

Effect of growth regulators on flowering of Norway spruce (*Picea abies* (L.) Karst.) grafts

By W. CHALUPKA

Institute of Dendrology, 63-120 Kornik, Poland

Received November 1978/October 1979

Summary

Triple application of GA, towards the end of May 1977 significantly increased the percentage of grafts with male flowers in 1978. A mid-June spray with CCC had no effect but jointly the two treatments acted synergistically to stimulate male flowering. The time interval between the two treatments appears important.

Key words: *Picea abies* (L.) KARST., flowering, GA₃, CCC.

Zusammenfassung

Durch dreimaliges Besprühen von Fichtenpfropflingen einer Samenplantage mit Gibberellinsäure (GA₃), gegen Ende Mai 1977, konnte der Prozentsatz im Frühjahr 1978 männlich blühender Pfropflinge signifikant erhöht werden. Das Besprühen mit Chlorcholinchlorid (CCC), Mitte Juni, hatte dagegen keine blühhördernde Wirkung, jedoch die Kombination beider Stimulantien zusammen. Der Zeitabstand zwischen den beiden Behandlungen erscheint wichtig.

Introduction

Studies in recent years have shown that gibberellins may induce flowering in trees of the family Pinaceae including *Picea abies*. DUNBERG (1974) first indicated that endogenous gibberellins participate in flower induction in Norway spruce. The same author also obtained a positive effect with gibberellin A₉ and a mixture of GA_{4/7} on flowering in *Picea abies* (DUNBERG 1976, after PHARIS and Kuo 1977). BLEYMÜLLER (1976) treated grafts of *Picea abies* with GA, and CCC which combination, in contrast to several others, is claimed to have a positive effect on female and a negative one on male flowering. On the other hand in Finland GA₃, with or without GA, had no effect on the flowering of Norway spruce grafts (LUUKKANEN and JOHANSSON 1979). The GA_{4/7} mixture had a positive effect on the flowering of *Picea glauca* (MOENCH) Voss in the experiments of KISS and PHARIS and in those of RAUTER and PHARIS (PHARIS and Kuo 1977). TOMPSETT (1977) studied the effect of various growth regulators on the flowering of *Picea sitchensis* (BONG.) CARR. grafts growing in a greenhouse or a polythene tent. He obtained among others a positive effect of gibberellin A₉ on female and male flowering. Also the GA_{4/7} mixture gave a several-fold increase in female and male flowering.

In 1976 experiments were begun at the Institute of Dendrology of the Polish Academy of Sciences in Kornik, on the effect of growth regulators on flowering of spruce and pine. The results of the pine study have already been reported (CHALUPKA 1978).

Materials and Methods

In spring 1976 in a 9-year old seed orchard of Norway spruce growing in Kornik 4 clones were selected, each represented by 12 grafts. On individual grafts two branches were selected in the same whorl, near to mid-height of the crown. One branch was girdled at its base with a full girdle 4-5 mm wide. Both branches were sprayed with one of the following combinations of growth regulators: GA₃, CCC, GA₃ + CCC or water as control. Thus the individual spraying treatments were replicated three times on three grafts

within each clone (4 treatments X 4 clones X 3 grafts). The growth regulators were applied in water solution at the following concentrations: GA₃ — 100 mg/l, CCC — 2000 mg/l with 0.1% Tween 20 added. Spraying with GA₃ was performed on the 11th, 18th und 25th May and with CCC on the 16th, 23rd and 30th June.

In the spring of 1977 the experiment was repeated on the same clones and grafts, using the same growth regulators, but spraying whole grafts. GA₃ was applied on 24th and 28th May and on 1st June and CCC on 16th and 20th June. In the spring of 1978 the male (Table 1) and female flowers were counted on treated grafts. There were only 17 female cones in the whole experiment. Data on the observations were treated by analysis of variance (Table 2).

Table 1. — Number of male flowers on treated spruce grafts.

		CLONES				
		01-16	01-24	03-34	03-35	
0	I	1	1	0	1	3
	II	0	1	0	30	31
	III	0	0	0	0	0
	Σ	1	2	0	31	34
GA ₃	I	5	49	16	0	70
	II	65	0	0	9	74
	III	0	3	0	4	7
	Σ	70	52	16	13	151
CCC	I	123	0	0	1	124
	II	0	0	0	0	0
	III	0	0	0	0	0
	Σ	123	0	0	1	124
GA ₃ CCC	I	291	4	173	2	470
	II	0	1	0	13	14
	III	11	0	234	52	297
	Σ	302	5	407	67	781
		496	59	423	112	1090

Results

A. The 1976 experiment

The assessments made in spring 1977 showed that the treatments had no effects. The branches did not flower.

B. The 1977 experiment

Male flowering

The triple spraying with GA₃ at 4 day intervals towards the end of May had a clear effect, significantly (Table 2) increasing the percentage of grafts with male flowers from 29.4% to 64.5% (Fig. 1). There was also a significant interaction between GA₃ and CCC (Table 2), the effects of which were of synergistic nature (Fig. 2). Clonal differences were significant.

The number of male flowers on the treated grafts, in spite of the considerable variability of this character (Table 1), was also significantly influenced by the growth regulators used (Table 2). Jointly GA₃ and CCC substantially

Table 2. — Results of analysis of variance.

Source of variance	% of grafts flowering ♂ (after arc-sine transformation)			Number of ♂ flowers per graft		
	DF	MS	F	DF	MS	F
Total	15	281,25		47	3631,40	
Growth regulators	3	740,62	8,47**	3	9785,31	3,12*
GA ₃	1	1701,56	19,46**	1	12480,75	3,98
CCC	1	14,06	0,16	1	10800,00	3,47
GA ₃ × CCC	1	506,25	5,79*	1	6075,00	1,94
Clones	3	403,12	4,61*	3	3998,47	1,28
Growth reg. × Clones				9	3228,97	1,03
Residual	9	87,44		32	3133,25	

* significant at 0.05 level
** significant at 0.01 level

stimulated male strobiles relative to untreated control (Fig. 3) and the effect appears to be synergistic.

Female flowering

The number of female flowers obtained (17 on 48 treated grafts) was so small that no conclusion could be made about the effect of growth regulators used on these plants.

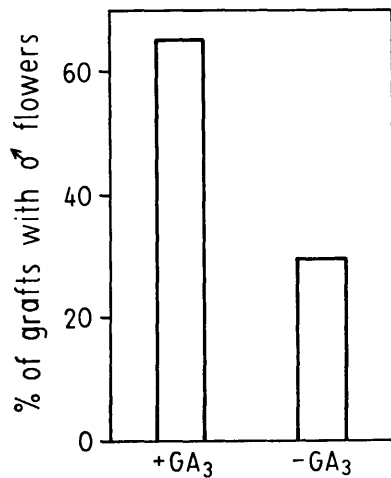


Figure 1. — The effect of GA₃ on the proportion of spruce grafts flowering male.

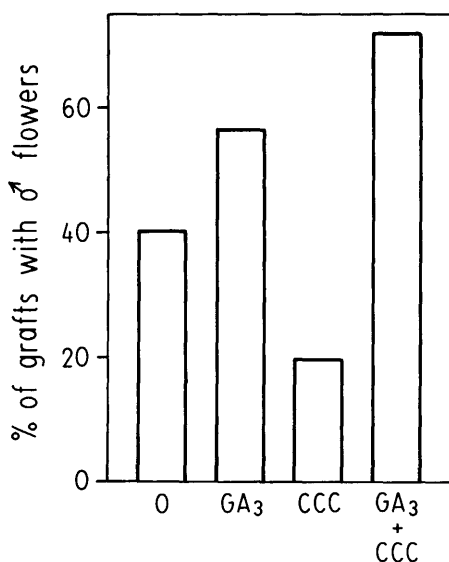


Figure 2. — The interaction of GA₃ and CCC effects on the proportion of spruce grafts flowering male.

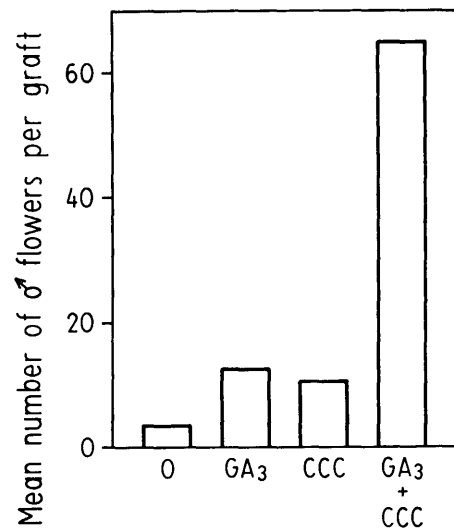


Figure 3. — Effect of GA₃ and CCC on the number of male flowers per treated graft.

Discussion

The synergistic effect of GA₃ and CCC on male flowering observed in this study confirms the less obvious results of BLEYMÜLLER (1976). The positive interaction of GA₃ and CCC is interesting in view of the expected mutual antagonism of these two growth regulators, since CCC is known to retard the processes stimulated by GAs. It is difficult therefore to explain why this combination of growth regulators leads to flowering. BLEYMÜLLER (1976) suggested that CCC is a factor blocking the biosynthesis of gibberellins and thus inhibiting GA activation of some genes. Possibly the time of application is important here. Both in BLEYMÜLLER's experiments and those reported here, GA₃ was supplied at the time when shoot elongation was beginning and could have acted positively on this process. CCC given a few weeks later towards the end of height growth, could have inhibited the increased growth activity causing a physiological shock that resulted in flower induction.

The difference in sex of flowers stimulated by the GA₃ and CCC combination in BLEYMÜLLER's work and mine appears of secondary importance. It happens that application of growth regulators stimulates both male and female flowers (eg. TOMPSETT 1977). These apparently contradictory results can be explained by the probable sequence of events leading to flower induction. It appears that flower formation in spruce (and possibly other species) takes place in two

stages. First, in some of the apical meristems induction of the generative state occurs leading to differentiation into vegetative and generative meristems. In the second stage male and female initials form and differentiate. This suggestion finds confirmation in some reports. OWENS and MOJDER (1976) have observed in *Picea sitchensis* differences in apical domes before initiation of bud scales, that is before the onset of formation of lateral organs. They found that in pith cells of future domes of vegetative buds there accumulated more phenolic and ergastic compounds than in pith cells of future domes of generative buds. These authors also report that apical domes of generative buds show greater mitotic activity during formation of the initials.

In 1978 I collected from Norway spruce grafts both male flowers and shoots with needles from analogous sites on the branches. It turned out that the number of stamens was twice as large as the number of needles. This observation confirms the greater mitotic activity of apical domes leading to male strobiles, which also agrees with the suggestions about the causal relation between rate of growth of the apical meristem and the type of bud that will develop from it (ROMBERGER and GREGORY 1974, TOMPSETT 1978). One suspects therefore that a difference in mitotic activity in favour of generative apices is caused by earlier induction of the generative state of these meristems.

Interpreting these results on the basis of this hypothesis one can say that GA₃ together with CCC favoured induction of the generative state in meristems, while the fact that male and not female initials were laid down was the result of other factors. This would explain why PHARIS *et al.* (1975) were able to induce female flowering in *Pinus contorta* DOUGL. when the GA_{4/7} application was made during differentiation of male flower initials. The reports in the literature on the possibility of changing sex of initials under the influence of growth regulators (SAITO 1957) represents indirect confirmation of the explanation suggested above concerning sequential induction of the generative state in apical meristems.

In any case it appears that there already are distinct differences between vegetative and generative meristems at the initial stage of flower induction that is during initiation of bud scales. These differences are much greater and more difficult to alter than those occurring later between female and male flower initials.

Acknowledgements

The author wishes to thank Dr. MACIEJ GIERTYCH from Institute of Dendrology in Kórnik for critical comments and translation of the text.

Literature

- BLEYMÜLLER, H.: Investigation on the Dependence of Flowering in Spruce (*Picea abies* (L.) KARST.) upon Age and Hormonal Treatment. *Silvae Genet.* 25 (2): 83—85 (1976). — CHALUPKA, W.: Effect of growth regulators on the flowering of Scots pine (*Pinus sylvestris* L.) grafts. *Silvae Genet.* 27 (2): 62—65 (1978). — DUNBERG, A.: Occurrence of gibberellin-like substances in Norway spruce (*Picea abies* (L.) KARST.) and their possible relation to growth and flowering. *Stud. For. Suec. Nr.* 111. (1974). — LUUKKANEN, O., and JOHANSSON, S.: Flower induction by exogenous plant hormones in conifers. *Silva Fenn.* 13 (in print) (1979). — OWENS, J. N., and MOJDER, M.: Bud development in Sitka spruce. II. Cone differentiation and early development. *Can. J. Bot.* 54 (8): 766—779 (1976). — PHARIS, R. P., and KUO C. G.: Physiology of gibberellins in conifers. *Can. J. For. Res.* 7 (2): 299—325 (1977). — PHARIS, R. P., WAMPLE, R. L., and KAMIENSKA, A.: Growth, Development, and Sexual Differentiation in *Pinus*, with Emphasis on the Role of the Plant Hormone, Gibberellin. In: *Management of Lodgepole Pine Ecosystems* (ed. D. M. BAUMGARTNER), Vol. I: 106—134, Pullman, Wash, State Univ. (1975). — ROMBERGER, J. A., and GREGORY, R. A.: Analytical morphogenesis and the physiology of flowering in trees. *Proc. 3rd North American Forest Biology Workshop*, 9—12 Sept., Colorado State Univ.: 132—147 (1974). — SAITO, Y.: Artificial Control of Sex Differentiation in Japanese Red Pine and Black Pine Strobiles. *Journ. Fac. Agr., Tottori Univ.*, Vol. III, No. 1. (1957). — TOMPSETT, P. B.: Studies of Growth and Flowering in *Picea sitchensis* (BONG.) CARR. 1. Effects of Growth Regulators Applications to Mature Scions on Seedlings Rootstocks. *Ann. Bot.* 41: 1171—1178 (1977). — TOMPSETT, P. B.: Studies of Growth and Flowering in *Picea sitchensis* (BONG.) CARR. 2. Initiation and Development of Male, Female and Vegetative Buds. *Ann. Bot.* 42: 889—900 (1978).

Comparison of a *Eucalyptus tereticornis* × *E. grandis* controlled hybrid with a *E. grandis* × *E.tereticornis* putative natural hybrid

By C. S. VENKATESH¹⁾ and V. K. SHARMA²⁾

(Received October 1978 / May 1979)

Summary

Species hybrids of a *E. tereticornis* × *E. grandis* controlled cross FRI-6 and a *E. grandis* × *E. tereticornis* putative natural cross FRI-10, are compared with the parental species. In more than half the total number of contrasting characters studied, the hybrids were intermediate between the parental species; in the rest, including lignotuber development, flowering precocity etc. they were more like

one or the other parent, inheritance apparently being dominant for such characters. Hybrids of the putative natural hybrid FRI-10, where *E. grandis* was involved as the maternal parent, were conspicuously heterotic in height and diameter growth as compared to their pure *E. grandis* half-sibs and non-sibs. The potential practical value of such species hybrids in Indian Forestry, is discussed.

Key words: *Eucalyptus tereticornis*, *E. grandis*, Interspecific hybridization.

Zusammenfassung

Arthybriden von *Eucalyptus tereticornis* × *E. grandis* aus einer kontrollierten Kreuzung (FRI-6) und einer wahr-

¹⁾ Head, Division of Genetics, Kerala Forest Research Institute, Peechi 680 653, Trichur, Kerala, India.

²⁾ Research Assistant, Forest Genetics Branch, Forest Research Institute, Dehra Dun 248 006, India.