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Cultivar and Seasonal Differences in the Response of Non Pre–cooled Tulip Bulbs to Gibberellin, TIBA and Root Excision

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Effects of GA_3, GA_3 with TIBA with/without root excision on growth and flowering of seven cultivars of non pre–cooled tulip bulbs were investigated. The treatments promoted the rapid growth and flowering of all the examined cultivars stored and grown at 20°C, although there was cultivar and seasonal variations of the response. Cultivars of Darwin hybrid group showed the highest response to GA_3 and GA_3+TIBA among the cultivars of other groups. The relationship between the duration of cold requirement of tulip bulbs and the response to the treatments with GA_3 and GA_3+TIBA was suggested. The seasonal difference of the response to the treatments indicates the endogenous changes of metabolism in the bulbs during the storage.

INTRODUCTION

Studies on hormonal regulation of growth and flowering of tulips have been carried out so far by using the pre–cooled bulbs with non pre–cooled bulbs (control) (for example, Aung and De Hertogh 1967; Rebers *et al.*, 1995), since the cool treatment gave the rapid growth and fastened the flowering that makes the comparison easy. Comparison using only the bulbs of non–cooling treatment has not yet been studied. Our approach is different from the past studies. We tried to induce growth and flowering in non pre–cooled bulbs by various means for the comparison with non treated non pre–cooled bulbs.

We previously clarified that 1) gibberellin A_3 (GA_3) partly replaced the cold requirement of non pre–cooled bulbs of tulip cv. Oxford, 2) when 2,3,5–triiodobenzoic acid (TIBA) was applied with GA_3, the growth and flowering were more promoted, 3) the later the treatment with GA_3 or GA_3+TIBA was, the faster the flowering was and the longer the flower stalk was, 4) excision of roots was more effective for early flowering and flower stalk elongation of the GA_3+TIBA treated bulbs when planted later, 5) naphthylphthalamic acid (NPA), another auxin transport inhibitor, with GA_3 showed the similar effects to that of TIBA and 6) endogenous indole–3–acetic acid (IAA) content in the basal plate and lower internodes was higher with GA_3+TIBA treatment than with GA_3 treat-

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ment alone (Geng *et al.*, 2005). And we concluded that TIBA blocked the polar auxin transport at the basal plate and increased auxin content in the stems and the root excision brought the better absorption of GA_3 from the cut surface, thereby promoted rapid internode elongation and flowering. Gibberellin–induced shoot growth and flowering of non pre–cooled derooted bulbs of tulip cultivars 'Apeldoorn' and 'Gudoshnik' has also been reported (Saniewski *et al.*, 1999).

Cultivated tulips have been classified into 15 groups based upon the time of outdoor flowering, morphology of the perianth parts and their origin (De Hertogh *et al.*, 1983). Cultivar difference in the time of outdoor flowering reflects the difference in low temperature requirement for subsequent growth and flowering, and this suggests the difference by cultivars or groups in the response to our treatments. In this study, effects of GA_3, GA_3+TIBA and root excision in different seasons were investigated.

MATERIALS AND METHODS

Seven cultivars of tulip (*Tulipa gesneriana* L.) bulbs (Table 2) with 12–13 cm circumferences, harvests of Niigata Prefecture, Japan in 2001, were used in this study. Upon arrival at the laboratory on 30 August the bulbs were disinfected with 1.0% Benlate (Du Pont) for 1 h and stored at 20 °C until the beginning of the experiments. The tunics were removed and 15 ('Oxford') or 10 (other cultivars) bulbs were used in each treatment. The cultivars covered early–flowering (Single early), mid–season flowering (Darwin hybrid and Triumph) and late–flowering (Single late) groups.

The bulbs were put on aluminum trays with distilled water (control), 200 mgl^{-1} GA_3 solution or 200 mgl^{-1} GA_3 solution after the lanolin paste containing 0.5% TIBA was smeared around the base of outer scale near the rim of root primordia of the bulbs with or without excision of the root primordia. The bulbs were held upright with plastic supports. They were grown on 6 September (except 'Kaiserin Maria Theresia') and 6 November at 15 °C in the phytotron (natural daylength) of The Biotron Institute, Kyushu University and transferred to 20 °C in the phytotron on the first day of the next month (1 October and 1 December, respectively). Water and GA_3 solution were renewed every two or three days. Roots that emerged after planting from the derooted bulbs were removed. The flowering date was determined when the petals were fully colored, and three days later, length of internodes was measured.

RESULTS

Response in September planting

Flowering was observed only in 'Golden Empire State' and 'Oxford' of Darwin hybrid group when the bulbs were treated with GA_3 or GA_3+TIBA in September planting (Table 1). When the bulbs were treated with only GA_3, flowering was observed in 'Oxford', but not in 'Golden Empire State'. The excision of the root primordia resulted in lower flowering rates, but it shortened the number of days to flowering of 'Golden Empire State' treated with GA_3+TIBA.

Table 1. Effects of GA_3, GA_3+TIBA and excision of roots on flowering of six cultivars of non pre-cooled tulip bulbs planted in September.

Group[z]	Cultivar	Treatment	Roots[y]	Flowering (%)	Days to flowering
SE	Merry Christmas	Water	+	0	–
			–	0	–
		GA_3	+	0	–
			–	0	–
		GA_3+TIBA	+	0	–
			–	0	–
DH	Golden Empire State	Water	+	0	–
			–	0	–
		GA_3	+	0	–
			–	0	–
		GA_3+TIBA	+	100	79.8±5.1[x]
			–	80	66.6±4.6
	Oxford	Water	+	0	–
			–	0	–
		GA_3	+	80	83.3±10.3
			–	40	80.2±13.0
		GA_3+TIBA	+	40	90.5±7.3
			–	13	92.5±3.5
T	Kees Nelis	Water	+	0	–
			–	0	–
		GA_3	+	0	–
			–	0	–
		GA_3+TIBA	+	0	–
			–	0	–
	Koki	Water	+	0	–
			–	0	–
		GA_3	+	0	–
			–	0	–
		GA_3+TIBA	+	0	–
			–	0	–
SL	Ile de France	Water	+	0	–
			–	0	–
		GA_3	+	0	–
			–	0	–
		GA_3+TIBA	+	0	–
			–	0	–

[z] SE; Single early, DH; Darwin hybrid, T; Triumph and SL; Single late.
[y] +; rooted bulbs, –; derooted bulbs.
[x]; values±standard deviation.

Response in November planting

All the cultivars flowered when they were treated with GA_3 or GA_3+ TIBA in November planting (Table 2). 'Merry Christmas', 'Ile de France' (Single early group) and 'Kaiserin Maria Theresia' (both Single late group) flowered even though their bulbs were

Table 2. Effects of GA_3, GA_3+TIBA and excision of roots on flowering of seven cultivars of non pre-cooled tulip bulbs planted in November.

Group[z]	Cultivar	Treatment	Roots[y]	Flowering (%)	Days to flowering
SE	Merry Christmas	Water	+	60	59.3±4.1[x]
			–	20	55.5±3.5
		GA_3	+	90	44.7±7.2
			–	90	38.1±7.6
		GA_3+TIBA	+	80	45.3±10.7
			–	70	33.9±5.8
DH	Golden Empire State	Water	+	0	–
			–	0	–
		GA_3	+	0	–
			–	100	46.0±4.4
		GA_3+TIBA	+	10	56.0
			–	100	43.9±2.9
	Oxford	Water	+	0	–
			–	0	–
		GA_3	+	60	56.6±5.1
			–	87	40.5±6.3
		GA_3+TIBA	+	93	57.7±5.1
			–	100	36.7±7.7
T	Kees Nelis	Water	+	0	–
			–	20	85.0±3.0
		GA_3	+	20	66.5±0.5
			–	20	63.0±3.0
		GA_3+TIBA	+	80	76.3±7.3
			–	20	60.5±1.5
	Koki	Water	+	0	–
			–	20	65.5±0.5
		GA_3	+	0	–
			–	50	40.6±6.2
		GA_3+TIBA	+	0	–
			–	60	47.2±12.8
SL	Ile de France	Water	+	90	72.4±7.0
			–	70	73.1±8.2
		GA_3	+	70	61.6±5.2
			–	80	57.6±4.6
		GA_3+TIBA	+	70	68.4±14.9
			–	90	59.4±7.2
	Kaiserin Maria Theresia	Water	+	90	76.0±5.6
			–	80	73.1±3.5
		GA_3	+	100	81.9±7.2
			–	90	58.7±6.6
		GA_3+TIBA	+	100	76.6±4.8
			–	100	54.8±6.2

[z] SE; Single early, DH; Darwin hybrid, T; Triumph and SL; Single late.
[y] +; rooted bulbs, –; derooted bulbs.
[x]; values±standard deviation.

grown on water, although they took longer days for flowering than those treated with GA_3 or GA_3+TIBA.

Single early

GA_3 and GA_3+TIBA treatments fastened the flowering date by 15 and 14 days with roots, respectively and by 17 and 22 days without roots, respectively (Table 2). Derooted plants flowered seven and 11 days earlier than rooted plants with GA_3 and GA_3+TIBA treatments, respectively, whereas the difference was only four days on water. All internodes were longer in derooted than in rooted plants with either GA_3 or GA_3+TIBA treatments, resulting in longer flower stalk (Fig. 1).

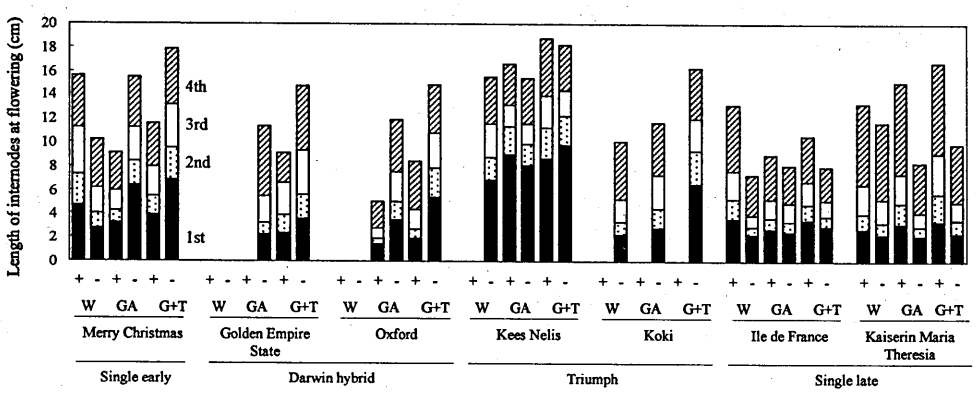

Fig. 1. Effects of GA_3, GA_3+TIBA and excision of roots on internode length at flowering of different cultivars of non pre-cooled tulip bulbs planted in November.
+; rooted, –; derooted.
W; water, GA; GA_3, G+T; GA_3+ TIBA.

Darwin hybrid

No flowering was observed on water with or without roots (Table 2). Rooted 'Golden Empire State' did not flower by the treatment with GA_3, but 'Oxford' did. GA_3+TIBA promoted the flowering date of the derooted plants by two days earlier than GA_3 in 'Golden Empire State' and four days earlier in 'Oxford'. With roots, the difference was only one day in 'Oxford'. Plant height in both cultivars was superior with GA_3+TIBA to with GA_3 treatment without roots, mainly due to the elongation of the lower internodes (Fig. 1). The fourth (top) internode was shorter instead.

Triumph

Both cultivars did not flower on water with roots, but flowered without roots (Table 2). Rooted 'Koki' did not flower even with GA_3 or GA_3+TIBA treatment. Flowering rate in both cultivars was low when compared with that in the cultivars in other group. 'Kees Nelis' without roots flowered 22 and 25 days earlier with GA_3 and GA_3+TIBA treatments, respectively than with water only. GA_3 and GA_3+TIBA accelerated the flowering in

derooted 'Koki' by 25 and 18 days, respectively. GA_3+TIBA treatment resulted the tallest plants at flowering with or without root excision, but the difference in other treatments was small in 'Kees Nelis' (Fig. 1).

Single late

Flowering rate was high in any treatment in both cultivars (Table 2). Flowering date was accelerated with GA_3 and GA_3+TIBA by 11 and four days with roots and by 16 and 14 days without roots, respectively in 'Ile de France'. Rooted 'Kaiserin Maria Theresia' flowered six and 0.6 days later with GA_3 and GA_3+TIBA, respectively, than with water, whereas root excision gave the opposite results; flowering date was 14 and 18 days earlier with GA_3 and GA_3+TIBA, respectively than with water. Root excision in both cultivars brought the shorter internode length of the plants grown on water, GA_3 or GA_3+TIBA solution (Fig. 1).

DISCUSSION

The results indicate that the treatments with GA_3 or GA_3+TIBA with or without root excision to non pre-cooled tulip bulbs are effective for rapid growth and flowering in wide range of the groups, even the response varied by cultivars, in which Darwin hybrid group showed the highest response. The role of TIBA and root excision for promoting flowering was previously clarified (Geng *et al.*, 2005).

The difference in the response to GA_3 may be related to that of cold requirement among the cultivars. 'Oxford' has longer cold requirement than 'Merry Christmas', 'Kees Nelis' and 'Ile de France' (Rees, 1977; De Hertogh, 1996). For cultivars that have longer requirement such as 'Oxford' of Darwin hybrid group, GA_3 appeared to have stronger replacement effects. It is in harmony with the previous investigations that GAs had the least effects on cultivars with the shortest cold treatment and more effects on those with longer cold treatment (Hanks, 1982).

'Oxford' and 'Golden Empire State' are both the hybrids of Darwin tulips with *T. fosteriana* (Lefeber, 1960; K. Okazaki, personal communication). Darwin tulips, formerly consisting of an independent group, are now included in the Single late group. The high response of Darwin hybrid tulips to the treatments in this study, therefore, indicates that the character might have come from *T. fosteriana*.

Single early tulips grown outdoors flower earlier in spring than other groups. It means that the cold requirement of the group is less than that of other groups. Outdoor-grown Single late tulips flower late in spring, indicating that they are less sensitive to low temperature. These may be the reason that the two groups flowered on water without GA_3 or GA_3+TIBA. The former received 15 °C (the temperature given for growth in this study) as enough low temperature during growth and the latter might have flowered at moderate temperature like 15 °C.

Difference in the flowering behavior in September and November planting suggests the endogenous changes of metabolism in tulip bulbs during the storage at 20 °C.

Root excision rather shortens the days to flowering from planting, while TIBA rather increase flower stalk length with GA_3.

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