Effects of Fulvic Acid on Photosynthetic Characteristics of Citrus Seedlings Under Drought Stress

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Abstract. The experimental material in the experiment was citrus seedling of Huangguogan. An experiment was conducted to study the effects of fulvic acid on photosynthetic characteristics of citrus seedlings under drought stress. Portable photosynthesis meter (LI-6400) was used to determine net photosynthetic rate and chlorophyll fluorescence. The results showed that the $P_n$ in citrus seedlings spraying with 200, 400, 600 mg·L$^{-1}$ fulvic acid was higher than that in control group, which increased with the increase of concentration of fulvic acid. With the increase of fulvic acid concentration, the $F_o$, $F_m$, $F_v$, $qP$, $F_v'$/[$F_m'$] in PSII and NPQ in citrus seedlings all increased significantly, while the $F_o$/[$F_v$] in PSII showed an opposite regulation pattern.

1. Introduction

Citrus is a subfamily of Citrus of Rutaceae [1]. According to FAO data, the proportion of citrus in the world’s total fruit production has increasing from 24% to 27% in the past 15 years [2]. Huangguogan is the natural hybrid citrus selected from China [3]. It has the characteristics of late ripening, high yield, having flowers and fruits on trees simultaneously, and has a broad sales prospect [4]. However, the special growing environment of Huangguogan (dry-heat valley areas) leads to the large demand for irrigation and high cost of management. Therefore, it is of great significance to seek an effective drought-resistant product in order to improve the efficient use of water and ensure the healthy and prosperous development of the industry.

It is reported that fulvic acid has been used in tomato, wheat, apple, strawberry and other crops to significantly improve its drought resistance. The regulation of fulvic acid on plant growth has been mainly manifested in promoting root development, increasing root activity and photosynthetic intensity of plants, and reducing the evaporation rate of crop leaves, followed by being able to improve the activity of multiple synthase and chlorophyll content in plants [5]. Photosynthesis is an important physiological process for the growth and development of plant, and plants accumulate dry matter through photosynthesis to complete the growth and metabolism. Drought stress is one of the most important abiotic factors, affecting crop growth and quality yield. It is reported that the net photosynthetic rate ($P_n$) of cucumber under mild drought stress could be significantly reduced. The value of $P_n$ can directly show the intensity of plant photosynthetic capacity. The Chlorophyll fluorescence in plants is an important index to detect and analyse the photosynthetic function of plants. Recently chlorophyll fluorescence has been used to estimate quantum efficiency and photosynthetic capacity [6].
In this experiment, the effect of fulvic acid on photosynthetic characteristics was analysed by measuring the net photosynthetic rate and chlorophyll fluorescence of Hugangguogan under drought stress condition.

2. Materials and Methods

2.1. Experimental materials
The one-year-old Huangguogan of 24 plants with the same growth were transplanted into a plastic pot.

2.2. Experimental design
The experiment was carried out in the greenhouse, and the selected potting soil was mixed with the field soil and nutrient soil (1:1). After a period of normal growth, seedlings without plant diseases and insect pest were selected for drought treatment. All materials were adequately irrigated three days before the drought treatment, and then the soil moisture content change was detected by weighing method. The control group had normal water supply, in which the soil water content was remained at 40% of field water capacity. The treatment group was under drought stress. Drought treatment began on 26 March, 2017 for 12 days, and the final soil moisture content maintained at about 20% of the field water capacity. Before the drought treatment, the plants neither in the treatment nor the control group were treated with fulvic acid respectively. The control group were: ① clear water was sprayed (CK1); ② 200mg·L⁻¹ fulvic acid was sprayed (F1); ③ 400mg·L⁻¹ fulvic acid was sprayed (F2); ④ 600mg·L⁻¹ fulvic acid was sprayed (F3); The treatment group were: ⑤ sprayed clear water (CK2); ⑥ 200mg·L⁻¹ fulvic acid (F4); ⑦ 400mg·L⁻¹ fulvic acid (F5); ⑧ 600mg·L⁻¹ fulvic acid (F6).

2.3. Test methods

2.3.1 Determination of net photosynthetic rate. From 9:30 to 13:00 on sunny days, the \( P_n \) of Huangguogan leaves was determined by portable photosynthesis apparatus (L1-6400). The parameters were set as follows: light intensity was 1400 \( \mu \text{mol} \cdot \text{m}^{-2} \cdot \text{sec}^{-1} \), \( \text{CO}_2 \) concentration was 400 \( \mu \text{mol} \cdot \text{mol}^{-1} \), and temperature was 25°C.

2.3.2 Determination of chlorophyll fluorescence properties. From 9:30 to 13:00 on sunny days, chlorophyll fluorescence properties of Huangguogan leaves was determined by portable photosynthesis apparatus (L1-6400). The \( \text{CO}_2 \) concentration was 400 \( \mu \text{mol} \cdot \text{mol}^{-1} \)[7]. The minimum fluorescence (\( F_o' \)), the initial fluorescence (\( F_o \)), the steady-state fluorescence (\( F_s \)), maximum fluorescence (\( F_m \)), variable fluorescence (\( F_v \)), photochemical quenching coefficient (\( qP \)), effective photochemical quantum yield of PSII (\( F'_{v'}/F_{m'} \)) and non-photochemical coefficient (NPQ) were detected and calculated.

3. Results and analysis

3.1 Effect of fulvic acid on \( P_n \) in leaves of Huangguogan
As was shown in figure 1, the \( P_n \) of the plants showed an upward trend except CK1 in the treatment group with time went on. In the F1 treatment, the value on 12th day was 4.6% and 9.7% higher than that on day 8 and day 4 respectively. \( P_n \) in the control group showed a downward trend. In the treatment of F6, \( P_n \) decreased by 8.4% and 14.1% on day 12 compared with the measured values on day 8 and day 4 respectively. It indicated that the \( P_n \) of plants decreased with the increase of drought degree.

In the control and treatment group, the plants treated with 600mg·L⁻¹ fulvic acid had the highest value of \( P_n \). On 6 April, \( P_n \) in F6 was higher than that of F5, F4 and CK2, and the difference was significant. The results showed that the \( P_n \) of Huangguogan could significantly be improved by spraying different concentrations of fulvic acid.
3.1 Effect of fulvic acid on the $P_n$ of Huangguogan leaves

It was shown in Fig. 5 that $F_o$, $F_m$, $F_v$, $qP$, $F_v'/F_m'$ and NPQ increased significantly in the control group and treatment group, which shown as $600 \text{ mg} \cdot \text{L}^{-1} > 400 \text{ mg} \cdot \text{L}^{-1} > 200 \text{ mg} \cdot \text{L}^{-1} > \text{CK}$. The potential activity of $F_o/F_v$ was the opposite as $600 \text{ mg} \cdot \text{L}^{-1} < 400 \text{ mg} \cdot \text{L}^{-1} < 200 \text{ mg} \cdot \text{L}^{-1} < \text{CK}$; The $F_v$, $F_v'/F_m'$, $F_v/F_m$ in control group were significantly lower than that of treatment group. The results showed that the photochemical activity of plants was inhibited under drought stress. With the increase of fulvic acid concentration, the plant was reduced by the inhibition of photochemical activity.

![Graph showing the effect of fulvic acid on $P_n$ of Huangguogan leaves](image)

Fig 1. Effect of fulvic acid on the $P_n$ of Huangguogan leaves

3.2 Effect of fluorescence parameters of chlorophyll in leaves of Huangguogan

It was shown in Fig. 5 that $F_o$, $F_m$, $F_v$, $qP$, $F_v'/F_m'$ and NPQ increased significantly in the control group and treatment group, which shown as $600 \text{ mg} \cdot \text{L}^{-1} > 400 \text{ mg} \cdot \text{L}^{-1} > 200 \text{ mg} \cdot \text{L}^{-1} > \text{CK}$. The potential activity of $F_o/F_v$ was the opposite as $600 \text{ mg} \cdot \text{L}^{-1} < 400 \text{ mg} \cdot \text{L}^{-1} < 200 \text{ mg} \cdot \text{L}^{-1} < \text{CK}$; The $F_v$, $F_v'/F_m'$, $F_v/F_m$ in control group were significantly lower than that of treatment group. The results showed that the photochemical activity of plants was inhibited under drought stress. With the increase of fulvic acid concentration, the plant was reduced by the inhibition of photochemical activity.
Fig. 2 Effect of fulvic acid on the chlorophyll fluorescence of Huangguogan leaves

4. Discussion and conclusion
In this experiment, the related photosynthetic indexes were determined by spraying different concentrations of fulvic acid on the seedlings of Huangguogan, and it was found that the $P_n$ of leaves
was inhibited under drought stress. However, spraying different concentrations of fulvic acid could significantly increase the net photosynthetic rate and promote photosynthesis of Huangguogan.

It was determined that spraying fulvic acid promoted the photosynthesis of Huangguogan leaves, and the optimum concentration of fulvic acid in seedlings was 600 mg·L⁻¹. The effect of fulvic acid on Huangguogan was only studied on seedlings. Whether it had the same effect in the field experiment needed further study.

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