



**Full Length Article**

## Effects of Hormone Soaking on Stem Physiology and Cell Structure of Carrot Seedlings

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### Abstract

Exogenous application of hormones can affect crop growth by regulating stem growth. To investigate the effects of exogenous hormones on stem physiology and cell structure of carrot (*Daucus carota* L. var. *sativa* DC) seedling, the effects of exogenous GA<sub>3</sub> and IAA on the stem growth of carrot seedlings were explored in this study. In particular, the following seven treatments were applied: water control (CK), 50, 150 and 250 mg·L<sup>-1</sup> GA<sub>3</sub>, and 50, 150 and 250 mg·L<sup>-1</sup> IAA. Endogenous hormone contents, seedling morphological indexes of the stem parts, and the vertical and horizontal cellular microstructures of the stems were evaluated. A concentration of 250 mg·L<sup>-1</sup> GA<sub>3</sub> (hereinafter called GA<sub>3</sub>) resulted in the longest carrot stems and 150 mg·L<sup>-1</sup> IAA (hereinafter called IAA) resulted in the highest root-to-shoot ratio. Therefore, these GA<sub>3</sub> and IAA treatments were used for subsequent analyses of endogenous hormone contents in carrot stems and cellular properties, since these two treatments clearly reflected the mechanism of action of exogenous hormones on stems. The endogenous GA<sub>3</sub>, ABA, and IAA contents were highest in the GA<sub>3</sub> treatment (*i.e.*, 12.15 ng/g FW, 151.74 ng/g FW and 77.75 ng/g FW) and were all greater than those for IAA treatment. The length and width of longitudinal single cells were largest for GA<sub>3</sub>. In the lateral cell structure of the stem, the cross-sectional diameter of the vascular bundle and the cross-sectional diameter of the stem decreased in the order of IAA > CK > GA<sub>3</sub>, but the diameter of the vascular bundle was equal for GA<sub>3</sub> and IAA. GA<sub>3</sub> and IAA total cross-sectional diameter ratio was 0.36, which was significantly larger than that for CK. Both 250 mg·L<sup>-1</sup> GA<sub>3</sub> and 150 mg·L<sup>-1</sup> IAA significantly changed the endogenous hormone content of the cylindrical stem part, and the morphology of the vertical and horizontal cell structure of the stem also changed significantly. Therefore, a certain concentration of exogenous hormones can pass. Changing the content of endogenous hormones in carrot seedlings changes the structure of certain cells in the stem, thereby affecting the morphology of the stem. © 2020 Friends Science Publishers

**Keywords:** *Daucus carota* L. var. *sativa* DC; Carrot seedling; Endogenous hormone; Cellular structure; Stem growth

### Introduction

*Daucus carota* L. var. *sativa* DC is a variant of wild carrot (*D. carota* L. var. *carota*), belonging to Apiaceae. It has a long history of cultivation, with extensive planting areas worldwide. Carrot seeds are small with leathery skin containing volatile oil, resulting in poor water permeability (Zhuang *et al.* 2006). Carrot is rich in vitamin A and β carotene, which could effectively prevent night blindness, delay aging, and strengthen immunity (Imsic *et al.* 2010). Seed soaking with exogenous hormones (such as GA<sub>3</sub> and IAA) is a common approach in production of crops, such as tomato (Luo *et al.* 2015), tall fescue (Xu *et al.* 2008), lettuce (Zagorski and Lcwak 1983) and radish (Jabir *et al.* 2017). Soaking seeds with exogenous hormones has a significant effect on the development of organs, such as the roots, stems, and leaves of the plant seedlings (Pill and Finch-Savage 2008). Exogenous hormones at certain concentrations can promote the growth of plant stems (Barratt and Davies

1997), affecting the performance of crops, such as cucumber (Qian *et al.* 2018), corn (de Souza and MacAdam 2001) and pea (Ross *et al.* 2000). Exogenous hormone could promote the growth of plant stems by increasing the number and length of cells or by altering the extensibility of the cell wall (Wolbang *et al.* 2004). For example, induced by exogenous GA<sub>3</sub>, cell cycle progression in rice stems could be shortened and the number of cell divisions could be increased, thereby affecting the morphology of rice (Wang *et al.* 2013). Furthermore, exogenous GA<sub>3</sub> not only promotes the growth of stalks by promoting the division of pea cells, but also affects the endogenous IAA content in metaphase cells of pea seedlings (Barratt and Davies 1997). However, the rapid growth rate of plant seedling stems could cause vein growth and lodging, which is not conducive to the growth and cultivation of seedlings. Therefore, it is important to study the mechanisms by which exogenous hormones promote stem growth.

In carrot production, seed soaking in exogenous GA<sub>3</sub> or

IAA is a common method (Lee 1990). Hormone treatment not only promotes the germination of carrot seeds, but also changes the morphology of plants by promoting the growth of seedlings (Barratt and Davies 1997). Changes in plant morphology are mainly due to changes in the cellular structure, which are determined, to some degree, by endogenous hormones in the plant (Li *et al.* 2019). Therefore, with the treatment of carrots with exogenous hormones, the relationship between exogenous hormones and the content of endogenous hormones, cell structure and morphology of carrot seedlings was explored. The effects of soaking with exogenous hormones on the growth and development of stems in carrot seedlings were further explored. The mechanism of action of hormones on carrot seedlings is to select the most suitable exogenous hormone treatment for carrot soaking, which provides a certain theoretical basis for obtaining high-yield and high-quality carrot seedlings in production. It is of great significance to improve the growth and development of carrot seedlings in production. Variation in the external morphology of plants is closely related to variation in physiological indexes. Morphological differences can better reflect the internal physiological and cellular changes of carrots (Pill and Finch-Savage 1988). Three concentrations of GA<sub>3</sub> and IAA were evaluated in this experiment. Accordingly, the GA<sub>3</sub> and IAA treatments with the largest morphological differences were first screened, and then the endogenous hormone contents were measured and cytological observations were performed to better reflect the physiology and cellular characteristics.

## Materials and Methods

### Materials and treatments

As test materials, Yibo Dadi carrot (*D. carota* L. var. *sativa* DC) seeds were obtained from Tongdeli Seed Industry Co., Ltd., Qingdao, China. This study was performed at the Vegetable Research Institute of Fujian Agriculture and Forestry University from January 1–23, 2019. Test seeds were divided into the following treatment groups: distilled water (CK), three gibberellic acid (GA<sub>3</sub>; 50, 150 and 250 mg·L<sup>-1</sup>) and three indole acetic acid (IAA; 50, 150 and 250 mg·L<sup>-1</sup>). After soaking seeds for 12 h, they were germinated at 30°C for 24 h and sown in the seedling substrate. Each treatment was repeated 3 times, and 100 carrot seeds were used for each repetition. The first day was seeded on the nursery substrate, and the test period was 20 days.

### Morphological indices of the carrot stem

The test period was 20 days. Three seedlings with the same growth potential were selected for each treatment to determine the plant attributes. The plants were cleaned, and the stem diameter, stem height, and plant height were measured using a Vernier caliper. In addition, 30 strains were randomly selected from each treatment, and the plants were cleaned. The fresh weights of the aboveground and

underground parts were measured, and the plants were placed in an oven (Model: DHG-9141A; Shanghai Jinghong Experimental Equipment Co., Ltd., Shanghai, China) and dried at 90°C for 1.5 h. Then, the dry weights of the aboveground and underground parts were determined.

The aspect ratio of the stem is defined as the ratio of the stem length to stem diameter; it reflects the growth of plant shoots. The ratio of the root to shoot is the ratio of the dry weight of the lower part to the dry weight of the shoot, reflecting the growth of the whole plant (Chinchilla-Ramírez *et al.* 2017). The stem aspect ratio was estimated according to the following formula:

$$\text{Stem aspect ratio (\%)} = (\text{Stem length/stem diameter}) \times 100$$

Where the stem diameter is measured at 1/2 the length of the stem

### Determination of endogenous hormone contents in carrot seedling stems

Enzyme-linked immunosorbent assays were used to determine endogenous hormone contents in carrot straw (O'Kennedy *et al.* 1990). A 0.2 g carrot stem segment was fully ground with liquid nitrogen, supplemented with 2 mL of extraction solution (containing 1 mmol/L BHT (Di-tert-butyl p-cresol) and 80% methanol mixture), ground in an ice bath, and maintained at 4°C for 4 h. After centrifugation at 1150 g for 8 min, the supernatant was obtained and 1 mL of extraction solution was added. The above steps were repeated twice, and the supernatants were combined. The supernatant was treated with a C18 solid-phase extraction column (Beijing Dikema Technology Co., Ltd., Beijing, China), and the samples were transferred to a 10 mL centrifuge tube. The methanol in the extraction solution was removed using a Cetivap 78120-03 centrifugal concentration dryer (Labconco, Kansas City, MO, USA). The volume was fixed to 0.4 mL of the sample diluent (including 1% Tween-20 and 1 g/L gelatin in PBS with pH 7.5). An IL-10 ELISA Kit (Thermo Fisher Scientific) and enzyme-linked immunosorbent spectrophotometer were used to measure the concentrations of the standards and the OD values at 490 nm for each sample. The concentrations of different endogenous hormones in each treatment were calculated by drawing the standard curve.

### Microscopic observations of whole plants and stem sections

On day 20 of the experiment, images of whole carrot seedlings were obtained using a 20-megapixel digital camera (Nikon, Tokyo, Japan; COOLPIX S2800). For microscopic observations of longitudinal sections of carrot stems, on day 20 of the experiment, 3 carrot seedlings with uniform growth were randomly selected from each treatment, and 0.5 cm samples were obtained at 1/2 the length of the stem. Then, 5 g of agar was dissolved in 100 mL of water. After melting at

a high temperature in a microwave oven (Shanghai Matsushita Microwave Co., Ltd.; NN-DS1100), the sample was poured into a culture dish with a diameter of 35 mm, tiled, fixed in 5% agar, and cooled to room temperature. The fixed stems were sectioned longitudinally by the freehand-sectioning method, stained with 0.04 mg of LPI (Hai De Biotechnology Co., Ltd., Beijing, China) dye for 10 min, washed with water to remove excess dye and dried. The longitudinal cell structure of the carrot stem was observed using a confocal microscope (FV1200, Olympus, Japan) at a wavelength of 535 nm.

For microscopic observations of transverse sections of carrot seedlings stems. On the last day of the experiment, 3 strains were randomly selected from each treatment. The same stem segments were cut, fixed with 5% agarose, and then sectioned using an oscillating slicer (Leica; VGA3000S) longitudinally and horizontally. The transverse sections were observed using an inverted fluorescence microscope (Leica DMI8). Then, the cross-sectional diameter of vascular bundle, stem cross-sectional diameter and ratio between vascular tube diameter and the total cross-sectional diameter of carrot stem cross-sections were measured.

### Data analysis

All indicators were estimated based on 3 biological replicates and technical replicates. Data were analyzed by LSD multiple comparison tests implemented in DPS7.5. The significance level was set to  $P < 0.05$ .

## Results

### Influence hormones on morphological indices of carrot stem segments

On day 20 of the test period, the stem length, plant height, and stem aspect ratio of GA<sub>3</sub>-treated carrot seedlings were 30.47, 74.21 and 55.37, respectively, and were significantly greater than those of other treatments, indicating that 250 mg·L<sup>-1</sup> GA<sub>3</sub> had the greatest impact on carrot morphology (Fig. 1). The main manifestation was an increase in the growth of stems; there were no significant differences in stem aspect ratio between CK, 50, 150 and 50 mg·L<sup>-1</sup> IAA treatments. Except for 50 mg·L<sup>-1</sup> GA<sub>3</sub>, the root-to-shoot ratios of other treatments were significantly greater than CK. The IAA treatment had the largest root-to-crown ratio. A concentration of 150 mg·L<sup>-1</sup> IAA had the greatest effect on the growth of carrot seedlings. Different concentrations of exogenous GA<sub>3</sub> and IAA affected plant morphology. When the exogenous GA<sub>3</sub> concentration was 250 mg·L<sup>-1</sup>, the growth of stems was greatest. When the concentration of IAA was 150 mg·L<sup>-1</sup>, the root-shoot ratio of carrots was highest. Therefore, the endogenous hormone contents in carrot stems were further evaluated in the CK, GA<sub>3</sub>, and IAA treatments. Observations at the hormone and cellular levels explained the effects of exogenous hormones on the growth of carrot stems.

### Determination of endogenous hormone contents in carrot seedling stems

The contents of the endogenous hormones GA<sub>3</sub>, ABA, and IAA in the carrot seedling stems for the GA<sub>3</sub> treatment was significantly higher than those for the CK treatment (see Table 1), *i.e.* 12.15, 151.74 and 77.75 ng/g FW, respectively. The ZR content for GA<sub>3</sub> treatment was 11.21 ng/g FW, which was second only to that of the CK treatment and was not significantly different from that of the CK treatment, indicating that 250 mg·L<sup>-1</sup> GA<sub>3</sub> applied to carrot seedling stems mainly increased endogenous GA<sub>3</sub>, ABA and IAA. It is possible that 250 mg·L<sup>-1</sup> exogenous GA<sub>3</sub> promoted stem elongation at the metabolic level by increasing the contents of endogenous GA<sub>3</sub>, ABA and IAA.

The contents of endogenous GA<sub>3</sub> and IAA in the carrot seedling stems of the IAA treatment were second only to those of the GA<sub>3</sub> treatment and were significantly higher than those of the CK treatment. The contents of endogenous ABA and ZR were the lowest. The contents of endogenous GA<sub>3</sub>, ABA, IAA, and ZR were 11.60, 97.52, 68.31 and 7.41 ng/g FW, respectively, indicating that the primary effects of 150 mg·L<sup>-1</sup> IAA at the metabolic level in carrot seedling stems were the increases in endogenous GA<sub>3</sub> and IAA and decreases in endogenous ABA and ZR.

Exogenous GA<sub>3</sub> and IAA can affect plant morphology by affecting the cellular structure (Barratt and Davies 1997; Kim *et al.* 2006). In this experiment, 250 mg·L<sup>-1</sup> GA<sub>3</sub> and 150 mg·L<sup>-1</sup> IAA significantly changed the endogenous hormone content in the stem parts of carrot seedlings; accordingly, it could be speculated that exogenous hormones at certain concentrations could alter the cellular structure of the stem by changing the endogenous hormone contents of carrot seedlings, which in turn could affect seedling morphology (Table 2).

### Microscopic observations of whole plants and stem sections of carrot seedlings

On day 20 of the experiment, the morphology of carrot seedlings varied significantly. IAA treatment had the highest plant height, GA<sub>3</sub> treatment was the second, and CK treatment was the smallest. The stem lengths of these three treatments were A (17.80 mm), B (30.47 mm) and C (21.79 mm), it suggested soaking with 250 mg·L<sup>-1</sup> GA<sub>3</sub> and 150 mg·L<sup>-1</sup> IAA promotes the growth of both the carrot plant and stem. In particular, 250 mg·L<sup>-1</sup> GA<sub>3</sub> had the most obvious effect on the seedling stem, and 150 mg·L<sup>-1</sup> IAA had the most obvious effect on the whole plant height (Table 3).

The longitudinal cell structure of carrot seedling stems and phloem sites were observed, the longitudinal cells of the stems were largest in 50 mg·L<sup>-1</sup> GA<sub>3</sub> treatment, with significant increases in the length and width of single cells, followed by the IAA treatment and CK treatment. This suggests that 250 mg·L<sup>-1</sup> GA<sub>3</sub> can promote stem growth by promoting the broadening and lengthening of stem longitudinal cells, and 150 mg·L<sup>-1</sup> IAA could also promote

**Table 1:** Endogenous hormone contents in the aboveground part of carrots under different seed-soaking treatments

Treatment	GA <sub>3</sub> content (ng/g FW)	ABA content (ng/g FW)	IAA content (ng/g FW)	ZR content (ng/g FW)
CK	9.43±1.20 c	102.39±4b	48.36±3.82 c	11.53±1.31 a
250 mg·L <sup>-1</sup> GA <sub>3</sub>	12.15±0.15 a	151.74±5a	77.75±2.21 a	11.21±1.35 b
150 mg·L <sup>-1</sup> IAA	11.60±0.24 b	97.52±3 b	68.31±4.10 b	7.41±1.27 c

Note: Data were analyzed using the LSD multiple comparison test implemented in DPS7.5. Different letters indicate significant differences ( $p < 0.05$ )

**Table 2:** Morphological indexes of carrots under different seed soaking treatments

Treatment	Stem diameter (mm)	Stem length (mm)	Plant height (mm)	Root dry height (mg)	Above ground dry weight (mg)	Root: shoot ratio	Aspect ratio
CK	0.53±0.03c	17.80±0.90e	61.70±1.47d	1.02±0.08d	5.35±0.31b	0.19±0.005e	33.79±0.21c
50 mg·L <sup>-1</sup> GA <sub>3</sub>	0.53±0.01c	23.48±1.22bc	65.32±1.31c	1.23±0.02b	6.14±0.38a	0.20±0.010de	44.35±3.12b
150 mg·L <sup>-1</sup> GA <sub>3</sub>	0.52±0.02c	25.29±1.31b	70.19±1.23b	1.25±0.03ab	5.97±0.65a	0.21±0.018cd	48.40±3.80b
250 mg·L <sup>-1</sup> GA <sub>3</sub>	0.55±0.03c	30.47±2.14a	74.21±1.15a	1.32±0.05a	6.02±0.29a	0.22±0.007c	55.37±0.87a
50 mg·L <sup>-1</sup> IAA	0.62±0.01ab	20.78±1.49d	62.27±0.75d	1.05±0.05cd	4.19±0.24c	0.25±0.004b	33.50±1.89c
150 mg·L <sup>-1</sup> IAA	0.64±0.02a	21.79±1.50cd	62.46±1.05d	1.11±0.01c	3.96±0.06c	0.28±0.005a	34.12±3.42c
250 mg·L <sup>-1</sup> IAA	0.60±0.02b	20.17±1.76de	61.99±0.90d	1.04±0.08cd	4.33±0.28c	0.24±0.008b	33.87±3.84c

Note: Data were analyzed using the LSD multiple comparison test using DPS7.5. Different letters indicate significant differences ( $p < 0.05$ )

**Table 3:** Cross-sectional indicators of carrot stem segments after different soaking treatments

Treatment	Cross-sectional diameter of vascular bundle (μm)	Stem cross-sectional diameter (μm)	Ratio between vascular tube diameter and the total cross-sectional diameter
CK	200.0±10b	590.0±10 b	0.340±0.011 b
250 mg·L <sup>-1</sup> GA <sub>3</sub>	180.0±5 c	500.5±10c	0.36±0.003 a
150 mg·L <sup>-1</sup> IAA	235.0±10 a	640.0±15a	0.36±0.004 a

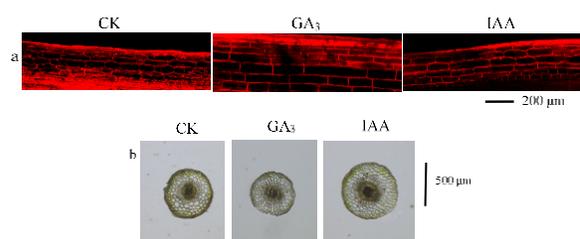
Note: Data were analyzed using the LSD multiple comparison test implemented in DPS7.5. Different letters indicate significant differences ( $p < 0.05$ )



**Fig. 1:** Carrot seedlings on day 20 of hormone treatment  
Note: A, B, and C indicate the stem length of CK, GA<sub>3</sub>, and IAA, respectively

the broadening and growth of stem longitudinal cells but the effect was weaker than that of 250 mg·L<sup>-1</sup> GA<sub>3</sub>.

GA<sub>3</sub> and IAA at certain concentrations can promote the growth and differentiation of vascular bundles in stems of plants (Fukaki *et al.* 2002; Xu *et al.* 2008). Fig. 2 depicts the cross-sectional diameters of the stem segments of carrot seedlings. The vascular bundle cross-sectional diameter, stem cross-sectional diameter, and vascular bundle diameter of the GA<sub>3</sub> treatment accounted for the largest proportion of the total cross-sectional diameter (235 and 640 μm, and 0.36, respectively) and the stem cross-sectional diameter of the IAA treatment was significantly larger than those of the CK and GA<sub>3</sub> treatments, indicating that 150 mg·L<sup>-1</sup> IAA has the most significant effect on stem thickening and vascular bundle enlargement. The vascular bundle cross-sectional diameter and the stem cross-sectional diameter of the GA<sub>3</sub> treatment were 180 and 500 μm, respectively, which were significantly smaller than those of the CK treatment,



**Fig. 2:** Morphological observations of cross-cut fragments of carrot stems treated with different hormones

Note: In panel a, from left to right, the transected cell structure of the stems of carrots treated with CK, GA<sub>3</sub>, and IAA are shown. In panel b, from left to right, the longitudinal cell structure of the stems of carrots treated with CK, GA<sub>3</sub>, and IAA are shown

suggesting that 250 mg·L<sup>-1</sup> GA<sub>3</sub> restricts the vascular bundle and stem diameters, resulting in a relatively thin carrot stem. However, the ratio between the vascular bundle diameter and the total cross-sectional diameter was 0.36, equal to that of the IAA treatment, and the diameters for both the GA<sub>3</sub> and IAA treatments were significantly larger than those of the CK treatment, suggesting that 250 mg·L<sup>-1</sup> GA<sub>3</sub> could make the carrot seedling stem thinner. However, the ratio between the vascular bundle diameter and total cross-sectional diameter was similar to that of the 150 mg·L<sup>-1</sup> IAA treatment (Table 3).

## Discussion

Huang *et al.* (2017) showed that soaking corn seeds with the exogenous plant growth regulator SPD can significantly

increase the endogenous SPD, gibberellin, and ethylene contents, and reduce the concentration of ABA in embryos, thus affecting seed vigor, increasing the germination index, vigor index, bud height, and root dry weight of corn seeds relative to those of the control, and promoting the growth of corn seedlings and dry matter. Furthermore,  $50 \mu\text{mol}\cdot\text{L}^{-1}$  IAA and  $50 \mu\text{mol}\cdot\text{L}^{-1}$  GA<sub>3</sub> can promote the elongation and thickening of the stem of *Narcissus* by changing the morphology of stem cells (Krug *et al.* 2006), indicating that the effect of exogenous hormones on plants may be mediated by changes in the cell structure, endogenous hormone contents, or physiological indexes. In this experiment, different concentrations of GA<sub>3</sub> and IAA had different effects on carrot stems. When the GA<sub>3</sub> concentration was  $250 \text{ mg}\cdot\text{L}^{-1}$ , carrot stems were the longest among all treatments, and when the IAA concentration was  $150 \text{ mg}\cdot\text{L}^{-1}$ , the carrot root: shoot ratio was the largest. These results showed that different concentrations of exogenous hormones had different effects on carrot stems, and GA<sub>3</sub> and IAA had the greatest effect on carrot seedling morphology. There was a positive correlation between the differences in plants and the differences in morphology, and the differences in external morphology were better able to reflect the differences in the physiological indicators of plants (Zotz *et al.* 2012). Therefore, the GA<sub>3</sub> and IAA treatments were used for further analyses of the endogenous hormone content in carrot stems and cytological observations to explore the relationship between exogenous and endogenous hormone contents, cell structure, and morphological compositions of carrot seedlings.

Certain concentration of exogenous GA<sub>3</sub> can induce hydrolase production in seed embryos, decompose storage macromolecules into small molecules, promote the maturation of embryos and the growth and development of plant seedlings, regulate plant endogenous hormones, promote cell division and tissue differentiation, and accelerate the growth process (Eriksson 2006). Wang *et al.* (2016) showed that exogenous GA<sub>3</sub> treatment could significantly reduce the content of endogenous ABA in wheat seeds during germination and seedling growth, while the amylase activity of wheat seeds was stronger and endosperm structure was fuller. In this experiment, the contents of the endogenous hormones GA<sub>3</sub>, ABA, and IAA in the stems of GA<sub>3</sub>-treated carrot seedlings were significantly higher than those in the CK treatment, indicating that  $250 \text{ mg}\cdot\text{L}^{-1}$  exogenous GA<sub>3</sub> promotes stem elongation at the metabolic level via increases in the contents of endogenous GA<sub>3</sub>, ABA, and IAA. The contents of endogenous GA<sub>3</sub> and IAA in the stems of IAA -treated carrot seedlings were second only to those of the GA<sub>3</sub> treatment and were significantly higher than those of the CK treatment. This indicated that the effect of  $150 \text{ mg}\cdot\text{L}^{-1}$  exogenous IAA on carrot seedling stems at the metabolic level was mainly related to an increase of endogenous GA<sub>3</sub> and IAA and reductions of endogenous ABA and ZR. Qian *et al.* (2018) reported that the elongation of cucumber stem segments is

very sensitive to the applied GA<sub>3</sub>, which could promote growth by changing the content of endogenous GA<sub>3</sub>. Another study showed that IAA at a certain concentration can promote the biosynthesis of GA<sub>1</sub> in the elongated internode of peas (Ross *et al.* 2000), thereby affecting the size of internode cells and promoting internodal elongation. This further indicated that the application of exogenous hormones could change the structure of cells by changing the content of endogenous hormones, affecting plant morphology. In this experiment, GA<sub>3</sub> and IAA significantly changed the endogenous hormone contents in seedling stems, and can also alter the content of endogenous hormones in the stems of carrot seedlings, which may have an effect on the cellular structure and morphology (Table 2).

The thickening of stems and enlargement of vascular bundles are conducive to the transport of water and nutrients. However, the growth of stems tends to result in vein growth and seedling lodging, which is unfavorable for the absorption of nutrients and the growth of shoots. Therefore, it is important to study the effects of exogenous hormones on plant stems (de Souza and MacAdam 2001; Krug *et al.* 2006). In this experiment, based on whole plant analyses and longitudinal and horizontal sections of the stem, the heights of the carrot were ranked: IAA > GA<sub>3</sub> > CK, and the stem lengths were ranked GA<sub>3</sub> > IAA > CK, indicating that both  $250 \text{ mg}\cdot\text{L}^{-1}$  GA<sub>3</sub> and  $150 \text{ mg}\cdot\text{L}^{-1}$  IAA can increase the plant height and stem length of carrots, but the effect of GA<sub>3</sub> was much stronger than that of IAA. Therefore, it is possible that GA<sub>3</sub> may cause the growth rate of stems to be too fast during the development of carrot seedlings, resulting in vein growth. However, chlormequat at certain concentrations can dwarf seedlings during the seedling stage, increasing robustness (Jiang *et al.* 2010). Therefore, in the actual production of carrots, seed soaking with GA<sub>3</sub> could result in a high germination rate, and chlormequat can be applied at the seedling stage to alleviate the effect of GA<sub>3</sub>. The specific approach requires further studies. GA<sub>3</sub> could promote the growth of plants by promoting cell division (Sauter *et al.* 1995; Wang *et al.* 2015). For example, induced by GA<sub>3</sub>, the cell cycle in rice stems could be shortened and the number of fission events could be increased, which resulted in continuous rice stem growth (Mao *et al.* 2018), while exogenous IAA affected cell elongation by adjusting the extensibility of cell walls (Barratt and Davies 1997). In this study, stem longitudinal cells were largest with GA<sub>3</sub> treatment, with an obvious increase of the length and width of single cells, following by the IAA treatment, indicating that GA<sub>3</sub> could promote stem growth by promoting the broadening and growth of longitudinal cells. The cross-section diameter of vascular bundle and stem cross-sectional diameter of the GA<sub>3</sub> treatment were the smallest, at  $180 \mu\text{m}$  and  $500 \mu\text{m}$ , respectively. However, the vascular bundle diameter accounts for the same proportion of the total cross-sectional diameter as that of the IAA treatment, indicating that the GA<sub>3</sub> treatment makes stems thinner. However, with respect to the cell structure, the ratio of the vascular bundle

area to the total cross-sectional area was the same for both GA<sub>3</sub> and IAA.

## Conclusion

Except for above ground dry weight, all morphological indicators of CK treatment in this experiment were smaller than other treatments, indicating that exogenous GA<sub>3</sub> and IAA soaking carrots had certain positive effects on carrot seedlings. In particular, 250 mg·L<sup>-1</sup> GA<sub>3</sub> and 150 mg·L<sup>-1</sup> IAA had the greatest effects on the morphological properties of carrot seedlings. Our results indicated that carrot stems are longest after treatment with 250 mg·L<sup>-1</sup> GA<sub>3</sub>, and the ratio of carrot roots to shoots was the largest in response to 150 mg·L<sup>-1</sup> IAA. Therefore, exogenous hormones could change the shape of stem cells by affecting the content of endogenous hormones in carrot seedlings, increase the size single cells in stems, promote the growth of stems, and affect the appearance of carrots. Accordingly, 250 mg·L<sup>-1</sup> GA<sub>3</sub> and 150 mg·L<sup>-1</sup> IAA were optimal for soaking carrots. The observed changes in hormone contents and cellular morphology during the seedling stage provide a basis for the selection of appropriate concentrations of exogenous hormones for large-scale carrot production.

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