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Effect of Acid Treatment on Root Architecture in Seedlings of *Malus hupehensis* var. *pingyiensis*

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Abstract

The purpose of this paper is to explore the effects of acid treatment on root morphology and architecture in seedlings of *Malus hupehensis* var. *pingyiensis*. The rootstock seedlings were cultured in 1/2 Hoagland nutrient solutions of different pH (pH 4, pH 4.5, pH 5 and pH 6), respectively. The parameters of root architecture were measured in the day 4, 8 and 12 with the professional WinRHIZO 2007. Compared with the control (pH 6), the treatments significantly decreased the fractal dimension, length, diameter, surface area and volume of roots in day 8 and 12, and they kept decreasing followed the increase of the acidity and treatment time. The growth of lateral roots was more susceptible to acid treatment than taproots. In addition, the acid treatment mainly inhibited the growth of rootlets, significantly decreased the proportion of rootlets that changed the composition of roots, and then simplified the space structure of roots.

Keywords

Malus hupehensis var. *pingyiensis*, Acid Treatment, Fractal Dimension, Root Architecture

1. Introduction

Soil acidification is one different form of soil degradation, the form resulted from

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many factors such as agricultural measures became more and more serious in recent decades [1] [2]. Soil acidification damaged plant roots and made the whole plant harmful [3]. The research on the change of organism root under soil acidification condition has an important significance for the scientific management of plant. The studied results showed that soil acidification increased exchangeable Pb and bio-availability of Pb in the soils [4], decreased the activity of PS Ⅱ and the photosynthetic rate of the leaf on *Malus hupehensis* var. *pingyiensis* [5]. However, soil acidification firstly hurt the plant roots, the present studies focused on the problem on the plant injuries of heavy metal stress and other factors [6].

Because the difference between the seedling *Malus hupehensis* var. *pingyiensis* is very small, it is an ideal material for researching the fruit roots [7]. Classification and objective description of root architecture is an important precondition for the study of plant root architecture [8]. Nowadays, it has no report on the effects of root architecture affected by soil acidification, the relationships between root architecture and soil acidification in the apple tree and other trees are not clear. In this article, the purpose is to explore the effects of acid treatment on root length, diameter, surface area, volume and other parameters in seedlings of *Malus hupehensis* var. *pingyiensis* with the professional WinRHIZO 2007, which could provide vital basis for the scientific management fruit production safety and the root of economic tree.

2. Material and Methods

2.1. Study Materials

According to the experiment method [9], the seedlings cultivated by laminated seed of *Malus hupehensis* var. *pingyiensis* were completed in the plastic containers. When the seedlings just sent second leaf, they were transfered into the above containers and nourished in the one-half Hoagland's solution with pH of 6, solution being changed per 3 days. When these seedlings grew out sixth leaf, the selected seedlings with relatively uniform growth were treated by acidification.

2.2. Acidification Treatment in Rhizosphere

According to the test method [10], the concentrated acid was dripped to make the one-half Hoagland's solutions with pH of 4.0, 4.5, 5.0, 6.0, the pH of each of the above solutions was determined by use of UB-10 pH meter, respectively. The seedlings were measured for 4, 8, 12 days after acidification treatment, repeated for three times.

2.3. Measurement of Root Architecture

The parameters were measured according to the test method [11], the scanned images of seedling roots were acquired, and the fractal dimension, length, diameter, surface area and volume of roots was analyzed through the professional WinRHIZO 2007.

2.4. Data Analysis

The data in the article were handled through Excel statistical software 2003 and SPSS 17.0 statistical software package, the significant difference was conducted by Duncan's new multiple range method.

3. Results

3.1. Length of Total Roots, Taproots and Lateral Roots

It was shown in Table 1 that length of total roots in seedlings of *Malus hupehensis* var. *pingyiensis* began to change, length of its taproots had a little variation, length of its lateral roots obviously varied in the treatment pH 4, pH 4.5, pH 5 in 4 day ($P < 0.05$). At day 8, length of the total roots decreased by 18.18%, 11.30%, 5.49%, length of taproots reduced by 15.83%, 10.15%, 4.97%, length of its lateral roots declined by 18.28%, 11.50%, 5.63% in the treatment pH 4, pH 4.5, pH 5 compared with pH 6, respectively. At day 12, lengths of the total roots in the treatment pH 4, pH 4.5, pH 5 were smaller 23.78%, 16.32%, 9.25% than that in the treatment pH 6, their lengths of taproots reduced by 15.83%, 10.15%, 4.97% than that in the treatment pH 6, their lengths of lateral roots declined by 18.28%, 11.50%, 5.63% than that in the treatment pH 6, respectively. Length of taproots in seedlings of *Malus hupehensis* var. *pingyiensis* gradually decreased with the increase of acidity. Length of its roots had differently decreased compared to that of the treatment pH6 with the increase of acidity and the experiment

Table 1. Effect of acid treatment on length of roots, taproots and lateral roots in seedlings of *Malus hupehensis* var. *pingyiensis.*

Treatment	Days of treatment	Total root length/cm	Taproot length/cm	Lateral root length/cm
pH4	4	145.32 ± 6.10a(a)	7.62 ± 0.73a(a)	137.10 ± 4.88a(a)
	8	153.52 ± 2.54b(a)	8.29 ± 0.17a(a)	144.72 ± 2.42b(a)
	12	159.26 ± 0.99b(a)	8.78 ± 0.76a(a)	150.05 ± 0.91b(a)
pH4.5	4	154.39 ± 1.19a(b)	8.14 ± 0.64a(a)	145.93 ± 0.58a(b)
	8	166.42 ± 3.57b(b)	8.85 ± 0.57ab(a)	156.73 ± 3.21b(b)
	12	174.85 ± 2.31c(b)	9.63 ± 0.61b(a)	163.34 ± 1.38c(b)
pH5	4	159.38 ± 0.97a(bc)	8.51 ± 0.29a(a)	150.88 ± 0.49a(c)
	8	177.31 ± 1.75b(c)	9.36 ± 0.27b(b)	167.12 ± 1.50b(c)
	12	189.63 ± 0.51c(c)	10.53 ± 0.46c(b)	178.88 ± 0.44c(c)
pH6	4	164.58 ± 2.43a(c)	8.78 ± 0.43a(a)	155.23 ± 2.15a(c)
	8	187.62 ± 1.50b(d)	9.85 ± 0.92b(b)	177.09 ± 0.48b(d)
	12	208.96 ± 1.49c(d)	11.25 ± 0.40c(c)	197.96 ± 1.59c(d)

Duncan's multiple test, different letter outside the brackets indicated the significant differences among days of treatments in the same treatment, different letter in the brackets indicated the significant differences among the different treatments in the same day, small letter stand for 5% level.

time, the sequence of length reduction in the following decreasing order: lateral roots > total roots > taproots.

3.2. Diameter, Surface Area and Volume of Roots

The mean diameter of the seedling roots became smaller under acid treatment (Figure 1(a)). At day 4, the diameter of the seedling roots had no obvious variations. At day 8, the changes on diameter of the seedling roots in the treatment pH 4, pH 4.5, pH 5 compared to pH 6 had reached the significant level ($P < 0.05$), their diameters grew smaller by 15.11%, 12.72% and 7.46% than that in the treatment pH 6, respectively. At day 12, the changes of the diameter in the treatment pH 4, pH 4.5, pH 5 compared to pH 6 had reached the extremely significant level ($P < 0.01$), the diameter in the treatment pH 4 was the smallest among these treatments.

At day 4, the surface area of the seedling roots had no significant changes (Figure 1(b)). At day 8, the surface area of the seedling roots in the treatment pH 4, pH 4.5, pH 5 reduced by 31.84%, 22.56%, 8.63% Compared with that of the treatment pH 6, respectively. At day 12, their surface areas of the seedling roots became smaller by 42.77%, 30.84%, 12.99% than that of the treatment pH 6, respectively.

The changes of volume of roots were consistent with that of surface areas of the seedling roots, the volume of the seedling roots had no apparent variations (Figure 1(c)). At day 8, their volumes of the seedling roots in the different treatments had obviously decreasing compared with the treatment pH 6. At day 8, their volumes of the seedling roots in the treatment pH 4, pH 4.5, pH 5 compared to pH 6 had reduced by 37.06%, 34.88%, 12.81%, respectively.

3.3. Composition of Roots in Seedlings

Composition of roots in seedlings was analyzed according to the diameter (Table 2). At day 4, the percentage of length of $0 < D \leq 0.5$ mm fine roots had a decrease, however, the percentage of length of $0.5 < D \leq 1.5$ mm middle roots had a rise with an increase of acid concentrations. At day 8, the length and percentage of length of $0 < D \leq 0.5$ mm fine roots obviously decreased ($P < 0.05$),

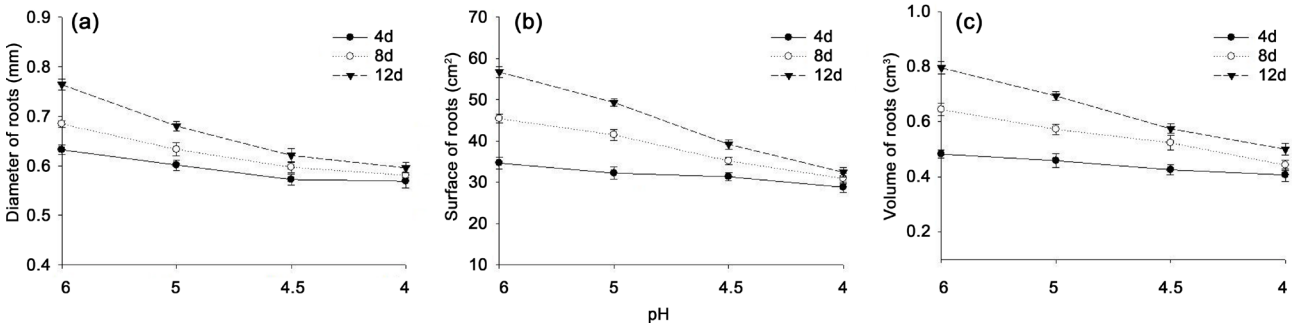

Figure 1. Effect of acid treatment on diameter of roots, surface of roots, volume of roots in seedlings of *Malus hupehensis* var. *pingyiensis*.

Table 2. The composition of roots in seedlings of *Malus hupehensis* var. *pingyiensis* under acid treatment.

Root diameter (mm)	pH	4d		8d		12d	
		Length (cm)	Percent (%)	Length (cm)	Percent (%)	Length (cm)	Percent (%)
0<D≤0.5	4	95.35 ± 2.19*	65.6	99.64 ± 1.98**	64.9	101.56 ± 0.84**	63.6
	4.5	107.52 ± 2.08*	69.6	115.11 ± 1.44*	69.2	120.03 ± 1.32**	68.6
	5	117.63 ± 1.01	73.6	130.99 ± 2.19*	73.8	139.36 ± 2.68*	73.6
	6	129.18 ± 1.78	78.3	149.24 ± 1.51	79.8	170.42 ± 1.38	81.2
0.5<D≤1	4	35.21 ± 1.14*	24.2	36.28 ± 1.90*	23.6	36.89 ± 0.56*	23.1
	4.5	33.28 ± 0.55*	21.5	34.67 ± 1.58*	20.9	35.42 ± 0.79*	20.3
	5	27.79 ± 0.79	17.4	29.72 ± 1.13	16.8	31.36 ± 1.07	16.6
	6	21.95 ± 0.83	13.3	23.75 ± 0.79	12.7	26.39 ± 1.00	12.7
1<D≤1.5	4	9.71 ± 0.66	6.7	13.61 ± 0.82	8.9	15.41 ± 0.66	9.6
	4.5	11.55 ± 1.39	7.5	12.62 ± 0.88	7.6	14.79 ± 1.08	8.5
	5	10.05 ± 0.97	6.3	11.26 ± 1.92	5.8	12.23 ± 0.42	6.5
	6	8.98 ± 0.66	5.5	9.91 ± 1.54	5.3	10.65 ± 0.69	5.1
1.5<D≤5	4	4.64 ± 1.02	3.2	4.83 ± 0.24	3.1	5.25 ± 0.47	3.3
	4.5	1.69 ± 0.47	1.1	3.28 ± 0.40	1.9	3.83 ± 0.55	2.2
	5	4.06 ± 0.76	2.5	5.13 ± 0.24	2.9	5.46 ± 0.52	2.9
	6	3.62 ± 0.46	2.2	4.11 ± 1.27	2.2	4.98 ± 1.28	2.4

*Indicate significant difference ($P < 0.05$), **Indicate extremely significant difference ($P < 0.01$).

however, the percentage of length of $0.5 < D \leq 1.5$ mm middle roots became larger with an increase of acid concentrations. At day 12, the percentage of length and length of $0 < D \leq 0.5$ mm fine roots significantly decreased ($P < 0.01$). These results showed that percentage of length of $0 < D \leq 0.5$ mm fine roots had a decreasing trend, the percentage of length of $0.5 < D \leq 1.5$ mm middle roots had an increase. In all, growth of $0 < D \leq 0.5$ mm fine roots was restrained, percentage of length of $0 < D \leq 0.5$ mm fine roots declined, composition of roots in seedling was affected by the acid treatment.

3.4. Fractal Dimension of Roots in Seedlings

It was shown from Figure 2 that fractal dimension of roots in seedlings became smaller with an increase of acid concentrations. At day 4, fractal dimension of roots in treatment pH 4 was largely affected and reduced by 7.26% compared with control treatment pH 6 among all the acid treatments. At day 8, fractal dimension of the seedling roots in the treatment pH 4, pH 4.5, pH 5 became smaller 31.84%, 22.56%, 8.63% than that of the treatment pH 6, respectively. At

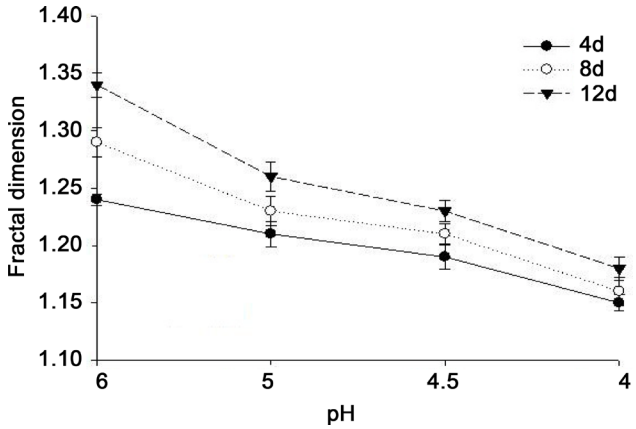

Figure 2. Effect of acid treatment on fractal dimension of roots in seedlings of *Malus hupehensis* var. *pingyiensis.*

day 12, their fractal dimension of the seedling roots also reduced by 42.77%, 30.84%, 12.99% compared with the treatment pH 6, respectively. The decrease of fractal dimension of roots in seedlings under acid treatment showed that acidification resulted in the reduction of branch and structure simple.

4. Conclusions

1) Length of total roots, taproots and lateral roots in seedlings of *Malus hupehensis* var. *pingyiensis* had apparent decrease under the increase of acidity and treatment time in the experiment, the reduction of lateral roots was the largest among the researched roots. The lateral roots were easily damaged compared to the taproots.

2) The parameters of the seedling roots in the different treatments such as diameter, surface area, volume of roots and fractal dimension obviously reduced under acidification treatment, had apparent decrease under the increase of acidity and treatment time in the experiment, it also demonstrated a dose effect relationship with acid concentrations. Those parameters of the seedling roots such as diameter, surface area, volume of roots and fractal dimension became much smaller than that of the treatment pH 6 with much higher concentration acidification.

3) Composition of roots in seedlings was changed significantly with the increase of acid concentrations and treatment days under acid treatment. Percentage of length of $0 < D \leq 0.5$ mm fine roots had a decreasing trend, percentage of length of $0.5 < D \leq 1.5$ mm middle roots had an increase. Growth of $0 < D \leq 0.5$ mm fine roots was restrained, percentage of length of fine roots declined, composition of roots in seedling was affected by the acid treatment.

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