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Research Paper

Effects of exogenous TIBA on dwarfing, shoot branching and yield of tea plant (*Camellia sinensis* L.)

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ABSTRACT

2,3,5-Triiodobenzoic acid (TIBA) is a safe and efficient synthetic auxin transport inhibitor. The mature tea plants of the cv. Longjing 43 were used in this study. The foliar portion of tea bushes were sprayed with different concentrations (50, 100, or 150 mg L^{-1}) of TIBA after heavy pruning (i.e., after two to three leaves grew out in late May). The effects of TIBA on the growth of the new shoots and lateral branches were explored. After 5 months, treatments with 50, 100, and 150 mg L^{-1} TIBA decreased plant height by 13.5%, 22.9%, and 23.3%, respectively; TIBA at 50, 100, and 150 mg L^{-1} decreased the number of lateral branches by 18.0%, 18.6%, and 8.4%, respectively. Treatments with 100 mg L^{-1} TIBA increased the length of lateral branches by 34.3%; 50 and 100 mg L^{-1} TIBA increased the lateral branch diameter by 9.45% and 12.46%, respectively. After 2 months, 50 and 100 mg L^{-1} TIBA did not significantly affect photosynthetic rate in comparison with the control. Furthermore, 100 mg L^{-1} TIBA significantly increased spring tea yield by 19.3% in comparison with the control. In conclusion, the treatment of 100 mg L^{-1} TIBA can promote dwarfing, and formation of effective lateral branches, and spring tea yield. It is suggested to spray 'Longjing 43' tea plants with 100 mg L^{-1} TIBA solution after heavy pruning in summer (i.e., after two to three leaves grew out in late May).

1. Introduction

The regulation of shoot branching is an important aspect for crop improvement in agricultural production (Chen et al., 2016; Muhr et al., 2016). The regulation mechanism of shoot branching is a major and difficult problem in the research field of plant sciences. Plant hormones play critical roles in regulating shoot branching, and plant growth regulators are commonly applied to promote shoot branching and thus minimize the occurrence of pruning-caused diseases and reduce labor cost (Kender and Carpenter, 1972).

Auxin is a primary hormone that regulates lateral branching of plant shoots (Hayward et al., 2009; Shimizu-Sato et al., 2009). Auxin is exported from the primary shoot apex, transports basipetally, and suppresses axillary bud outgrowth (Shimizu-Sato et al., 2009; Waldie et al., 2014; Dierck et al., 2016). The auxin transport inhibitor *N*-1-naphthylphthalamic acid (NPA) relieves bud growth inhibition by indoleacetic acid (IAA) (Tamas et al., 1989). 2,3,5-Triiodobenzoic acid (TIBA) is another auxin transport inhibitor that inhibits polar auxin transport inside the plant. Exogenous TIBA application can promote axillary bud outgrowth (Shimizu-Sato et al., 2009), increase branch number, and inhibit the increase of plant height (Wu, 1983).

Tea plant (*Camellia sinensis*) is an important economic crop with bud leaves in the lateral branches as its product organ (Tounekti et al., 2013; Wu et al., 2015). A sufficient number of leaf buds is necessary to obtain high yield and quality (Yao and Wu, 1990). Tea plant presents obvious apical dominance, and artificially regulating the development of its lateral branches is necessary in tea production (Yue et al., 2012). Mature tea plants are regularly pruned to promote the growth of lateral branches. However, over-pruning of tea trees increases labor cost and generates excessive biomass waste.

The effect of exogenous TIBA application on shoot branching of tea plants remains unknown to date. Mature tea plants of 'Longjing 43', which is a premium clone for green tea and widely cultivated in China, were used in the current study. Here, the effects of TIBA treatment on dwarfing, shoot branching and yield of tea plant were studied, which can lay a foundation for further studying of the regulation mechanism of plant hormones on shoot branching of tea plant. The objectives of this study were to screen and develop the chemical compounds which

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can promote shoot branching of tea plant, in order to reduce the pruning frequency and finally form a standardized comprehensive management technology for the canopy of tea plant.

2. Materials and methods

2.1. Plant material, growth conditions, and TIBA treatment

Mature tea plants (*C. sinensis* L.) of the widely grown cv. Longjing 43 in China were used in this study. The experiment was conducted in the tea garden of the Tea Research Institute, Chinese Academy of Agricultural Sciences, Hangzhou, Zhejiang province, China (longitude 120° 100′E and latitude 30°140′N, 16 m above sea level).

Graded levels of TIBA working solution (50, 100, and 150 mg L^{-1}, J & K Scientific Ltd.) were prepared by adding solute in distilled H_2O and the mixture was stirred until dissolution. 0.1% (v/v) Tween 20 was added before spraying. Control (0 mg L^{-1}) tea bushes were simultaneously sprayed with distilled H_2O containing the same ratio of Tween 20. The experiment was laid out in a randomized complete-block design consisting of three concentrations of TIBA solution with three replications each treatment, and each replication spans an area of 10 m^2 consisting of 20 tea bushes. For the current study, tea plants that were heavily pruned in late April were chosen. When the apical buds and two to three leaves below the bud grew out in late May, the foliar portion of tea bushes was sprayed with different concentrations of TIBA solution.

2.2. Index measurement and methods

The plant height, the number of lateral buds/branches, as well as the lengths of new shoots, internodes, and lateral buds/branches were measured 1, 2, and 5 months after TIBA application. The diameter of lateral branches was measured using a vernier caliper. For each treatment, 15–30 new shoots, lateral buds/branches, or tea bushes were tested.

At 1 and 2 months after TIBA application, photosynthetic rate (Pn) was measured on the third fully expanded leaves in 12 tea bushes under each treatment by using an open-flow infrared gas analyzer adapted with light and temperature control systems (Li-COR 6400, Lincoln, NE, USA). The measurement was performed within the time period from 8:00 am to 11:00 a.m.

The leaf area of the third fully expanded leaves was measured using a portable leaf area meter (LI-3000C, LI-COR, Lincoln, NE, USA), and five to six replicates were measured for each treatment.

2.3. Statistical analysis

Data were analyzed using Statistica (SAS Institute, Inc., Cary, NC, USA, http://www.sas.com/). In each figure and Table 1, the differences of each index among different concentrations of TIBA application at each time point were analyzed using one-way ANOVA; if the ANOVA analysis was significant ($P < .05$), Duncan's multiple range test was used to detect significant differences between means.

Table 1
Effects of 2,3,5-Triiodobenzoic acid (TIBA) on the growth and yield of spring tea.

TIBA concentration (mg L^{-1})	Density of bud (bud/m^2)	Weight of 100 buds (g)	Yield (g/m^2)
0	2480 ± 240.2a	5.80 ± 0.136a	143.9 ± 16.86a
50	2633 ± 164.4ab	6.17 ± 1.425ab	166.8 ± 13.57ab
100	2983 ± 451.8b	7.37 ± 0.236b	171.7 ± 10.26b
150	2593 ± 208.4ab	6.20 ± 0.346a	163 ± 12.27ab

Data shown were means ± SE ($n = 6$). Letters indicate significant differences in each index among different concentration of TIBA application ($P < 0.05$, Duncan's multiple range test).

3. Results

3.1. Effect of TIBA on plant height and number of lateral buds/branches of tea plants

After 1 month, the height of tea plants treated with 50, 100, and 150 mg L^{-1} TIBA was shorted by 18.0%, 20.1%, and 22.5%, respectively. After 2 months, it was shorted by 21.2%, 25.8%, and 26.4%, respectively. After 5 months, it was shorted by 13.5%, 22.9%, and 23.3%, respectively (Fig. 1a). The TIBA application significantly inhibited the growth of tea plants at the three time points, and the inhibiting degree of the plant height induced by TIBA treatment increased with the increase in TIBA concentration. After 5 months, the height of tea plants treated with 50, 100, and 150 mg L^{-1} TIBA was shorted by 17.1, 29.0, and 29.5 cm, respectively, compared to the control.

Treatment with 50, 100, and 150 mg L^{-1} TIBA increased the number of lateral buds by 211%, 123%, and 292%, respectively after 1 month, compared to the control. After 2 months, no significant differences in the number of lateral buds were found among the four treatments. After 5 months, treatment with 50, 100, or 150 mg L^{-1} TIBA decreased the number of lateral branches by 18.0%, 18.6%, or 8.4%, respectively, in comparison with the control (Fig. 1b).

3.2. Effect of TIBA on growth of new shoots and lateral branches of tea plants

After 2 months, all three concentrations of TIBA inhibited new shoot growth and promoted lateral bud growth in comparison with the control (Fig. 2). As shown in Fig. 3a, the different concentrations of TIBA inhibited new shoot growth after 1, 2, and 5 months. No differences were noted between 100 and 150 mg L^{-1} TIBA after 1 month, and no differences were found among the three concentrations of TIBA after 2 months. After 5 months, 50, 100, and 150 mg L^{-1} TIBA application inhibited new shoot length by 23.6%, 23.6%, and 28.8% (i.e. 17.5, 17.5, and 21.4 cm), respectively, in comparison with the control. The result implies that the inhibition degree of TIBA on new shoot growth increased with the increase in TIBA concentration after 5 months.

After 1 month the three concentrations of TIBA did not affect the internode length of new shoots in comparison with the control. After 2 months, 50 mg L^{-1} TIBA inhibited the internode length by 22.9% (i.e. 0.98 cm) in comparison with the control. By contrast, no significant differences were noted among the three other treatments. After 5 months, 50, 100, and 150 mg L^{-1} TIBA all inhibited the internode length while no differences were noted among the three concentrations of TIBA (Fig. 3b). After 1 month, the lateral bud lengths of the tea plants treated with 100 and 150 mg L^{-1} TIBA were 73.5% and 99.1% (i.e. 1.8 and 2.5 cm) longer than that of the control plants, respectively. After 2 months, the lateral bud lengths treated with 150 mg L^{-1} TIBA significantly increased by 33.2% (i.e. 1.7 cm) compared with that of the control plants. At 5 months after TIBA application, the lateral branches of the tea plants treated with 100 mg L^{-1} TIBA were 34.3% (i.e. 2.4 cm) longer than the control plants (Fig. 3c). At the same period, 50 and 100 mg L^{-1} TIBA significantly increased the lateral branch diameter by 9.45% and 12.46% (i.e. 0.017 and 0.022 cm) in comparison with the control. On the contrary, no significant differences were noted between 150 mg L^{-1} TIBA and the control (Fig. 3d).

3.3. Effect of TIBA on growth of tea leaves

The leaf areas of the third fully expanded leaves were measured using a portable leaf area meter. As shown in Fig. 4a, after 1 and 2 months, TIBA spraying significantly decreased the leaf areas in comparison with those of the control plants, and no differences in inhibition degree were observed among three concentrations of TIBA treatment.