

Full Length Research Paper

Accumulation of cinnamic acid and vanillin in eggplant root exudates and the relationship with continuous cropping obstacle

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Accepted 12 November, 2010

The contents of cinnamic acid and vanillin in eggplant root exudates and soil were determined by HPLC. The results showed that cinnamic acid and vanillin might remain in soil after the root of eggplant is released. With the extending growth stage and planting year of eggplant, the contents in root exudates, rhizosphere and continuous cropping soil increased. In bioassay and field studies, the allelopathy of cinnamic acid and vanillin to the verticillium wilt (*V. dahliae*) and the eggplant was observed, and when the concentration of cinnamic acid or vanillin was at 1 and 4 mmol L⁻¹, it inhibited the eggplant growth, while the occurrence of verticillium wilt was promoted. A high concentration of cinnamic acid and vanillin caused eggplant autotoxicity and increased the risk of disease infection, which finally led to continuous cropping obstacle.

Key words: Cinnamic acid, vanillin, root exudates, soil, verticillium wilt, continuous cropping obstacle.

INTRODUCTION

Eggplant (*Solanum melongena* L.) is a popular vegetable crop worldwide, but its yield and quality sharply reduced annually owing to continuous monoculture, and as such, the adverse effect was called continuous cropping obstacle, which is a key reason that restrains eggplants' sustainable production (Yu et al., 2000). There are many factor responses for continuous cropping obstacle, such as soil-born diseases, nutrition unbalance and allelopathy (Gao et al., 1998; Zheng et al., 2004; Yu et al., 2000). As such, soil-born diseases had been paid more attention in eggplants continuous cropping obstacle studies (Liu et al., 2009; Shipton, 1977; Wang, 2005; Yin et al., 2008; Zhou et al., 2001). Recently, many researches showed that allelopathy had close relationship with continuous cropping obstacle. When the allelochemicals in root exudates or residues of previous crop were left in soil and accumulated to a certain level, the growth and yield of the following crop in continuous cropping were inhibited, that is, autotoxicity takes place (Cao et al., 2001; Fisher, 1980; Huang et al., 2000, 2002; Wu et al., 1999; Yu et al.,

1993, 1999).

Autotoxicity, which is one of the obvious characters of allelopathy, especially for root exudates, was one of the main reasons for continuous cropping obstacle (Wang et al., 2005b; Wu et al., 2003; Yu et al., 1999). The former study of eggplants has indicated that cinnamic acid and vanillin existing in the root exudates were the two basic autotoxicity substances (Wang, 2005). So far, the regulations of cinnamic acid and vanillin exudated from root, accumulation in rhizosphere and continuous cropping soil have not been clear. The report on the relationship between accumulation of cinnamic acid and vanillin and continuous cropping obstacle is still little. The study aims to illuminate the relationship between the two autotoxicity substances and the eggplant growth and verticillium wilt, understand the mechanism of eggplant autotoxicity and how to overcome the problem of monoculture.

MATERIALS AND METHODS

Extraction and determination of cinnamic acid and vanillin

The experiment was carried out in the Vegetable Experiment Station, Shenyang Agricultural University. The commonly grown eggplant (*Solanum melongena* L.) cv. 'Xi an lv' was used as test

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plant and the seeds were provided by Shenyang Agricultural University. Seeds were sown on March 5, 2007, while the seedlings were transplanted into 12.5 cm diameter plastic pots on March 20. On April 15, some eggplant seedlings were transplanted into 25 cm diameter container with substrates of soil, manure and turf (1:1:1 ratio).

Collection of root exudates and soil supernatants

Root exudates

Root exudates were collected with root soaking method at bud stage, first fruit expanding stage and second fruit expanding stage, respectively. The seedlings were taken out of substrate and the soil was completely washed away with water, and then with distilled water. These seedlings were put into beakers covered with black plastic containing 200 ml distilled water. After 3 h incubation, the distilled water was collected. This action was repeated 5 times and the collected solution (5 times) was mixed and then filtered.

Soil supernatants

The rhizosphere soil of seedlings were collected at bud stage, first fruit expanding stage and second fruit expanding stage, respectively, and the soil was collected from the place where eggplants were planted for 0, 1, 2 and 3 years in eggplant monoculture garden, respectively. The aforementioned soil samples were air-dried at room temperature ($20 \pm 5^\circ\text{C}$) and passed through a sieve with 0.3 mm openings. Soil solutions were obtained by adding sieved soil at the rates of 40% (w/v) to distilled water in 500 ml flasks, and then, the flasks were covered with stoppers and agitated on a platform shaker for 1 h. Then the soil solutions were passed through a filter paper and centrifuged at 12,000 rpm for 10 min. Finally, the soil supernatants that are to be used for the determination test (20) were collected. There were 3 replications in each treatment.

Determination of cinnamic acid and vanillin

The root exudates and soil supernatants were adsorbed with XAD-4 resin produced by Sigma Company and concentrated after dissolved with alcohol, then cinnamic acid (CA) and vanillin (VN) were determined with Waters high pressure liquid chromatography (HPLC, USA) (Wang et al., 2005b).

Effects of cinnamic acid and vanillin on verticillium wilt

According to the results of HPLC analysis and previous experiments on eggplants, concentrations of cinnamic acid or vanillin were selected as 0 (control), 0.1, 0.5, 1.0 and 4 mmolL^{-1} , respectively (Wang et al., 2006). Firstly, cinnamic acid or vanillin was dissolved in alcohol and then diluted with distilled water, keeping the alcohol content to 1.5% (Zheng et al., 2007) to make the aforementioned solutions of different concentration.

Mycelium growth

The *V. dahliae* was the tested pathogen used in the concentrated solutions of 0, 10, 50, 100 and 400 mmolL^{-1} cinnamic acid or vanillin that were ready. Subsequently, 1 ml of the aforementioned concentrated solutions was added to 99 ml PDA cultural medium, and was then sterilized and cooled to 40°C . Afterwards, it was taken out with 25 ml and poured into each Petri dish (8.5 cm in

diameter). However, the final concentration in the PDA cultural medium was 0, 0.1, 0.5, 1.0 and 4 mmolL^{-1} . The 6 mm-diameter pure cultured *V. dahliae* spot was inoculated into the center of the PDA cultural medium and incubated at 25°C . There were three replications for each concentration. From the 3rd day to the 7th day of incubation, the colony diameter was measured every 24 h with the cross method. The mycelium growth was assayed using the growth rate method (Li et al., 2007) and the RI value of mycelium was calculated using the same method.

Field disease (eggplant verticillium wilt) investigation

The 'Xi and lv' eggplant seeds were sown on April 5, 2008. When the seedlings reached 4 - 5 leaf stage, they were transplanted into 20.5 cm diameter container with 1.5 kilogram substrate containing garden soil, manure and peat (1:1:1 ratio). Two weeks later, the seedlings were irrigated with 0, 0.1, 0.5, 1.0 and 4 mmolL^{-1} of cinnamic acid or vanillin every three days for four times, respectively. Three days after the fourth irrigation, each seedling was applied with 100 ml pathogen suspension of *V. dahliae* around the root (10 cm). Each treatment had 3 replications in random permutation and 10 seedlings in each replication. Typical wilt symptoms of disease appeared, and plant infection was investigated every 4 days according to the methods of Zhou (Zhou et al., 1998).

Effects of cinnamic acid and vanillin on germination and growth of eggplants

The seeds of 'Xi and lv' eggplants were used for testing germination. Concentrations of cinnamic acid or vanillin were selected as 0 (control), 0.1, 0.5, 1.0 and 4 mmolL^{-1} , respectively. Germination bioassays were done in Petri dishes (9 cm in diameter) and each contained 30 seeds with two sheets of filter paper. A 3 ml solution of cinnamic acid or vanillin was added as per treatments and 3 ml (1.5%) alcohol solution was added as control. The Petri dishes were incubated in a growth chamber at 30°C for 16 h and 18°C for 8 h in darkness. There were three replicates for each concentration and the dishes were in a complete randomization design. After germination, the number of germinated seeds of each dish was counted every 24 h. On the seventh day after germination, germination rate, the index of germination speed, the root and shoot length were measured and the RI value was calculated.

Eggplant growth and physiological characteristics

The seeds of 'Xi and lv' eggplant were sown on April 5, 2008. When the seedlings reached 4 to 5 leaf stage, they were transplanted into 20.5 cm diameter container with 1.5 kilogram substrate containing garden soil, manure and peat (1:1:1 ratio). After 1 week, the seedlings were irrigated around the root (10 cm) once every 5 days by cinnamic acid or vanillin of 0 (control), 0.1, 0.5, 1.0 and 4 mmolL^{-1} , respectively. Each seedling was irrigated in 100 ml each time and each treatment had 9 replications in random permutation. After 1 month, plant height, stem diameter, fresh weight, relative electrical conductivity, malondialdehyde (MDA) content and superoxide dismutase (SOD) activity of the plant were measured (Chen et al., 2008), and the RI values were counted.

Statistical analysis

The allelopathic effect of cinnamic acid or vanillin on mycelium growth and the seed germination and eggplant growth were expressed in RI value. RI indicated the intensity of allelopathic effect, which

Table 1. The contents of cinnamic acid and vanillin in root exudates and rhizosphere soil of eggplant.

Growth stages	Root exudates (ug.g ⁻¹ root DM)		Rhizosphere soil (ug.g ⁻¹ soil DW)	
	Cinnamic acid	Vanillin	Cinnamic acid	Vanillin
Bud stage	0.029 bB	2.520 cC	0.011 cC	0.260 cC
First fruit expanding stage	0.060 bB	4.430 bB	0.320 bB	2.210 bB
Second fruit expanding stage	0.540 aA	12.260aA	0.610 aA	2.440 aA

Numbers with different small and capital letters within each column show significant differences from control at 0.05 and 0.01 levels, respectively.

Table 2. The contents of cinnamic acid and vanillin in eggplant soil of different continuous cropping years (ug.g⁻¹ soil DW).

Planting years	Cinnamic acid	Vanillin
0	0.000 dD	0.000 dD
1	0.097 cC	0.568 cC
2	0.180 bB	0.810 bB
3	0.302 aA	1.115 aA

The results showed that cinnamic acid and vanillin not only existed in root exudates, rhizosphere and replanting soil, but also their content increased with the extending growth stage and planting years (Tables 1 and 2). Numbers with different small and capital letters within each column show significant differences from control at 0.05 and 0.01 levels, respectively.

was presented by Williamson (Williamson, 1988) that $RI = 1 - C/T$ ($T \geq C$) and $RI = T/C - 1$ ($T < C$). However, C was the control datum, T was the treatment datum and RI was the allelopathic index. $RI > 0$ indicated stimulation, while $RI < 0$ indicated inhibition. In the germination test, the index of germination speed (I) were counted by the following formula:

$$I = 2 \times (7 \times X_1 + 6 \times X_2 + 5 \times X_3 + 4 \times X_4 + 3 \times X_5 + 2 \times X_6 + 1 \times X_7),$$

where X_n : is the number of germinated seed for every 24 h. In the disease (eggplant verticillium wilt) infection test, disease index was counted according to the following equation:

$$\text{Disease index} = \frac{[\sum(\text{rating number} \times \text{number of infected plants})]}{(\text{total number of plants} \times \text{highest rating})} \times 100\%$$

The original experimental data were conducted with Microsoft Office Excel 2003, and the statistical analysis was done using SPSS 11.5 software package (SPSS Inc, USA). Meanwhile, significant differences test at 5 and 1% level respectively, was calculated using LSD significant difference test of SPSS software.

RESULTS

Cinnamic acid and vanillin determination

The results showed that cinnamic acid and vanillin existed in root exudates, rhizosphere and replanting soils. Their content varied with growth stage and planting years and there was significant difference in the content between them (Tables 1 and 2). Contents of cinnamic acid and vanillin in rhizosphere and replanting soil were rising

increasingly with growth stages and planting years. At the second fruit expanding stage, their content in root exudates and rhizosphere reached its maximum, and in replanting soil, it lasted for 3 years. Also, both contents were at the highest level. The content of cinnamic acid was generally lower than that of vanillin, with cinnamic acid accounting for 1.15~4.40%, 3.30~25.00% and 14.58~27.09% of vanillin in root exudates, rhizosphere and replanting soil, respectively.

Effects of cinnamic acid and vanillin on verticillium wilt

Allelopathy intensity (RI) of cinnamic acid and vanillin on mycelium growth of verticillium wilt were shown in Figure 1. The effect of cinnamic acid on mycelium showed similar trend to vanillin, and both had stimulatory or inhibitory growth on mycelium, depending on the concentration and incubation time. The mycelium growth was suppressed by cinnamic acid at the concentration of 0.1 and 0.5 mmolL⁻¹ (except day 6 and 7, 0.5 mmolL⁻¹), and was then promoted at the concentration of 1 and 4 mmolL⁻¹. Inhibition of vanillin on mycelium decreased with the concentration increasing from 0.1 to 1 mmolL⁻¹ (except 5d, 6d and 7d, 1 mmolL⁻¹), while 1 and 4 mmolL⁻¹ vanillin indicated promotion. When the mycelium was incubated for 5 to 6 days, each concentration treatment for cinnamic acid and vanillin provided the strongest allelopathy intensity (stimulation or inhibition). The treatment of 0.1 mmolL⁻¹ cinnamic acid incubated for 6d or 0.1 mmolL⁻¹ vanillin incubated for 5d, has the maximum inhibitions with RI of -0.058 and -0.056.

Field disease (eggplant verticillium wilt) investigation

Eggplants treated with cinnamic acid and vanillin were different from the control in disease resistance to *V. dahliae*, with small disease index at a lower concentration and big disease index at a higher concentration (Table 3). On May 12, inhibitory disease index of 0.1 mmolL⁻¹ cinnamic acid and 0.5 mmolL⁻¹ vanillin over control increased more than 50%, and then on June 1, it remained at a higher level (40.53 and 47.97%, respectively) over control. When compared with the control's disease index of 83.41, 4 mmolL⁻¹ cinnamic

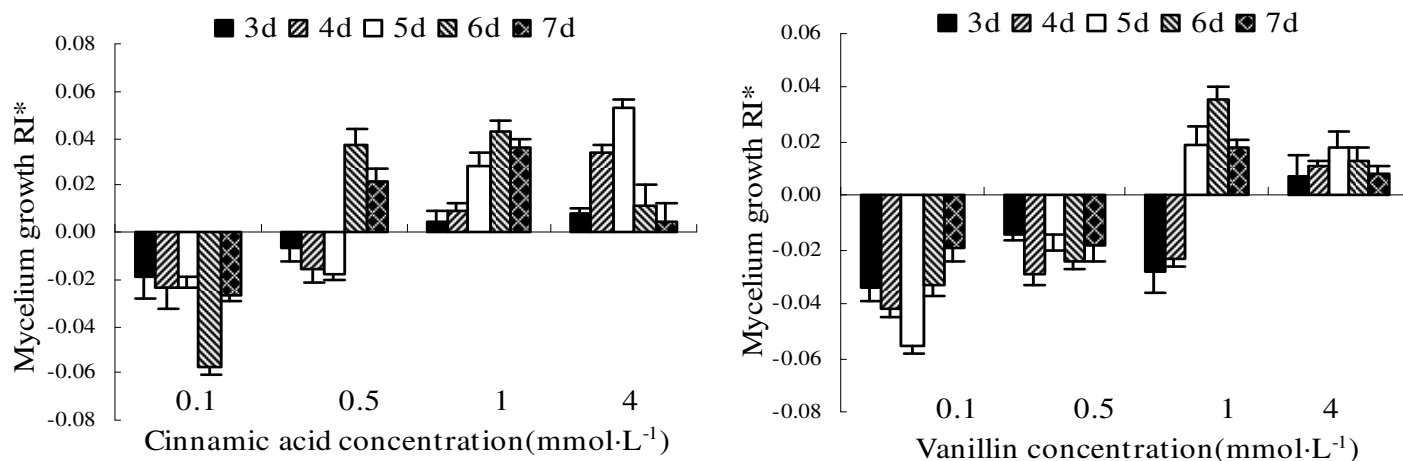


Figure 1. Effects of cinnamic acid and vanillin on mycelium growth of eggplants verticillium wilt (*over control). The mycelium growth was suppressed at cinnamic acid concentration of 0.1 and 0.5 $\text{mmol}\cdot\text{L}^{-1}$ (except 6d and 7d, 0.5 $\text{mmol}\cdot\text{L}^{-1}$), and was then promoted at 1 and 4 $\text{mmol}\cdot\text{L}^{-1}$ concentration. Inhibition of vanillin on mycelium decreased with concentration increasing from 0.1 to 1 $\text{mmol}\cdot\text{L}^{-1}$ (except 5d, 6d and 7d, 1 $\text{mmol}\cdot\text{L}^{-1}$), and vanillin of 1 and 4 $\text{mmol}\cdot\text{L}^{-1}$ indicated promotion. In general, lower concentration of cinnamic acid (0.1 and 0.5 $\text{mmol}\cdot\text{L}^{-1}$) and vanillin (0.1, 0.5 and 1 $\text{mmol}\cdot\text{L}^{-1}$) showed suppression, while higher concentration showed promotion on mycelium growth of *V. dahliae*.

Table 3. Effects of cinnamic acid and vanillin on the disease index of verticillium wilt of eggplant.

Treatment	Concentration ($\text{mmol}\cdot\text{L}^{-1}$)	Disease index				
		May 12	May 17	May 22	May 27	June 1
Control	0.0	45.20aA	54.20aA	58.40cB	70.25bB	83.41bB
	0.1	17.20fF	23.40dD	45.20eD	50.29fF	49.60eF
Cinnamic acid	0.5	27.14dC	30.52cC	40.06fE	57.60eE	68.50dD
	1.0	30.80cC	46.70bB	64.52aA	65.23cC	79.60cB
	4.0	32.16cC	39.60cC	52.40dC	80.23aA	89.15aA
Vanillin	0.1	24.60eD	39.20cC	44.57eD	43.15gG	56.50eE
	0.5	15.20gF	23.60dD	31.10gF	39.65hH	43.40eG
	1.0	35.80cB	46.23bB	58.93cB	60.40dD	74.20cC
	4.0	39.30 bB	56.10 aA	71.20bA	89.10aA	92.16aA

Numbers with different small and capital letters within each column show significant differences from control at 0.05 and 0.01 levels, respectively. This table indicated that cinnamic acid or vanillin of higher concentration eliminated eggplant resistance, while that of lower concentration enhanced resistance to *verticillium* wilt. That is to say, accumulation of allelochemicals, that is, cinnamic acid and vanillin made eggplant *verticillium* wilt more serious. Allelochemicals of higher concentration weakened suppression on growth of *V. dahliae*, which made it possible for eggplant to raise *verticillium* wilt occurrence.

acid or vanillin was observed to be significantly higher in disease index with 89.15 and 92.16 on June 1. The present data indicated that cinnamic acid or vanillin with higher concentration eliminated eggplant resistance, while that of lower concentration enhanced resistance to verticillium wilt.

Effects of cinnamic acid and vanillin on germination and growth of eggplants

Figure 2 suggested that cinnamic acid and vanillin had allelopathic effects on seed germination and seedling growth, with promotion at a lower concentration and

inhibition or slight promotion at a higher concentration. For cinnamic acid, eggplants germination and seedling growth were stimulated at 0.1 $\text{mmol}\cdot\text{L}^{-1}$ concentration, whereas inhibitory effects were enhanced with the concentration increasing from 0.5 to 4 $\text{mmol}\cdot\text{L}^{-1}$ (except shoot height at 1 $\text{mmol}\cdot\text{L}^{-1}$). At 0.5 $\text{mmol}\cdot\text{L}^{-1}$ cinnamic acid, the RI of germination rate index, germination rate, root length and shoot height was -0.24, -0.076, -0.182 and -0.077, after which the inhibition intensity increased by 238.33, 777.63, 98.90 and 62.33% at 4 $\text{mmol}\cdot\text{L}^{-1}$, respectively. We conclude that germination rate was the easiest to be inhibited, and root length was more aptly inhibited than shoot height by higher concentrations of cinnamic acid. As such, vanillin of each concentration presented

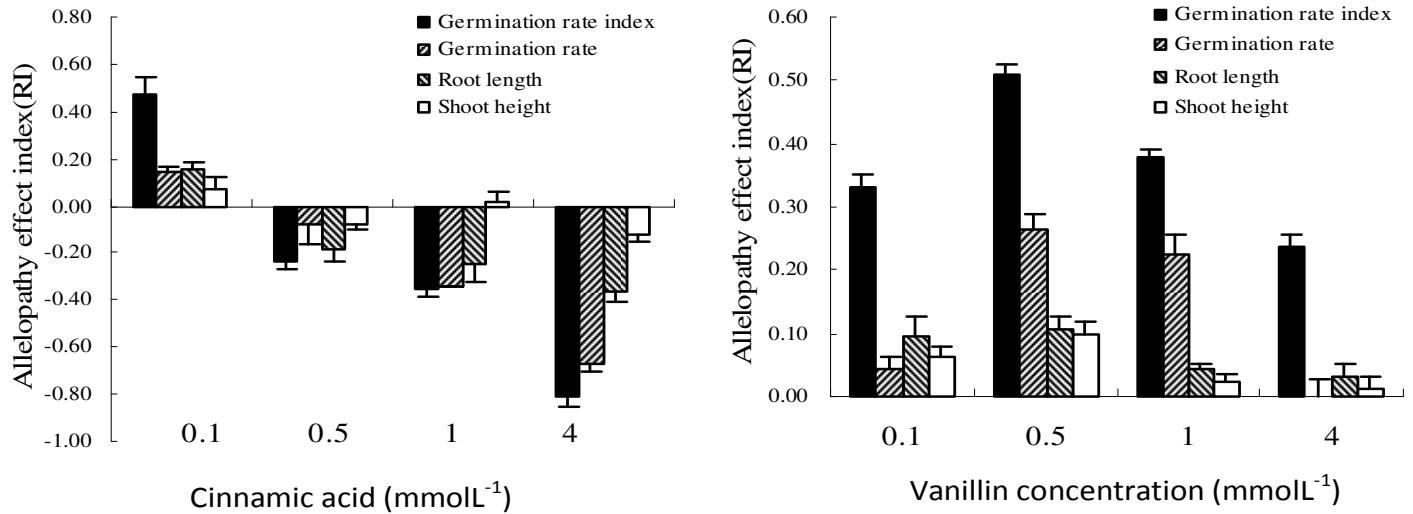


Figure 2. Effects of cinnamic acid and vanillin on RI of seed germination and seedling growth of eggplant (over control*). This figure suggested that cinnamic acid and vanillin had allelopathic effects on seed germination and seedling growth, with promotion at lower concentration and inhibition or slight promotion at higher concentration. For cinnamic acid, eggplants germination and seedling growth were stimulated at 0.1 mmol⁻¹ concentration, whereas inhibitory effect was enhanced with concentration increasing from 0.5 to 4 mmol⁻¹ (except shoot height, at 1 mmol⁻¹). Vanillin of each concentration presented promotion with lower concentration (from 0.1 to 0.5 mmol⁻¹) enhanced promotion with higher concentration (from 0.5 to 4 mmol⁻¹) and decreased promotion with no concentration.

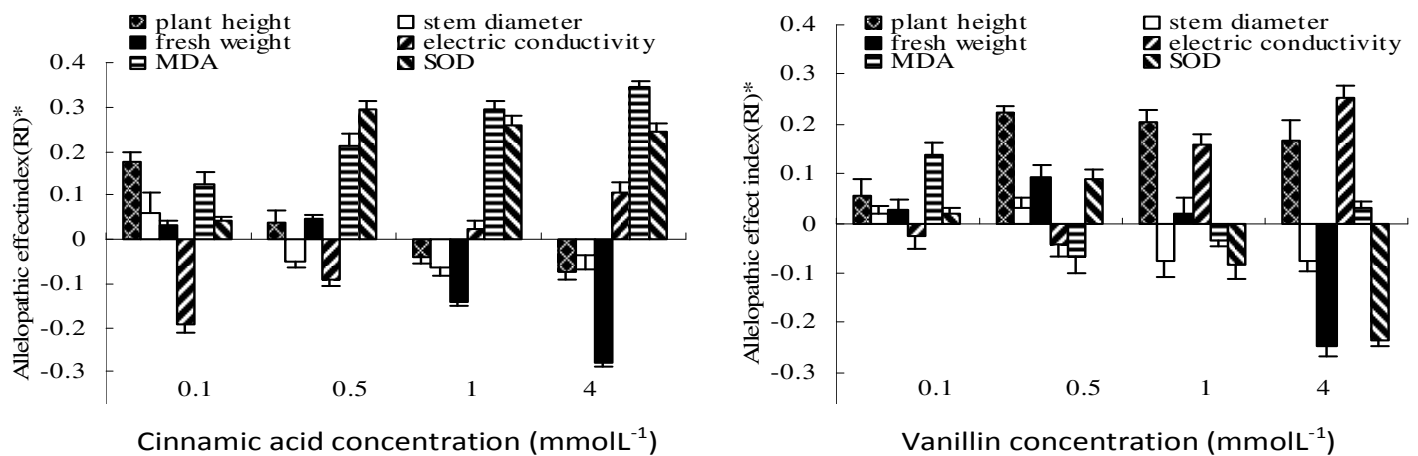


Figure 3. Effects of cinnamic acid and vanillin on RI of growth and some physiological characteristics of eggplant (*over control). This figure indicated that 0.1 and 0.5 mmol⁻¹ of cinnamic acid and vanillin had stimulatory effect on eggplant growth (plant height, stem diameter and fresh weight), while lower stimulation or inhibition was observed at higher concentration. The effects on physiological characteristics of cinnamic acid and vanillin exhibited stimulation or inhibition, depending on their concentrations. Electric conductivity and MDA significantly increased at higher concentrations of 1 and 4 mmol⁻¹. Malondialdehyde (MDA) content and relative electrical conductivity were higher for 4 mmol⁻¹ concentration. However, SOD activity was stimulated in cinnamic acid treatments, but in lower concentration treatment of 0.1 to 1 mmol⁻¹.

promotion with lower concentrations (from 0.1 to 0.5 mmol⁻¹) enhanced promotion with higher concentrations (from 0.5 to 4 mmol⁻¹) and then decreased promotion with no concentration. At 0.5 mmol⁻¹ of vanillin, the promotions on germination rate index, germination rate, root length and shoot height were the strongest with RI of 0.510, 0.266, 0.108 and 0.10, which decreased by 55.10, 100, 70.37 and 87.00% at 4 mmol⁻¹, respectively.

However, germination rate was the most susceptible to high concentration of vanillin.

Eggplant seedling growth and physiological characteristics

From Figure 3, cinnamic acid and vanillin of 0.1 and 0.5

mmolL⁻¹ had stimulatory effects on eggplant growth, and thus, lower stimulation or inhibition was observed at a higher concentration. The plant height, stem diameter and fresh weight were markedly suppressed by 4 mmolL⁻¹ concentration of cinnamic acid and vanillin, with the inhibition of cinnamic acid on eggplant growth being the most intense.

The allelopathic effect of cinnamic acid and vanillin on physiological characteristics of eggplant exhibited stimulation or inhibition, depending on their concentrations. As such, when compared with the control, electric conductivity and malondialdehyde (MDA) significantly increased at 1 and 4 mmolL⁻¹ concentrations. The MDA content and relative electrical conductivity were higher at the concentration of 4 mmolL⁻¹. When the concentration was 4 mmolL⁻¹, cinnamic acid had bigger stimulation with RI of 0.1956 and 0.3468, respectively. As such, a concentration of 4 mmolL⁻¹ vanillin was more stimulated with the RI of 0.07814 and 0.2514, respectively. When compared to the control, the superoxide dismutase (SOD) activity in cinnamic acid treatments was stimulated, exhibiting a stronger effect with the RI of 0.2944 at 0.5 mmolL⁻¹. For vanillin, SOD activity was stimulated by a lower concentration from 0.1 to 1 mmolL⁻¹, but was significantly inhibited by 4 mmolL⁻¹.

DISCUSSION

In the previous studies, soil-borne disease was the main problem caused by continuous cropping. The studies on the relationship between the eggplant root exudates and the plant diseases had begun since 1930's. Studies showed that the resistance on soil-borne disease of plant root is related to root exudates compositions. However, plant root exudates contained several compounds. The release of these compounds by root exudates leads to intense microbial activity (including pathogen) in the rhizosphere and soil.

Based on the present result of this study, cinnamic acid or vanillin was not only exudated to rhizosphere by root, but was retained in soil. With their extending growth stage and planting years, their content in root exudates, rhizosphere and soil increased. The result is accorded with the reports of Xu and Ma (Ma et al., 2005; Xu et al., 2008). The relationship between the content and soil diseases and the eggplant growth is needed for further research. However, it would be helpful to well understand the effect of allelochemicals exudated from eggplants root on continuous cropping obstacle.

Verticillium wilt was one of the serious soil diseases in eggplant production. The study indicated that lower concentration of cinnamic acid (0.1 and 0.5 mmolL⁻¹) and vanillin (0.1, 0.5 and 1 mmolL⁻¹) showed suppression, and thus, higher concentration showed promotion on mycelium growth of *V. dahliae*, which supported the previous study by Wang (Wang et al., 2006). In addition,

the present data suggested that cinnamic acid and vanillin of lower concentration contribute to reduce verticillium wilt of eggplant, while higher concentration increase disease occurrence. That is to say, accumulation of cinnamic acid and vanillin, as important allelochemicals, made eggplant verticillium wilt more serious. Allelochemicals of higher concentration weakened suppression on growth of *V. dahliae*, which made it possible for eggplant to raise verticillium wilt occurrence. Therefore, more emphasis should be placed on the effect of allelochemicals with higher concentration on eggplant, to further reveal cropping obstacle problem.

It has been recognized that allelochemicals could affect the physiological processes of plant growth and can ultimately cause allelopathic effect on seed germination and seedling growth. Allelopathy of phenolic acids on seed germination and seedling growth of crops have been reported. Cotton seed germination and seedling growth were suppressed by higher concentration of para-hydroxybenzoic benzoic acid, ferulic acid and vanillic acid (Wang et al., 2001). 2,4-Dihydroxy-1,4-benzoxazin-3-one and 2-benzoxazinone reduced the growth of lentil seedling (Sampietro et al., 2006a, b). However, the root growth of lettuce and weed species was affected by vanillic acid (Sampietro et al., 2007).

The present study suggested that cinnamic acid and vanillin presented allelopathic effects on seed germination and seedling growth of eggplants. Germination was promoted by cinnamic acid at 0.1 mmolL⁻¹ or vanillin of each concentration. Therefore, germination was more susceptible for cinnamic acid than vanillin. Promotive effect on plant height, stem diameter, fresh weight and superoxide dismutase (SOD) activity of eggplant seedling were observed in the study in the presence of 0.1 and 0.5 mmolL⁻¹ concentrations. Although they promoted the aforementioned indexes, they did not affect the malondialdehyde (MDA) content and relative electrical conductivity. The weakened promotion or suppression enhanced with a concentration from 1 to 4 mmolL⁻¹ significantly increased the MDA content and relative electrical conductivity and thus, the SOD significantly decreased. MDA content, relative electrical conductivity and SOD can reflect the ability to clean reactive oxygen. The results showed that higher concentration of cinnamic acid and vanillin decreased the ability to clean reactive oxygen, so that the degree of cell membrane lipid peroxidation became severer and severer. In consequence, the seed germination and seedling growth were prevented, which might be a reason for enhancing disease infection. The present results supported the fact that allelochemicals of higher concentration had inhibition, which were in accordance with reports on other allelochemicals (Chen et al., 2002; Wu et al., 2002). As a result, when allelochemicals such as cinnamic acid and vanillin were accumulated in the soil to a certain level, germination, seedling growth and eggplant growth were prohibited, and the soil disease such as verticillium wilt, which finally

led to continuous cropping obstacle, occurred greatly.

Conclusion

We conclude that exudated cinnamic acid and vanillin from root might go into soil and remain in soil. They were accumulated gradually in soil with the extending growth stage and planting years. As such, they presented allelopathic effect on verticillium wilt (*V. dahliae*) and eggplant seedling growth, once their concentration reached a high level. Higher concentrations promoted verticillium wilt pathogen, and most importantly, they eliminated seedling growth vigor to cause eggplant disease resistance decrease, which leads to the occurrence of verticillium wilt greatly. Allelochemicals of higher concentration in soil prohibited eggplant growth and aggravated soil-borne disease, which finally caused continuous cropping obstacle. However, the role that allelochemicals play in soil nutrient and microorganism is unknown. Further study to reveal the effect of cinnamic acid and vanillin on soil biological characteristics, and a relationship of cinnamic acid and vanillin to overcome continuous cropping obstacle are required.

ACKNOWLEDGEMENTS

The research was supported by National Natural Science Foundation of China (30771469), the 11th Five Year Key Support Programs for Science and Technology Development of China (2008BADA6B02) and the Innovation Group of Higher Education of Liaoning Province (2009T087).

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