Effects of a Plant Immune Inducer on Cotton under Nanfan Conditions

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Abstract [**Objectives**] This study was conducted to mitigate the impact of delayed sowing on cotton production caused by typhoons, cotton-rice rotation and other factors. [**Methods**] Five representative elite breeding lines with excellent comprehensive performance were selected to investigate the effects of a plant immune inducer on the growth period, yield and other traits of cotton. Two treatments were established: foliar application of the plant immune inducer (at a dilution ratio of 1∶500) and foliar application of an equal volume of water as the control. [**Results**] Compared with the control, foliar application of the plant immune inducer shortened the interval from squaring to flowering by 2.79 d, and increased plant height, seed cotton weight per 25 bolls, lint yield, cottonseed yield and the number of harvested bolls by 20.36%, 19.17%, 20.95%, 18.44% and 25.45%, respectively. Foliar application of the plant immune inducer had no consistent significant effects on squaring, the number of fruit branches, lint percentage, boll opening or seed index of cotton. [**Conclusions**] The inducer acted mainly after the squaring stage in cotton and effectively accelerated late-stage development and increased yield. Among these effects, the marked increase in cottonseed yield is of particular significance for Nanfan breeding under delayed sowing.

Key words Cotton; Nanfan; Plant immune inducer; Growth period; Yield

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Cotton is an important economic crop[1] with a wide range of uses, including apparel and home textiles, edible oil and food, chemical engineering and the military industry, cosmetics and medicine, papermaking and currency printing, feed and microbial culture media, building materials, fuels and fertilizers, and ornamental horticulture[2]. Improved cotton varieties are the core of cotton production, and breeders have long striven to overcome various technical bottlenecks in the development of improved varieties, with the aim of achieving comprehensive improvement of cotton varieties. Among these efforts, Nanfan (off-season propagation in Hainan) for generation advancement is an important approach to shortening the breeding cycle[3]. Since cotton is a typical warm-season crop[4] and Hainan has unique tropical light and temperature conditions in winter and spring, cotton can be advanced from one generation per year to two or even three, thereby greatly shortening the time required to breed new varieties[5]. However, the annual sowing season for Nanfan coincides with the peak typhoon season. Typhoons can cause serious consequences such as delayed sowing, damage to or loss of seedlings, and even complete destruction of experimental materials[6]. Therefore, sowing is generally delayed until the typhoon-prone period ends. However, the most direct impact of delayed sowing is that by the time harvesting begins at the end of March of the following year, the cotton plants have failed to mature and the bolls remain unopened. In northern China, particularly in the Northwest Inland region, the main cotton-producing area, the window suitable for cotton growth is relatively fixed. If the Nanfan experimental materials are harvested late, sowing in the north will be delayed, making it impossible to continue planting there, and Nanfan would thus lose its significance[7]. Furthermore, owing to limited land resources and the ever-increasing demand for Nanfan, competition for land and the pressure of continuous cropping persist. For example, in cotton-rice rotation, sowing can begin only after the rice has been harvested and the land has dried and been prepared, which also leads to delayed sowing and consequently affects timely harvesting in the following year[8]. Therefore, sowing and harvesting in Nanfan are essentially carried out against a tight schedule. Breeders have made various efforts to alleviate this dilemma. Among multiple attempts, our team found that foliar application of a water-soluble fertilizer containing a plant immune inducer could promote cotton growth. In this study, elite lines developed by our team were used as experimental materials, and two treatments were established — foliar application of a water-soluble fertilizer containing a plant immune inducer and foliar application of water alone, to analyze the effects of the inducer on cotton growth and development. The study aimed to scientifically evaluate the effectiveness of the plant immune inducer in mitigating the adverse effects of delayed sowing during cotton Nanfan, and to provide a scientific basis for fertilizer application under late-sowing conditions.

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Materials and Methods

General situation of the experimental site

The experiment was conducted at the Yacheng Base, which belongs to the Institute of Cotton Research of Chinese Academy of Agricultural Sciences, and is located in Yazhou District, Sanya, Hainan Province. The experimental field had uniform soil fertility, flat terrain and good drainage and irrigation conditions, and had been planted with rice as the preceding crop. During the experimental period (November to April), the weather was stable with no natural disasters, meeting the basic requirements for cotton growth.

Experimental materials

The cotton materials tested were 25YC711, 25YC717, 25YC721, 25YC737 and 25YC743. They are non-homologous elite lines developed by our team, characterized by high fiber quality, high and stable yield, strong stress resistance and broad adaptability.

The water-soluble fertilizer containing the plant immune inducer was purchased from Sichuan Hongteng Biotechnology Co., Ltd. The product composition was as follows: organic matter ≥50 g/L, pH 6.0 – 8.0, and water-insoluble matter ≤50 g/L.

Experimental design

A randomized complete block design was adopted, consisting of a treatment group and a control group. Three replicates were established for each variety and each treatment, and each plot had an area of 20 m^2. Cotton was planted with plastic film mulching. The treatment and control groups were planted in isolation to avoid interference from spray drift.

To investigate the effects of the plant immune inducer on the growth and development of late-sown cotton in Nanfan, a water-soluble fertilizer containing the plant immune inducer was applied four times during the growth period. In the treatment group, the inducer was applied twice at the seedling stage (beginning at the three-leaf-one-heart stage), and once each at the squaring stage (when the squaring rate reached 30%) and at the flowering and boll-setting stage (full flowering stage). The inducer was diluted at a ratio of 1∶500 and applied with a knapsack electric sprayer to ensure uniform coverage of the leaves. The control group was sprayed with an equal volume of water at the same time. Other field management measures (irrigation, chemical regulation, and pest and disease control) were identical between the two groups.

Because the previous crop was rice and cotton is sensitive to waterlogging, sowing of Nanfan cotton could begin only after the rice had been harvested and the soil had dried. Sowing was carried out on November 10, 2025, which was relatively late. A film-mulched hole-punching planting pattern was adopted, with an actual average row spacing of 71 cm and a planting density of 73 350 plants per hectare. The dates of the squaring and flowering stages were subsequently recorded. On March 23, 2026, plant height, number of fruit branches, total number of bolls and number of open bolls were measured. Following the application of a defoliant, harvesting for seed testing was conducted on April 3, 2026.

Measurement items and methods

The growth period, agronomic traits, yield, and yield components of each experimental material were measured in three replicates according to the relevant standards, and all measurements were completed on the same day.

Observation of the growth process The squaring stage was defined as the date on which 50% of the plants in a plot had produced their first effective square, and the flowering stage as the date on which 50% of the plants had opened their first flower. The durations from sowing to squaring and from squaring to flowering were calculated.

Agronomic traits Plant height and the number of fruit branches of cotton were measured according to the *Cotton Agronomic Trait Investigation Standard* (NY/T 1300-2007). Plant height was defined as the vertical distance from the cotyledon node to the top of the plant (cm), and the total number of fruit branches referred to the total number of all fruit branches borne on the main stem (number).

Determination of yield and yield components Total number of bolls and total number of open bolls in a 3-m plot: A sample segment of 10 consecutive plants was randomly selected within each plot, and the total number of bolls, the number of open bolls and the plant spacing were recorded and converted to the corresponding values for a 3-m plot.

Seed cotton weight per 25 bolls and lint weight per 25 bolls: Twenty-five normal open bolls were randomly picked from each plot. After air-drying, the seed cotton weight (g) was measured. The lint weight (g) was measured following ginning.

Cottonseed yield per 25 bolls and lint percentage: Cottonseed yield per 25 bolls = Seed cotton weight per 25 bolls-Lint weight per 25 bolls; Lint percentage = Lint weight per 25 bolls/Seed cotton weight per 25 bolls × 100%.

Seed index: One hundred cottonseeds were randomly counted and weighed using an electronic balance with a precision of 0.01 g, and the weight (g) was recorded.

Data analysis

Excel 2016 was used for data processing and preliminary calculation, and DPS software for analysis of variance (ANOVA). Multiple comparisons were performed using Duncan's new multiple range test, with significance set at $P < 0.05$ and high significance at $P < 0.01$. Data were expressed as mean ± standard deviation. Different lowercase letters indicate significant differences among treatments ($P < 0.05$), and different uppercase letters indicate highly significant differences ($P < 0.01$).

Results and Analysis

Effects of plant immune inducer application on cotton growth and development

As shown in Table 1, only 25YC711 showed a significantly shorter duration from sowing to squaring in the treatment group than in the control group, whereas the other four lines showed no significant difference between treatments. These results indicate

that the plant immune inducer had no consistent effect in accelerating cotton squaring.

For four of the five lines, the duration from squaring to flowering was significantly shortened by foliar application of the plant immune inducer. The greatest reductions were observed in 25YC717 and 25YC743, both by 3.50 d, and the corresponding reductions for 25YC711 and 25YC721 were 1.83 and 2.12 d, respectively. Only 25YC737 showed no significant difference between the treatment and control groups. These results indicate that the plant immune inducer began to promote the reproductive growth of cotton after the squaring stage.

Table 1 Effects of plant immune inducer application on growth period of cotton

Determined indicators	Duration from sowing to squaring//d		Duration from squaring to flowering//d	
	Treatment group	Control group	Treatment group	Control group
25YC711	50.50 ±0.55 a	53.67 ±0.52 b	5.67 ±0.52 a	7.50 ±0.55 b
25YC717	57.67 ±0.58 a	58.33 ±0.58 a	6.75 ±0.50 a	10.25 ±0.50 b
25YC721	57.50 ±0.76 a	57.63 ±0.52 a	4.88 ±0.64 a	7.00 ±1.07 b
25YC737	57.33 ±0.82 a	57.50 ±0.55 a	6.33 ±1.21 a	7.33 ±1.75 a
25YC743	57.50 ±0.76 a	57.75 ±0.46 a	8.63 ±0.74 a	12.13 ±0.99 b

Data in the table are presented as mean ± standard deviation. Within the same row for the same material, the same letter indicates no significant difference between treatments ($P>0.05$); different lowercase letters indicate significant differences ($P<0.05$), and different uppercase letters indicate extremely significant differences ($P<0.01$). The same applies below.

Effects of plant immune inducer application on vegetative growth of cotton

As shown in Table 2, plant height in the treatment group was significantly greater than that in the control group for all tested lines. The greatest increase was recorded for 25YC721 (24.49% over the control). The increases in plant height for 25YC711 and 25YC717 also reached a highly significant level. These results indicate that foliar application of the plant immune inducer significantly promoted plant height in cotton.

With respect to the number of fruit branches, except for 25YC717, the differences between the treatment and control groups for the other lines did not reach significance, and in 25YC737 the number of fruit branches in the treatment group was even slightly lower than in the control group. These results indicate that foliar application of the plant immune inducer exerted no consistent regulatory effect on the development of cotton fruit branches.

Table 2 Effects of plant immune inducer application on vegetative growth of cotton

Determined indicators	Plant height//cm		Number of fruit branches	
	Treatment group	Control group	Treatment group	Control group
25YC711	86.00 ±1.00 A	71.67 ±3.51 B	9.67 ±0.58 a	9.33 ±1.15 a
25YC717	84.00 ±3.61 A	69.67 ±1.53 B	11.33 ±0.58 a	9.33 ±0.58 b
25YC721	83.00 ±3.46 A	66.67 ±1.53 B	9.67 ±0.58 a	9.67 ±0.58 a
25YC737	77.00 ±3.61 a	64.00 ±2.65 b	8.67 ±0.58 a	9.00 ±1.00 a
25YC743	71.00 ±6.56 a	61.00 ±3.61 a	7.33 ±0.58 a	6.33 ±0.58 a

Effects of plant immune inducer application on yield components of cotton

Boll number As shown in Table 3, the total number of bolls increased after foliar application of the plant immune inducer. For 25YC711 the increase in total boll number was significant, whereas for the other four lines the differences reached a highly significant level compared with their respective controls. These results indicate that foliar application of the plant immune inducer significantly promoted boll formation in cotton.

With respect to boll opening, the overall number of open bolls was low owing to the relatively late sowing date. Although the treatment group had more open bolls than the control group, the differences were not significant for all five tested lines, indicating that foliar application of the plant immune inducer had no significant promoting effect on boll opening in late-sown cotton under Nanfan conditions.

Table 3 Effects of plant immune inducer application on cotton bolls

Determined indicators	Total number of bolls		Number of open bolls	
	Treatment group	Control group	Treatment group	Control group
25YC711	249.31 ±26.52 a	199.33 ±9.52 b	12.53 ±1.08 a	9.34 ±7.05 a
25YC717	226.71 ±11.88 A	181.61 ±7.20 B	4.93 ±1.15 a	2.77 ±2.05 a
25YC721	232.70 ±4.92 A	186.23 ±5.71 B	5.30 ±4.59 a	3.40 ±3.17 a
25YC737	227.08 ±12.07 A	180.98 ±8.94 B	5.93 ±3.76 a	7.77 ±4.95 a
25YC743	171.09 ±10.49 A	134.82 ±10.55 B	5.89 ±1.21 a	1.90 ±3.28 a

Cotton yield As shown in Table 4, seed cotton yield increased significantly in all five lines after foliar application of the plant immune inducer compared with the control. The increases for 25YC711, 25YC717, 25YC737 and 25YC743 all reached a highly significant level, indicating that foliar application of the plant immune inducer markedly increased seed cotton yield.

Lint yield also increased significantly after foliar application of the plant immune inducer. Specifically, the increases in lint yield for 25YC737 and 25YC743 reached a highly significant level compared with the control. These results indicate that foliar application of the plant immune inducer could promote cotton fiber development and increase lint yield.

Table 4 Effects of plant immune inducer application on yield-related indicators of cotton

Determined indicators	Seed yield//g		Lint yield//g		Cotton lint percentage//%	
	Treatment group	Control group	Treatment group	Control group	Treatment group	Control group
25YC711	147.59 ±3.23 A	121.62 ±8.94 B	69.67 ±1.75 a	56.40 ±5.06 b	47.21 ±0.00 a	46.34 ±0.01 a
25YC717	140.33 ±5.32 A	119.12 ±2.81 B	64.06 ±3.04 a	54.06 ±0.97 b	46.06 ±0.01 a	46.32 ±0.00 a
25YC721	157.43 ±7.77 a	137.42 ±5.66 b	75.17 ±4.92 a	63.98 ±1.49 b	47.72 ±0.01 a	46.58 ±0.01 a
25YC737	170.39 ±3.66 A	146.98 ±7.21 B	81.58 ±2.05 A	68.16 ±2.81 B	47.88 ±0.01 a	46.39 ±0.01 b
25YC743	219.43 ±5.50 A	183.01 ±11.42 B	101.99 ±1.62 A	84.54 ±5.91 B	46.49 ±0.00 a	46.35 ±0.01 a

Determined indicators	Cottonseed yield//g		Seed index	
	Treatment group	Control group	Treatment group	Control group
25YC711	77.91 ±1.68 A	65.22 ±3.89 B	12.72 ±0.07 a	12.96 ±0.17 a
25YC717	76.26 ±2.36 A	65.07 ±1.87 B	12.91 ±0.17 a	12.69 ±0.02 a
25YC721	82.26 ±3.22 a	73.44 ±4.18 b	13.45 ±0.06 a	13.51 ±0.03 a
25YC737	88.81 ±2.13 a	78.81 ±4.52 b	14.08 ±0.06 a	14.01 ±0.12 a
25YC743	117.44 ±3.88 a	98.14 ±5.14 b	14.01 ±0.12 a	14.24 ±0.03 b

No consistent significant difference in lint percentage was observed between the treatment and control groups after foliar application of the plant immune inducer; only slight differences were found in individual lines, indicating that this trait remained stable overall. These results suggest that foliar application of the plant immune inducer had no obvious effect on cotton lint percentage.

After foliar application of the plant immune inducer, cottonseed yield increased significantly in all five tested lines compared with the control, and the increases for 25YC711 and 25YC717 reached a highly significant level. However, no consistent significant differences were observed in seed index, indicating that the inducer increased seed yield rather than seed size.

Discussion and Conclusion

Foliar application of the plant immune inducer promoted the overall growth and development of cotton under Nanfan conditions. In terms of the growth period, although the inducer was applied twice at the seedling stage, no consistent promoting effect on squaring was observed. The effect of the plant immune inducer on the reproductive growth of cotton began to manifest after the squaring stage, and the interval from squaring to flowering was shortened, so that flowering occurred earlier overall. From the perspective of vegetative growth, foliar application of the inducer promoted vegetative growth and significantly increased plant height, but had no consistent effect on the number of fruit branches. With respect to yield, foliar application of the plant immune inducer significantly increased seed cotton yield, lint yield and cottonseed yield, with some of these effects reaching a highly significant level. However, it had no consistent significant effect on the lint percentage of any of the tested lines. In summary, the main effect of the inducer on cotton was manifested in greater plant height, earlier flowering and higher yield, while its effect on the growth period was mainly observed after the squaring stage. The number of fruit branches, lint percentage and seed index were not consistently affected during this process.

This study was conducted under delayed-sowing conditions, with reference to previous studies. Throughout the growth period, uncontrollable factors such as natural disasters and severe pest or disease outbreaks were absent[9]. The tested materials were new upland cotton breeding lines developed by our team, which perform excellently and are representative of production in terms of yield, quality and resistance. This study aimed to investigate the regulatory effects of the plant immune inducer on cotton growth and development and its potential value for Nanfan cotton breeding. It was anticipated that foliar application of the plant immune inducer would promote cotton growth and development, thereby avoiding delayed sowing in Xinjiang in the following year. The results indicate that foliar application of the plant immune inducer is of positive value for Nanfan cotton breeding. In this study, the earlier flowering stage promoted earlier boll setting and earlier boll opening, which could compensate to some extent for the adverse effects of late sowing[10]. The pursuit of high yield is an enduring theme for breeders[11]. The overall improvement in yield-related traits following foliar application of the plant immune inducer has positive implications for both cotton production and breeding. Moreover, plant height increased consistently with the yield-related traits, reflecting the synergistic promotion of vegetative and reproductive growth by the inducer. In this study, the squaring stage and the number of open bolls remained relatively stable, and foliar application of the plant immune inducer exerted no consistent

significant effect on them. Although individual lines showed differences, none reached a highly significant level. It is therefore speculated that the inducer promoted reproductive growth mainly during the period from squaring to boll opening. During this period cotton grows rapidly, leaf function is fully established and metabolism is vigorous, making it the optimal window for foliar fertilization[12]. No consistent significant differences were observed in the number of fruit branches, lint percentage or seed index between the sprayed and control groups. This may be because these traits are influenced by the environment but, to a greater extent, determined by the inherent characteristics of the varieties themselves, whereas the inducer does not disrupt the expression of the corresponding genetic traits. In previous studies, cotton lint percentage has been shown to be a yield component strongly influenced by varietal characteristics and only slightly affected by the environment[13]. By contrast, the number of fruit branches and the seed index of cotton varieties result from the combined effects of heredity and environment. Under conditions of abundant water and fertilizer supply, stress, or cultivation in different ecological regions, the genetic potential for fruit branch number and seed index may be activated or suppressed. However, in field production, the effective number of fruit branches and the seed index of cotton are generally relatively stable. Different topping measures can affect the number of fruit branches in cotton, but the effective number of fruit branches still shows a relatively stable trend[14-15]. This study was conducted in a Nanfan field with the aim of compensating for the adverse effects of late maturity caused by delayed sowing. The application rate of the plant immune inducer was determined according to actual field production conditions and the product instructions. The applied dose may not have reached the level required to alter genetic traits, or the formulation itself may not be designed to modify the genetic characteristics related to the number of fruit branches and the seed index. This remains to be further investigated.

In summary, foliar application of the plant immune inducer exerted positive effects during Nanfan generation advancement of cotton: it effectively promoted growth and development and increased yield. In particular, foliar application of the plant immune inducer at the squaring stage and at the flowering and boll-setting stage could effectively promote flowering and boll formation, thereby compensating for the negative effects caused by delayed sowing. Among the yield-related traits examined, seed cotton yield, lint yield and cottonseed yield all increased significantly under the effect of the plant immune inducer, whereas lint percentage, seed index and the number of open bolls were not significantly affected. In particular, the increase in cottonseed yield obtained under late-sowing conditions in Nanfan is of great significance for breeders in multiplying experimental materials[16-17].

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