ISSN: 2472-3703 DOI: 10.6911/WSRJ.202308_9(8).0016

Effects of Phenolic Allelochemicals on Plant Photosynthesis and Chlorophyll Fluorescence

Yu Dong[1, a], Weiguo Fu[1, b]

[1]College of Agricultural Engineering, Jiangsu University, Zhenjiang, Jiangsu, China

[a]2622214609@qq.com, [b]fuweiguo@ujs.edu.cn

Abstract

In this study, a control pot experiment was used to investigate the effects of different concentrations of p-hydroxybenzoic acid and cinnamic acid on the parameters of photosynthetic gas exchange and chlorophyll fluorescence in lettuce and tomato. The results showed that with the increase of phenolic acid concentration, the net photosynthetic rate, transpiration rate and stomatal conductance of recipient plants showed a downward trend, while the intercellular carbon dioxide concentration showed an upward trend; the PSII maximum light energy conversion efficiency, PSII electron transfer quantum efficiency, and photochemical quenching coefficient of recipient plants all showed a downward trend, while the non-photochemical quenching coefficient showed an upward trend. It can be obtained that the treatment of phenolic allelochemicals inhibits the photosynthesis of recipient plants.

Keywords

Phenolic acids; Photosynthesis; Chlorophyll fluorescence.

1. INTRODUCTION

Allelopathy refers to the favorable or unfavorable effects (mostly unfavorable) of the metabolic secretions of plants or microorganisms on other plants or microorganisms in the environment[1], these metabolic secretions with allelopathic effects are called allelopathic substances. Among them, phenolic allelochemicals are one of the main allelochemicals secreted by allelopathic plants[2]. The number of phenolic allelochemicals isolated and identified at this stage accounts for more than half of all allelochemicals[3]. Among the numerous phenolic allelochemicals, p-hydroxybenzoic acid and cinnamic acid are important and widely present in higher plants, microorganisms, mosses and soils[4,5]. Therefore, p-hydroxybenzoic acid and cinnamic acid are often used as representatives of phenolic allelochemicals for various researches on allelopathic effects.

In recent years, agricultural harvests have been bumper year after year, and the production of crop straws has also increased year by year. Straw burning is a serious phenomenon, which not only wastes resources, but also brings a series of environmental problems, such as causing air pollution, greenhouse gas emissions, and reducing soil fertility[6,7]. Therefore, the current treatment of straw is mainly to return the straw directly to the field or make it into an organic matrix[8-10]. And due to the increase in demand for lettuce and tomato, the cultivation of crop straw as an organic substrate to achieve high yields has gradually spread across the world. However, crop straw usually contains various allelochemicals that inhibit plant growth. Among the many allelochemicals, phenolic acids such as p-hydroxybenzoic acid and cinnamic acid are relatively common and representative allelochemicals[4,5]. The photosynthetic gas exchange parameter is the most important index to express the strength of plant photosynthesis, and it is

ISSN: 2472-3703 DOI: 10.6911/WSRJ.202308_9(8).0016

also one of the means to understand the response of plant photosynthesis to adversity stress[11]. When plants are under allelopathic stress, photosynthesis is usually affected, resulting in growth inhibition, and the degree of photosynthesis is usually reflected by photosynthetic gas exchange parameters. Chlorophyll fluorescence signals emitted by plants can usually be used to quickly, sensitively and non-invasively study and detect the true behavior of photosynthesis of plants under stress conditions[12,13]. Therefore, by analyzing the effects of different concentrations of p-hydroxybenzoic acid and cinnamic acid on the photosynthetic gas exchange parameters and chlorophyll fluorescence parameters of lettuce and tomato, this study explored whether phenolic allelochemicals reduced the photosynthetic capacity of lettuce and tomato, inhibited Its growth provides theoretical guidance for the scientific return of straw.

2. MATERIALS AND METHODS

2.1. Experimental environment

The experiment was carried out in the artificial climate chamber and laboratory of Agricultural Engineering College of Jiangsu University. During the whole experiment, the parameters of the artificial climate chamber were set as follows: The light intensity is set to 400 μmol • m^{-2} • s^{-1}.The temperature is set to 25/20 ℃ (day/night), The light cycle is set to 10/14h (light/dark), and the air humidity is set to about 70%.

2.2. Experimental design

Experimental materials: In this study, p-hydroxybenzoic acid and cinnamic acid were used as allelochemicals, and lettuce and tomato were used as allelopathic receptor plants (these two plants can be planted every year, which is conducive to the full arrangement of the experiment). Select good and consistent lettuce and tomato seedlings for transplanting, and after transplanting, pour enough water and place them in a cool place to slow down the seedlings. After the slow seedlings survived, they were all moved into the artificial climate chamber.

Experimental method: All experimental recipient plants were planted in plastic pots (upper diameter 16.5 cm, lower diameter 8.9 cm, height 11 cm). Each pot is filled with 1 kg loam, and its nutrient status is 16.05 g kg^{-1} for C, 1.16 g kg^{-1} for N and 0.72 g kg^{-1} for P. In this experiment, four treatment concentrations of p-hydroxybenzoic acid and cinnamic acid were respectively set, P-hydroxybenzoic acid: CK (0 g L^{-1})、T1(1 g L^{-1})、T2(2 g L^{-1})、T3(3 g L^{-1}), cinnamic acid: CK(0 mg L^{-1})、T1(5 mg L^{-1})、T2(50 mg L^{-1})、T3(500 mg L^{-1}). Each treatment was replicated 4 times, and each pot represented one replicate, a total of 64 pots. Lettuce and tomato were irrigated once every three days with solutions containing different concentrations of phenolic allelochemicals. The indicators were measured after one month of treatment.

2.3. Determination index

(1) Determination of photosynthetic gas exchange parameters

The photosynthetic gas exchange parameters were measured from 9:00 to 11:00 in the morning, and the photosynthetic physiological indicators of lettuce and tomato under different concentration treatments were measured with LI-6400 portable photosynthetic measuring instrument. Four replicates were selected for each treatment, and the 3-4 expanded leaves from top to bottom were selected. The flow rate of the photosynthetic instrument was set to 500umol·s^{-1}, and the light was set to 500μmol·m^{-2}·s^{-1}.

(2) Determination of chlorophyll fluorescence parameters

Chlorophyll fluorescence parameters were measured at 9:00-11:00 in the morning, using the IMAGING-PAM chlorophyll fluorescence imager. The selection criteria of the leaves are the same as above, and the relevant indicators are measured after the plants are dark-treated for 20 minutes.

ISSN: 2472-3703 DOI: 10.6911/WSRJ.202308_9(8).0016

2.4. Data processing

After the data were processed with Excel 2016, one-way One variance Way ANOVA and multiple comparisons (Multiple Comparisons) were performed with SPSS 22.0.

3. RESULTS AND ANALYSIS

3.1. Effects of different intensities of allelopathy on photosynthetic gas exchange parameters of recipient plants

In this study, with the increase of p-hydroxybenzoic acid and cinnamic acid concentration, the changes of photosynthetic gas exchange parameters of lettuce and tomato are shown in Table 1 and Table 2.

Table 1. Changes of photosynthetic gas exchange parameters in recipient plants under different concentrations of p-hydroxybenzoic acid

p-HA Treatment	Pn/(μmol·m^{-2}s^{-1})		Gs/(mol·m^{-2}s^{-1})		Ci/(μmol·m^{-2}s^{-1})		Tr/(mmol·m^{-2}s^{-1})	
	Lettuce	Tomato	Lettuce	Tomato	Lettuce	Tomato	Lettuce	Tomato
CK	13.88 ±0.02a	10.33 ±0.57a	0.31 ±0.01a	0.48 ±0.01a	235.29 ±1.17c	216.81 ±1.02c	2.00 ±0.06a	3.36 ±0.16a
T1	12.69 ±0.33b	9.91 ±0.52b	0.29 ±0.02a	0.35 ±0.00b	248.30 ±1.02b	231.66 ±0.76b	1.72 ±0.08b	2.89 ±0.19b
T2	12.08 ±0.56c	8.36 ±0.21c	0.26 ±0.01b	0.27 ±0.01c	257.30 ±1.07a	243.25 ±0.98b	1.27 ±0.07c	2.19 ±0.19c
T3	11.29 ±0.47d	7.19 ±0.32c	0.22 ±0.01c	0.22 ±0.01d	261.17 ±0.59a	258.86 ±0.45a	0.92 ±0.09d	1.70 ±0.16d

Note: Different lowercase letters indicate that the photosynthetic indicators are significantly different under different treatments (P<0.05).

Table 2. Changes of photosynthetic gas exchange parameters in recipient plants under different concentrations of cinnamic acid

CA Treatment	Pn/(μmol·m^{-2}s^{-1})		Gs/(mol·m^{-2}s^{-1})		Ci/(μmol·m^{-2}s^{-1})		Tr/(mmol·m^{-2}s^{-1})	
	Lettuce	Tomato	Lettuce	Tomato	Lettuce	Tomato	Lettuce	Tomato
CK	13.66 ±0.14a	8.94 ±0.29a	0.39 ±0.01a	0.48 ±0.01a	227.04 ±1.39d	217.27 ±1.70d	1.85 ±0.05a	3.52 ±0.15a
T1	12.57 ±0.18b	7.91 ±0.18b	0.35 ±0.03b	0.39 ±0.02b	247.43 ±0.26c	234.89 ±1.51c	1.57 ±0.02b	2.82 0.13b
T2	11.33 ±0.36c	7.22 ±0.19c	0.27 ±0.03c	0.31 ±0.01c	268.48 ±0.35b	246.49 ±0.97b	1.41 ±0.04c	2.25 0.17c
T3	10.71 ±0.24d	6.31 ±0.53d	0.23 ±0.02d	0.28 ±0.01d	286.55 ±0.34a	258.87 ±1.61a	1.24 ±0.03d	1.76 0.11d

Note: Different lowercase letters indicate that the photosynthetic indicators are significantly different under different treatments (P<0.05).

Net photosynthetic rate (Pn) can be used as a very critical index to test the photosynthetic response ability of plants under stressful environmental conditions. It can be seen from Table 1 and Table 2 that with the increase of the concentration of p-hydroxybenzoic acid and cinnamic acid, the Pn of lettuce and tomato showed a downward trend. Under different concentrations of p-hydroxybenzoic acid, the Pn of lettuce decreased by 8.57%, 12.96% and 18.66% compared with CK at T1, T2 and T3; the Pn of tomato decreased by 4.06%, 19.07% and 30.39%. Under different concentrations of cinnamic acid, the Pn of lettuce decreased by 7.98%, 17.06% and

ISSN: 2472-3703 DOI: 10.6911/WSRJ.202308_9(8).0016

21.59% at T1, T2 and T3, respectively. Compared with CK, the Pn of tomato decreased by 11.52%, 19.24% and 29.42% at different concentrations, and lettuce and tomato decreased significantly at T1 treatment (P<0.05). It was shown that both p-hydroxybenzoic acid and cinnamic acid could lead to a decrease in the net photosynthetic rate of lettuce and tomato, and the greater the concentration, the greater the decrease.

With the increase of concentration of p-hydroxybenzoic acid, the stomatal conductance (Gs) of lettuce decreased by 7.88%, 40.79% and 53.79% compared with CK in T1, T2 and T3 treatments, and the transpiration rate (Tr) decreased by 14.08%, 36.73% and 59.12% respectively. The Gs of tomato decreased by 25.94%, 43.93%, and 53.35% compared to CK at each treatment concentration, while Tr decreased by 14.06%, 34.86%, and 49.41%, respectively. Under different concentrations of Cinnamic acid, the Gs of lettuce under T1, T2 and T3 treatments decreased by 6.11%, 19.33% and 27.42% respectively compared with CK; Tr decreased by 14.63%, 23.49%, and 32.66% respectively; The Gs of tomato decreased by 17.39%, 36.44%, and 55.9% compared to CK under each treatment concentration, while Tr decreased by 19.84%, 35.95%, and 49.79%, respectively. lettuce and tomato had significantly decreased under T1 treatment (P<0.05).

It can be seen from Table 1 and Table 2 that with the increase of p-hydroxybenzoic acid concentration, the intercellular carbon dioxide concentration (Ci) of lettuce and tomato increased by 5.5%, 9.4% and 10.99% respectively compared with CK, and the φ ΦPSII of tomato at each treatment concentration increased by 6.8%, 12.19% and 19.39% respectively. With the increase of cinnamic acid treatment concentration, the ΦPSII of lettuce at each treatment concentration increased by 8.98%, 18.25% and 26.21% compared with CK, and the ΦPSII of tomato at each treatment concentration increased by 8.11%, 13.45% and 19.15% compared with CK, respectively, and lettuce and tomato have been significantly increased at T1(P<0.05).

3.2. Effects of different intensities of allelopathy on chlorophyll fluorescence parameters of recipient plants

In this study, with the increase of phenolic acid allelochemical stress, the changes of chlorophyll fluorescence parameters of recipient plants are shown in Table 3 and Table 4.

Photosystem II maximum light energy conversion efficiency (Fv/Fm) reflects the level of plant photochemical efficiency. It can be seen from Table 3 and Table 4 that with the increase of p-hydroxybenzoic acid treatment concentration, the Fv/Fm of lettuce and tomato decreased by 0.41%, 2.09% and 3.01% respectively compared with CK, and the Fv/Fm of tomato at each treatment concentration was lower than that of CK respectively. Decreased by 5.3%, 7.06% and 10.63%; With the increase of cinnamic acid treatment concentration, the Fv/Fm of lettuce decreased by 1.34%, 2.35% and 4.1% compared with CK at each concentration, and the Fv/Fm of tomato at each concentration decreased by 2.86%, 3.99% and 6.22% compared with CK. %, and they all decreased significantly at T1 (P<0.05).

PSII electron transfer quantum efficiency (ΦPSII) reflects the actual light energy conversion efficiency and activity of PSII, and its size can be used as an important indicator of the speed of photosynthetic electron transfer in plant leaves[46]. It can be seen from Table 3 and Table 4 that with the increase of p-hydroxybenzoic acid treatment concentration, the PSII electron transfer quantum efficiency (ΦPSII) of lettuce and tomato decreased by 22.29%, 31.9% and 43.96% compared with CK, respectively. Compared with CK, the ΦPSII of tomato at each treatment concentration decreased by 7.1%, 21.53% and 28.02%, respectively; with the increase of cinnamic acid treatment concentration, the ΦPSII of lettuce at each treatment concentration decreased by 7.59%, 11.48% and 30.28% compared with CK, respectively. The ΦPSII of tomato at each concentration decreased by 22.32%, 29% and 36.66% compared with CK, respectively, and both lettuce and tomato decreased significantly at T1 (P<0.05).

ISSN: 2472-3703 DOI: 10.6911/WSRJ.202308_9(8).0016

It can be seen from Table 3 and Table 4 that with the increase of p-hydroxybenzoic acid and cinnamic acid treatment concentrations, the photochemical quenching (qP) values of lettuce and tomato decreased gradually, indicating that p-phenolic acid treatment would reduce the photosynthetic activity of lettuce. And while the qP of lettuce and tomato showed a downward trend, the non-photochemical quenching (NPQ) showed a continuous upward trend, and qP and NPQ showed a trend of trade-off, indicating that the two were in a competitive relationship. The NPQ of lettuce and tomato continued to rise with the increase of phenolic acid concentration, which was beneficial to protect their own photosynthetic mechanism and function from excessive damage.

Table 3. Changes of chlorophyll fluorescence parameters of recipient plants under different concentrations of p-hydroxybenzoic acid

p-HA Treatment	Fv/Fm		ΦPSII		qP		NPQ	
	Lettuce	Tomato	Lettuce	Tomato	Lettuce	Tomato	Lettuce	Tomato
CK	0.74 ±0.03a	0.72 ±0.06a	0.54 ±0.03a	0.40 ±0.03a	0.58 ±0.02a	0.84 ±0.01a	0.66 ±0.01d	0.31 ±0.05d
T1	0.73 ±0.05a	0.68 ±0.01b	0.42 ±0.02b	0.37 ±0.08b	0.50 ±0.01b	0.75 ±0.01b	0.76 ±0.04c	0.43 ±0.02c
T2	0.72 ±0.01b	0.67 ±0.02c	0.37 ±0.01c	0.31 ±0.02c	0.46 ±0.02c	0.63 ±0.04c	0.81 ±0.02b	0.54 ±0.04b
T3	0.71 ±0.01c	0.64 ±0.01d	0.30 ±0.02d	0.29 ±0.01d	0.39 ±0.02d	0.54 ±0.02d	0.90 ±0.04a	0.66 ±0.01a

Note: Different lowercase letters indicate that the fluorescence measurement indicators are significantly different under different treatments (P<0.05).

Table 4. Changes of chlorophyll fluorescence parameters of recipient plants under different concentrations of cinnamic acid

CA Treatment	Fv/Fm		ΦPSII		qP		NPQ	
	Lettuce	Tomato	Lettuce	Tomato	Lettuce	Tomato	Lettuce	Tomato
CK	0.74 ±0.02a	0.70 ±0.01a	0.57 ±0.01a	0.43 ±0.02a	0.63 ±0.04a	0.83 ±0.02a	0.54 ±0.00d	0.27 ±0.03d
T1	0.73 ±0.01b	0.68 ±0.00b	0.53 ±0.00b	0.33 ±0.02b	0.55 ±0.02b	0.75 ±0.05b	0.64 ±0.03c	0.35 ±0.05c
T2	0.72 ±0.02b	0.67 ±0.00c	0.51 ±0.01c	0.30 ±0.01c	0.44 ±0.00c	0.65 ±0.02c	0.71 ±0.02b	0.47 ±0.02b
T3	0.71 ±0.02c	0.66 ±0.01d	0.40 ±0.02d	0.27 ±0.00d	0.36 ±0.02d	0.57 ±0.04d	0.95 ±0.06a	0.66 ±0.03a

Note: Different lowercase letters indicate that the fluorescence measurement indicators are significantly different under different treatments (P<0.05).

4. DISCUSS

More and more studies have shown that changes in external environmental factors usually significantly affect the photosynthesis of plants, and allelopathic stress is one of the forms of adversity that plants often suffer[14]. In this study, the effects of exogenous phenolic acids on the photosynthetic gas exchange parameters of lettuce and tomato were studied. The results showed that with the increase of p-hydroxybenzoic acid and cinnamic acid concentration, the net photosynthetic rate of lettuce and tomato showed a downward trend. Stomata are the main channels for gases such as CO2 and O2 to enter and exit mesophyll cells. Its size directly determines the level of plant photosynthetic rate. In this study, the stomatal conductance of recipient plants decreased with the increase of the concentration of phenolic acid substances.

ISSN: 2472-3703 DOI: 10.6911/WSRJ.202308_9(8).0016

The trend of change is consistent. Plants mainly rely on transpiration to complete the growth and physiological metabolism in the body, absorb and transmit nutrients, and maintain normal body temperature. The main way for plants to consume water is also transpiration, and changes in surrounding environmental factors can easily affect the transpiration of plants. Influence[15]. In this study, the transpiration rate of the recipient plants gradually decreased with the increase of the concentration of phenolic acid substances. The results of this study indicated that the treatment of phenolic allelochemicals inhibited the photosynthesis of recipient plants.

From the perspective of energy metabolism and conversion, the measurement of chlorophyll fluorescent signal molecules in plants can quickly, sensitively and non-destructively detect their photosynthesis. Therefore, this method is usually used to detect the effects of stress on plant photosynthesis Influence[16]. Fv/Fm and ΦPSII are usually important parameters used to characterize the efficiency of plant photochemical reactions. In this study, with the increase of the concentration of phenolic acid substances, the Fv/Fm and ΦPSII of recipient plants showed a downward trend. The size of qP and NPQ can reflect whether the light energy absorbed by plants is used for photosynthesis or dissipated in the form of heat. The two are in a competitive relationship. While the qP of lettuce and tomato showed a downward trend with the increase of phenolic acid concentration, NPQ showed a continuous upward trend, indicating that phenolic acid inhibited the photosynthesis of recipient plants and the recipient plants increased NPQ to prevent its The photosynthetic apparatus was highly damaged, which reflected a self-protection mechanism of lettuce and tomato in the face of allelopathic stress.

5. CONCLUSION

The results showed that with the increase of the concentration of phenolic acid allelochemicals, the photosynthetic inhibition of allelopathy on recipient plants was significantly enhanced, and the phenomenon of allelopathic stress appeared and intensified continuously. The main results were as follows: net photosynthetic rate, transpiration rate and stomatal conductance showed a downward trend, intercellular carbon dioxide concentration showed an upward trend, PSII maximum light energy conversion efficiency, PSII electron transfer quantum efficiency and photochemical quenching coefficient showed a downward trend, non-photochemical quenching coefficient showed an upward trend, and plant growth was inhibited.

REFERENCES

[1] Rice E L. *Allelopathy* (2rk edition) (Academic Press, Florida 1984).

[2] Cheng J, Wang F Q, Li X S, et al. Research progress on types, extraction, separation and detection of phenolic allelochemicals[J].Jiangsu Agricultural Sciences,2022,50(06):8-15.

[3] Weidenhamer J D, Macias F A, Fischer NH, et al. Just how insoluble are monoterpenes? [J]. Chem Ecol. 1993, 19(8): 1799-1807.

[4] Sun W. *The effect of coumarin and cinnamic acid on the occurrence of muskmelon wilt* (MS., Shenyang Agricultural University, China 2022).

[5] Wang L. *Effects of exogenous p-hydroxybenzoic acid on the nitrogen metabolism of grape rhizosphere soil and plants* (MS., Shenyang Agricultural University, China 2022).

[6] Wang H P, Meng F H. Hazards and causes of straw burning [J]. Modern Agricultural Science and Technology, 2015,No. 661(23): 216-217.

[7] Bernard A. Goodman. Utilization of waste straw and husks from rice production: A review[J]. Journal of Bioresources and Bioproducts, 2020, 5(3).

ISSN: 2472-3703 DOI: 10.6911/WSRJ.202308_9(8).0016

[8] Liu H X. Research progress of straw returning on soil improvement [J]. Special Economic Animals and Plants, 2022, 25 (02): 123,124.

[9] Fan R Q, Luo J, Yan S H, et al. Research Progress on substrate Utilization Technology of crop Straw [J]. Journal of Ecology and Rural Environment, 2016, 32 (03): 410-416.

[10] Yang M. Main problems and countermeasures of returning corn straw to field in China [J]. Agricultural Technology and Equipment, 2016, No. 259 (01): 65-66.

[11] Liu Z B, Cheng R M, Xiao W F, et al. Effects of flooding stress on photosynthetic physiology and ecology of plants [J]. World Forestry Research, 2013, 26 (03): 33-38.

[12] Wang D. *Interference of p-hydroxybenzoic acid on ecostoichiometric balance and growth physiological inhibition of lettuce* (MS., Jiangsu University, China 2021).

[13] Li X, Feng W, Zeng X C. Progress in chlorophyll fluorescence analysis technology and its application [J]. Acta Botanica Sinica, 2006 (10): 2186-2196.

[14] Pan R C. Plant Physiology. 5th ed. Beijing: Higher Education Press, 2004: 56-57.

[15] Ma M W. *Exogenous phenolic acid on Trichosanthes kirilowii (Trichosanthes kirilowii Maxim.). Study on physiological characteristics and effects of Rhizosphere soil Environment* (MS., Zhejiang Normal University, China 2020).

[16] Wen G S, Tian H T, Zhang M R, et al. Application of chlorophyll fluorescence analysis technique in forest cultivation [J]. Journal of Applied Ecology, 2006 (10): 1973-1977.