CA during Chilling Storage Suppresses the Induction of Daughter Bulb Enlargement in Tulips

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Abstract

Tulip (*Tulipa gesneriana* L. ‘Gander’) bulbs just after lifting were stored dry at -2°C for 20 weeks under ambient (AA) or 3% O₂ - 3% CO₂ (CA; controlled atmosphere) conditions. Following the storage at -2°C, the bulbs were stored at 20°C for 8 weeks followed by 2°C for 12 weeks in ambient air and then forced hydroponically at 20°C under 12 h photoperiods. At the end of storage at 20°C and at 2°C, the dry weight of inner and outer daughter bulbs (offsets) of the bulbs that had been stored at -2°C in AA (AA bulbs) was greater than that of CA bulbs. The length and dry weight of shoots in the CA bulbs at planting were significantly greater than those of shoots in the AA bulbs. Thirty one percent of the plants from the AA bulbs (AA plants) failed to flower due to the abortion of flower buds during the forcing period, whereas all CA plants flowered. The cut flower quality of the CA plants was superior to that of the AA plants. The dry matter distributed to shoot and daughter bulbs in CA plants was greater than that in AA plants. Thus, the low O₂ and high CO₂ condition during the storage of vegetative bulbs just after lifting suppressed the chilling sensitivity of daughter bulbs needed for their enlargement during the subsequent storage period, and consequently promoting the growth of shoots which compete with the daughter bulbs for assimilate stored in mother bulb scales.

INTRODUCTION

For the practical cut tulip production, successive two-bulb storage phases are necessary after bulbs are lifted. The first process (phase A) is storage at an intermediate temperature (15-20°C) to promote differentiation of leaves and floral organs and the second process (phase B) is chilling (-2-9°C) storage required for subsequent flower bud development and stem elongation (Hartsema, 1961; Imanishi et al., 1997; Le Nard and De Hertogh, 1993). Since the period of bulb lifting is limited to June and July in the Northern Hemisphere, long-term storage, for retaining the forcing potential of bulbs, needs to be developed for year-round production.

Extension of the time of phase A at 20°C up to 20 weeks resulted in emergence of shoots from bulbs and flower abortion during this period (Inamoto et al., 2000a). We previously reported that the bulbs stored at -2°C in phase B retained forcing potential longer than those stored at 2°, 5° or 9°C (Inamoto et al., 2000b). However, when the bulbs were stored at -2°C for 36 weeks or longer, daughter bulbs rapidly enlarged and the shoot growth was severely depressed during forcing.

The purpose of long-term storage of bulbs is to keep the bulbs in a vegetative stage after bulb lifting and delay the start of phase A. Previously, we used a high temperature for this purpose and succeeded in obtaining bulbs that produced summer and autumn flowering (Imanishi et al., 1993). However, the high temperature storage during flower bud formation induced abnormal flowers. Moreover, most tulip cultivars are sensitive to ethylene under high temperature conditions and those bulbs show abnormalities such as gummosis (De Munk and Saniewski, 1989). A high temperature at this phase enhanced the development of daughter bulbs and suppressed the development of shoots after planting.
(Koster, 1980). Dry storage of vegetative bulbs at 5°C induces rapid growth of inner daughter bulbs and outer ones (offsets), and depression of shoot growth occurred during subsequent 20°C storage since the shoot and daughter bulbs compete for assimilates stored in mother bulb scales (Aoba, 1976; Aoba and Shibuya, 1976; Inamoto et al., 2000a, b). Since the growth of daughter bulbs is induced by chilling, it would be beneficial to suppress the chilling sensitivity of daughter bulb primordia even under a low-temperature condition.

Aoba and Shibuya (1976) observed that the chilling effect on the induction of daughter bulb enlargement was diminished by subsequent high temperature treatment. This phenomenon is analogous to vernalization and devernalization for flower bud initiation. Salisbury (1963) mentioned the necessity of oxygen for the induction of flower buds by chilling. Thus, we considered the possibility of suppressing chilling sensitivity of bulbs by a low oxygen condition.

In this study, we stored tulip bulbs at vegetative stage at a low temperature combined with low oxygen and high carbon dioxide concentration. We estimated the growth and development of shoots and daughter bulbs, which occurred during subsequent storage period and finally we evaluated the flowering potential of the bulbs by forcing.

**MATERIALS AND METHODS**

Fig. 1 shows the schedule of the experiment. Bulbs of ‘Gander’ tulips at vegetative stage were purchased from Toyama Bulb Growers Association in Japan and shipped to our laboratory on June 17, 1997. They were wrapped in low density polyethylene bags (300 mm$^3$ x 250 mm$^2$ x 0.18 mm$^1$) having two 5 mm$^0$ holes. Immediately after packing, they were stored at -2°C for 20 weeks under two atmospheric compositions as follows; 1) ambient atmospheric condition (AA) and 2) low O$2$ - high CO$2$ atmospheric condition (CA; controlled atmosphere) in a 4 L desiccator into which the air consisting of 94% (v/v) N$2$, 3% O$2$ and 3% CO$2$ was flushed at a flow rate of 200 mL min$^{-1}$. Following the -2°C storage, the bulbs stored in AA and CA (called AA and CA bulbs respectively, hereafter) were stored dry at 20°C for 8 weeks in ambient atmosphere. Flower bud completion (G stage) was confirmed under a binocular microscope and then the bulbs were stored at 2°C for 12 weeks. Before and after each storage period, ten bulbs were sampled and the length of shoot, and the fresh and dry weights of component parts, i.e., scales and basal plate of mother bulb, the shoot and daughter bulbs were measured.

After the series of dry storage mentioned above, twenty bulbs for each treatment were planted on March 23, 1998 and forced hydroponically. Tunica and outer daughter bulbs were removed before planting and those bulbs were inserted into holes (5 x 4) on a foamed polystyrene plate (620 mm$^L$ x 278 mm$^W$ x 15 mm$^H$). A plastic container (620 mm$^L$ x 278 mm$^W$ x 153 mm$^H$) was filled with nutrient solution (1000 mg L$^{-1}$ CaNO$_3$·4H$_2$O) and the foamed polystyrene plates with bulbs were floated on the solution. The forcing environment was regulated at 20°C and 12 h photoperiods with the light at 100 µmol m$^{-2}$ s$^{-1}$ PPFD at bulb level supplied from the combination of 40 W three-band fluorescent tubes and 100 W incandescent lamps. At anthesis, the plants were harvested and stem length and fresh and dry weights of component parts were measured.

**RESULTS AND DISCUSSION**

At the end of -2°C storage, there was no significant difference in fresh and dry weight of the whole bulb and its component parts between the AA and CA bulbs (data not shown). At the end of 20°C (Table 1) and 2°C (Table 2) storage, although fresh and dry weight of the whole plant was not significantly different between the AA and CA bulbs, dry weight of outer daughter bulbs of the AA bulbs was significantly greater than that of the CA bulbs. The dry weight of daughter bulbs of the AA bulbs was also greater than that of the CA bulbs but the difference was not significant. At the end of 2°C storage (i.e., at planting), shoot length and dry weight of the AA bulbs were significantly greater than those of the CA bulbs (Table 2).

Thirty one percent of the plants from the AA bulbs (AA plants) failed to flower due
to the abortion of flower buds during the forcing period, whereas all of the plants from the CA bulbs (CA plants) flowered (Table 3). Cut flowers of the CA plants had significantly heavier shoot, longer 1st internode and longer perianth than those of the AA plants. The dry matter distributed to the shoot, floral organs and the daughter bulbs in CA plants was greater than that in AA plants (Table 4).

There are few reports on the gaseous control of flower bulb storage. Kabe and Nakashizuka (1974) reported that chilling of tulip bulbs after completion of flower bud formation under a CA condition hastened flowering and improved the cut flower quality. For shipment, under a moderate temperature condition, low pressure storage (De Hertogh et al., 1978), low oxygen storage (Prince et al., 1981) and modified atmosphere storage (Prince et al., 1986) were applied to bulbs after chilling storage. Prince et al. (1981) suggested that the optimum oxygen concentration for low oxygen storage was 3-5%.

In this experiment, the CA condition suppressed the enlargement of daughter bulbs resulting in improved shoot growth at forcing. The fact that dry matter distributed to shoot and floral organs at anthesis was greater in the CA plants than in the AA plants (Table 4) suggests that the assimilate stored in mother bulb scales was well translocated after planting in the CA plants. Furthermore, the dry weight of daughter bulbs at anthesis was greater in the CA plants than in AA plants although it was smaller at planting in the CA bulbs than in the AA bulbs. This suggests that the CA condition enhanced the growth of newly developing organs. The participation of atmospheric environment in chilling sensitivity is an interesting issue and its biochemical aspect is to be studied.

**Literature Cited**


Prince, T.A., Herner, R.C. and De Hertogh, A.A. 1981. Low oxygen storage of special

Tables

Table 1. Effects of atmospheric condition during storage of vegetative ‘Gander’ bulbs at -2°C for 20 weeks on shoot length and dry weight of their component parts measured at the end of subsequent 20°C storage for 8 weeks (December 30).

<table>
<thead>
<tr>
<th>Atmospheric condition</th>
<th>Shoot length (mm)</th>
<th>Whole bulb (g)</th>
<th>Mother bulb (g)</th>
<th>Shoot (mg)</th>
<th>Daughter bulbs</th>
</tr>
</thead>
<tbody>
<tr>
<td>AA³</td>
<td>19.9</td>
<td>9.1</td>
<td>8.6</td>
<td>39</td>
<td>6</td>
</tr>
<tr>
<td>CA⁴</td>
<td>22.6</td>
<td>9.5</td>
<td>9.4</td>
<td>30</td>
<td>4</td>
</tr>
<tr>
<td>Significance⁵</td>
<td>ns</td>
<td>ns</td>
<td>ns</td>
<td>ns</td>
<td>ns</td>
</tr>
</tbody>
</table>

1 Including stem, leaves and floral organs
2 Including scales and basal plate
3 Ambient atmosphere
4 Controlled atmosphere (3% O₂ - 3% CO₂)
5 *, P<0.05; ns, P≥0.05

Table 2. Effects of atmospheric condition during storage of vegetative ‘Gander’ bulbs at -2°C for 20 weeks on shoot length and dry weight of their component parts measured at the planting (March 23).

<table>
<thead>
<tr>
<th>Atmospheric condition</th>
<th>Shoot length (mm)</th>
<th>Whole bulb (g)</th>
<th>Mother bulb (g)</th>
<th>Shoot (mg)</th>
<th>Daughter bulbs</th>
</tr>
</thead>
<tbody>
<tr>
<td>AA³</td>
<td>22.4</td>
<td>8.9</td>
<td>8.2</td>
<td>80</td>
<td>36</td>
</tr>
<tr>
<td>CA⁴</td>
<td>32.0</td>
<td>9.4</td>
<td>9.2</td>
<td>113</td>
<td>7</td>
</tr>
<tr>
<td>Significance⁵</td>
<td>**</td>
<td>ns</td>
<td>ns</td>
<td>*</td>
<td>*</td>
</tr>
</tbody>
</table>

1 Including stem, leaves and floral organs
2 Including scales and basal plate
3 Ambient atmosphere
4 Controlled atmosphere (3% O₂ - 3% CO₂)
5 **, P<0.01; *, P<0.05; ns, P≥0.05
Table 3. Effects of atmospheric condition during storage of vegetative ‘Gander’ bulbs at -2°C for 20 weeks on cut flower quality.

<table>
<thead>
<tr>
<th>Atmospheric condition</th>
<th>Flowering (%)</th>
<th>No. of days from planting to anthesis</th>
<th>Length</th>
<th>Fresh weight</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td>Total Stem (cm)</td>
<td>1st internode (cm)</td>
</tr>
<tr>
<td>AA²</td>
<td>69</td>
<td>22</td>
<td>34.9</td>
<td>7.8</td>
</tr>
<tr>
<td>CA³</td>
<td>100</td>
<td>23</td>
<td>*</td>
<td>**</td>
</tr>
<tr>
<td>Significance⁴</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

1 Including stem, leaves and floral organs
2 Ambient atmosphere
3 Controlled atmosphere (3% O₂ - 3% CO₂)
4 **, P<0.01; *, P<0.05; ns, P≥0.05

Table 4. Effects of atmospheric condition during storage of vegetative ‘Gander’ bulbs at -2°C for 20 weeks on dry weight of their component parts measured at anthesis.

<table>
<thead>
<tr>
<th>Atmospheric condition</th>
<th>Whole plant (g)</th>
<th>Mother bulb (g)</th>
<th>Shoot (g)</th>
<th>Floral organs (mg)</th>
<th>Inner daughter bulbs (mg)</th>
<th>Roots (mg)</th>
</tr>
</thead>
<tbody>
<tr>
<td>AA²</td>
<td>7.8</td>
<td>4.8</td>
<td>2.0</td>
<td>275</td>
<td>947</td>
<td>54</td>
</tr>
<tr>
<td>CA³</td>
<td>8.2</td>
<td>4.6</td>
<td>2.3</td>
<td>350</td>
<td>1249</td>
<td>53</td>
</tr>
<tr>
<td>Significance⁵</td>
<td>ns</td>
<td>ns</td>
<td>*</td>
<td>*</td>
<td>**</td>
<td>ns</td>
</tr>
</tbody>
</table>

1 Including scales and basal plate
2 Including stem, leaves and floral organs
3 Ambient atmosphere
4 Controlled atmosphere (3% O₂ - 3% CO₂)
5 **, P<0.01; *, P<0.05; ns, P≥0.05

Figures

Fig. 1. Schedule of the experiment.