added potassium (+K and −K)

Inoculation treatment	−P treatments			+P treatments		
	−K	+K	Means	−K	+K	Means
Uninoculated	0.18	0.12	0.15	1.70	1.19	1.45
G. graminis var. *tritici*	0.20	0.14	0.17	1.39	0.84	1.12
P. radicicola var. *radicicola*						
Isolate A	0.16	0.12	0.14	1.69	1.32	1.51
Isolate B	0.25	0.17	0.21	1.80	1.37	1.59
P. radicicola var. *graminicola*						
Isolate A	0.16	0.11	0.14	1.58	1.17	1.38
Isolate B	0.13	0.09	0.11	1.65	1.20	1.43
Means	0.18*	0.13*		1.64**	1.18**	
LSD			0.03			0.12

Figures in columns 1, 2, 4, 5 are means of 4 replicates.
LSD = Least significant difference ($p = 0.05$).
* Figures significantly different ($p < 0.01$).
** Figures significantly different ($p < 0.001$).

shoots of the seedlings (Table 5), were considerably lower in plants exposed to phosphorus deficiency than in plants supplied with plentiful phosphorus. When the data in Table 3 and Table 4 are calculated in terms of the oxides of phosphorus and potassium which would appear in the ash, the amounts of these oxides can be subtracted from the total ash yields to evaluate the levels of minerals other than phosphorus and potassium. No statistical differences could be established between the effects of any of the treatments on the levels of minerals other than phosphorus and potassium. The effects of the treatments on the percentage yields of ash could therefore be accounted for by the changes brought about in the phosphorus and potassium levels, which together contributed a large proportion of the total ash percentage; the mean yield of ash from minerals other than phosphorus and potassium was 4.16 ± 0.11 (SE) per cent shoot dry weight. The reduced ash yields brought about by phosphorus de-

Table 4. Potassium content (per cent shoot dry weight) of the shoots of wheat seedlings colonized by *G. graminis* or *P. radicicola*, and grown in media with or without added phosphorus (+P and −P), and with or without added potassium (+K and −K)

Inoculation treatment	−K treatments			+K treatments		LSD
	−P	+P	Means	−P	+P	
Uninoculated	0.43	0.47	0.45	1.37	1.71	0.53
G. graminis var. *tritici*	0.58	0.66	0.62	2.04	1.85	0.43
P. radicicola var. *radicicola*						
Isolate A	0.54	0.54	0.54	1.48	2.26	0.22
Isolate B	0.47	0.56	0.52	1.88	2.51	0.37
P. radicicola var. *graminicola*						
Isolate A	0.45	0.47	0.46	1.30	1.90	0.28
Isolate B	0.46	0.50	0.48	1.49	1.88	0.32
Means	0.49*	0.53*				
LSD			0.04	0.27	0.31	

Figures in columns 1, 2, 3, 4, 5 are means of 4 replicates.
LSD = Least significant difference ($p = 0.05$).
* Figures significantly different ($p < 0.001$).

ficiency are thus attributable to the considerable reduction in phosphorus and potassium levels in plants exposed to conditions of low phosphorus.

The improvements in the levels of phosphorus and potassium observed in seedlings colonized by *P. radicicola* var. *radicicola* may be explained by assuming that the presence of this fungus in the rhizosphere exerts a similar influence on uptake to that found in mycorrhizal roots[5,6,7]. Colonizing hyphae of *P. radicicola* var. *radicicola* penetrate the cortex of wheat roots as far as the endodermis, whereas hyphae of *P. radicicola* var. *graminicola* penetrate only as far as the first few layers of cortical cells[9]. This may be one reason for the different effects of these two fungi on the uptake of nutrients. On the basis of evidence currently available, it was not possible to explain why water use by the plants was affected by *P. radicicola* var. *graminicola*, but not by *P. radicicola* var. *radicicola*; both of these fungi were found to cause extra suberin deposition in the endodermal cell

Table 5. Quantities of ash (per cent shoot dry weight) obtained from the shoots of wheat seedlings colonized by *G. graminis* or *P. radicicola*, and grown in media with or without added phosphorus (+P and −P), and with or without added potassium (+K and −K)

Inoculation treatment	−K treatments		+K treatments		LSD
	−P	+P	−P	+P	
Uninoculated	5.7	10.1	6.4	10.4	0.7
G. graminis var. *tritici*	5.2	8.4	8.5	10.6	2.0
P. radicicola var. *radicicola*					
Isolate A	6.0	10.0	6.3	11.5	0.4
Isolate B	5.6	9.9	7.9	12.4	2.5
P. radicicola var. *graminicola*					
Isolate A	5.8	10.1	5.8	10.3	1.1
Isolate B	5.4	9.2	6.6	10.8	1.0
LSD	0.4	1.1	1.3	2.2	

Figures in columns 1, 2, 3, 4 are means of 4 replicates.
LSD = Least significant difference.

walls of wheat roots[9], and this might have been expected to give a greater resistance to water flow, regardless of which variety of *P. radicicola* was the cause.

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