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RESEARCH ARTICLE

Effect of Solution Phosphorus Concentration on the Exudation of Oxalate Ions by Wheat (*Triticum aestivum* L.)

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Abstract A solution culture experiment was conducted to investigate the effect of solution phosphorus concentration on the exudation of citrate and oxalate ions by five different varieties of the wheat (*Triticum aestivum* L). Wheat varieties were grown in a modified Hoagland nutrient solution containing five phosphorus levels (0, 0.2, 0.3, 0.4 and 0.5 µg/mL). Increasing the concentration of P significantly decreased oxalic acid exuded from roots. The exudation of oxalic acid was maximum at 10 days time interval. All the varieties of wheat were equally effective in exudating oxalic acid. The time interval had no significant effect on P concentration in plants. But significant interactive effect on plant P was found between varieties and different time intervals.

Keywords Citrate · Oxalate · Phosphorus · Wheat

Introduction

Phosphorus is one of the essential nutrients for plant growth and crop production. The importance of phosphorus in maintaining soil fertility and improving crop productivity has now been recognized much more than before in view of the fact that about 66 % of Indian soils are known to give universal response to P application and remaining 34 % to heavy feeding crops [1]. Phosphorus deficiency is a limiting factor of crop production in agricultural soils worldwide [2–4]. Correcting P deficiency with application of P fertilizers is not possible for the resource-poor farmers in the tropics and subtropics, especially on soil with high P fixing capacity. Knowledge about the principle mechanisms involved in efficient P acquisition by plants increased substantially during recent years [5, 6]. It has been attempted to use organic acids like citric acid and oxalic acid to extract labile P as an indicator of soil P availability [7, 8]. Low-molecular-weight oxalic acid plays an important role in the mobilization of soil phosphorus [9]. Oxalic acid exudation from roots is considered to be one of the mechanisms for plants to adapt to P deficiency [10] by mobilizing P in soil. The objective of the present study was to investigate the effect of solution phosphorus on the exudation of oxalic acid, in different varieties of wheat at different time intervals in sand culture.

Material and Methods

Seeds of different potential yield and genetic characters of wheat varieties viz. HD 2687; HD 2733; HD 2643, HD 2932; HD 2894 (Table 1) obtained from division of genetics, IARI, New Delhi were germinated on moist blotting paper in petriplates. After 7 days, the seedlings were taken out from petriplates and washed carefully under tap water to remove the adhering particles. Three seedlings of uniform size were selected and transplanted in PVC pots (3 cm diameter × 7 cm high) containing sand and graded doses of P ($P_1 = 0.0$, $P_2 = 0.2$, $P_3 = 0.3$, $P_4 = 0.4$ and $P_5 = 0.5$ µg/mL) with Hoagland solution. Solution from the pots was collected through outlet by displacement method intermittently at 5, 10, 20 and 30 days after transplanting and stored in refrigerator and then analyzed

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Table 1 Potential yield and genetic characters of the varieties

Variety	Characteristics
HD 2687	It is called Shresth. Released by IARI New Delhi, timely sown variety. Irrigated conditions of NWPZ region. It is bread wheat variety suitable for North Western Plains Zone, normal sown. The average yield varies 38.2–41.9 q/ha.
HD 2733	Bread wheat, North Eastern Plains Zone, normal sown. The average yield of HD 2733 is varies 40.0–42.2 q/ha.
HD 2643	Bread wheat, North Eastern Plains Zone, late sown variety. The average yield varies 39.5–41.3 q/ha.
HD 2932	It is called wheat PUSA 111. The variety HD 2932 has proved to be the most widely adapted and stable variety in the entire area falling in the central and peninsular parts of the country and exhibits high magnitude of adult plant resistance to leaf and stem rust diseases. The average yield of HD 2932 is 42.0 and 43.3 q/ha in central zone and peninsular zone respectively.
HD 2894	High yielding variety for NCR Delhi. It is developed by IARI New Delhi. Average yield of 5.2 t/ha. Protein content of 12.9 %, high gluten score, and good chapatti making. It is a replacement for PBW 343.

for citrate and oxalate ions with HPLC (Water 600 controller as a Water's USA) using a PDA detector. At each time interval, plants were harvested, oven dried at 70 °C and analyzed for P concentration by yellow color method [11]. Statistical analysis was done for factorial Complete Randomized Design [12]. During the experiment temperature was maintained between 21.5 and 39.8 °C and relative humidity between 51 and 90 %.

Results and Discussion

Oxalic and Citric Acids

The data on the organic acids revealed that wheat varieties exuded only oxalic acid. No citric acid was exuded. The concentration of oxalic acid decreased with the increasing level of phosphorus. P_1 recorded the highest concentration of oxalic acid (1.88 μg/mL) and was followed by P_2 (1.56 μg/mL), P_3 (1.33 μg/mL), P_4 (1.24 μg/mL) and P_5 (1.20 μg/mL). The varieties did not show any significant effect on exudation of oxalic acid but, their interactive effect with levels of phosphorus concentration was significant (Table 2). It was maximum (2.79 μg/mL) in case of P_1 (0 μg/mL) treatment with variety HD 2932 and minimum (0.84 μg/mL) in P_4 (0.4 μg/mL) with variety HD 2733.

It is known that low molecular oxalic acid is secreted from plant roots in P deficient soil conditions and it is one of the mechanisms for plants to adapt to P deficiency [13]. Several studies have suggested that the exudation of organic acids by plant roots might be a physiological adaptation rather than a passive response of plants to P deficiency [14]. It has been postulated that increased exudation was due to increased membrane permeability induced by decrease in phospholipids in P deficient roots [15]. If this is true, then greater the degree of P starvation, the greater the amount of root exuded organic acids that would be expected. The addition of inorganic P to starved roots results in both depolarization of the plasma membrane and acidification of the cytoplasm by secretion of low molecular organic acids [16]. Present results have confirmed this hypothesis. Addition of P to the nutrient solution significantly decreased the exudation of oxalic acid. The exudation of oxalic acid was maximum (1.59 μg/mL) at 10 days time interval, followed by 20 days time interval, while the least concentration (1.15 μg/mL) was measured at 30 days time interval. The interactions between levels of phosphorus in solution and time interval were not significant (Table 3). Between 10 and 25 days after germination of wheat crop, root and shoot growth accelerated secretion of more amounts of organic substances, which enhanced microbial activity releasing more amount of photosynthate in the soil through roots [17]. It is largely accepted that up to

Table 2 Effect of P concentration in solution and different varieties on oxalic acid (μg/mL) exudation during sand culture

Variety	Phosphorus concentration (μg/mL)					Mean
	P_1 (0.0)	P_2 (0.2)	P_3 (0.3)	P_4 (0.4)	P_5 (0.5)	
HD 2687	2.08	1.71	0.88	0.91	1.46	1.41
HD 2733	0.99	1.40	1.79	0.84	1.23	1.25
HD 2643	1.22	1.79	1.21	1.43	0.94	1.32
HD 2932	2.79	1.49	1.68	1.49	1.34	1.76
HD 2894	2.30	1.40	1.09	1.52	1.01	1.46
Mean	1.88	1.56	1.33	1.24	1.20	
LSD ($P < .01$)	Variety = NS	P = 0.60	Variety × P = 1.18			

Table 3 Effect of P concentration in solution and time interval on oxalic acid (μg/mL) exudation during sand culture

P concentration (μg/mL)	Time interval (days)				Mean
	5	10	20	30	
P_1 (0)	1.84	1.67	2.57	1.42	1.88
P_2 (0.2)	2.07	1.39	1.65	1.12	1.56
P_3 (0.3)	1.18	1.83	1.33	0.97	1.33
P_4 (0.4)	1.23	1.73	0.10	0.10	1.24
P_5 (0.5)	1.12	1.36	1.09	1.21	1.20
Mean	1.49	1.59	1.52	1.15	
LSD ($P < .05$)	P = 0.60	Time interval = 0.38	P × time interval = NS		

20–30 % of total C assimilated by higher plants is released in the rhizosphere as diverse exudates including respired CO_2 [18].

Phosphorus Concentration in Plants

The concentration of P in plant tissue increased with increased level of applied P (Table 4). In case of P_1, it was 0.157 % while in case of P_5, was 0.169 %. Different time interval had no significant effect on P concentration in plants, while the interaction effect between phosphorus concentration and time intervals was significant. The concentration of P was maximum (0.204 %) at 5 days time interval in the treatment P_4 while it was minimum (0.105 %) at 5 days time interval in P_3 (0.3 μg/mL). Increasing phosphorus concentration in soil solution enhanced the P uptake in crop plants. It might be due to more availability of labile phosphorus in soil solution [19]. More addition of inorganic P fertilizers at the period of vigorous growth of crop plant, directly enhanced the nutrient concentration in plant [20]. The concentration of P in different varieties was significant (Table 5). The lowest concentration was found in case of HD 2687 as 0.139 % while, highest in case of HD 2894 as 0.166 %. The interaction between phosphorus levels and varieties was also significant. The concentration of plant P was highest (0.193 %) in variety HD 2643 in P_4, whereas lowest concentration (0.103 %) was seen in variety HD 2733 in P_2 treatment. Phosphorus uptake by different varieties is affected by genetic potential of the varieties [21] as well as concentration of phosphorus in solution [20]. Transfer of inorganic P from the solution to the plant [22] involves a different set of thermodynamic parameters to those applying to the plasma membrane, mainly because of the millimolar concentrations in the cytoplasm and vacuoles [23]. The interaction between varieties and time interval was also found significant (Table 6). Maximum P-concentration (0.197 %) was recorded after 10 days time interval and minimum (0.129 %) after 30 days time interval in variety HD 2894. Different varieties having different genetic potential and biochemical reaction respond to particular ions from soil solution. In the present experiment, plant P concentration was improved substantially by the addition of P to the nutrient solution. This is due to the fact that increase in P concentration in solution near the root surface, increases P uptake rate in accordance with Michaelis and Menten equation [24]. Application of P is beneficial to the growth of wheat plant both in the field as well as under glasshouse condition. Exudation of oxalic acid plays an important role in the mobilization of soil phosphorus and ultimately enhances the concentration of P in plants [9].

In conclusion, the present results suggest that the organic acid exudation from wheat crop roots follow the thumb rule of organic acid exudation under P stress conditions. It will be better if one identifies maximum exudation of oxalic acid at various time intervals and high organic acid exudation varieties in different crops, before

Table 4 Effect of P concentration in solution and time interval on plant P concentration (%) during sand culture

P concentration (μg/mL)	Time interval (days)				Mean
	5	10	20	30	
P_1 (0)	0.159	0.173	0.158	0.138	0.157
P_2 (0.2)	0.125	0.145	0.145	0.152	0.142
P_3 (0.3)	0.105	0.156	0.142	0.128	0.133
P_4 (0.4)	0.204	0.151	0.152	0.134	0.160
P_5 (0.5)	0.167	0.180	0.163	0.165	0.169
Mean	0.152	0.161	0.152	0.143	
LSD ($P < .05$)	P = 0.029	Time interval = NS	P × time interval = 0.041		

Table 5 Effect of P concentration in solution and different varieties on plant P concentration (%) in sand culture

Variety	Phosphorus concentration (μg/mL)					Mean
	P_1 (0.0)	P_2 (0.2)	P_3 (0.3)	P_4 (0.4)	P_5 (0.5)	
HD 2687	0.151	0.139	0.120	0.145	0.141	0.139
HD 2733	0.158	0.103	0.132	0.153	0.183	0.146
HD 2643	0.153	0.154	0.126	0.193	0.164	0.158
HD 2932	0.173	0.151	0.141	0.153	0.139	0.151
HD 2894	0.150	0.161	0.146	0.157	0.217	0.166
Mean	0.157	0.142	0.133	0.160	0.169	
LSD ($P < .05$)	Variety = 0.285	P = 0.029	Variety × P = 0.045			

Table 6 Effect of varieties and time interval on P concentration (%) during sand culture

Variety	Time interval (days)				Mean
	5	10	20	30	
HD 2687	0.130	0.133	0.147	0.147	0.139
HD 2733	0.144	0.132	0.140	0.167	0.146
HD 2643	0.170	0.179	0.149	0.134	0.158
HD 2932	0.164	0.164	0.138	0.139	0.151
HD 2894	0.153	0.197	0.185	0.129	0.166
Mean	0.152	0.161	0.152	0.143	
LSD ($P < .05$)	Variety = 0.029	Time interval = NS	Variety × time interval = 0.041		

applying P fertilizers accordingly. It will help to reduce the amount of P fertilizers application and secure the global food security.

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