林試研報 Bull. For. & For. Prod. Res. Inst. No. 313, 1981 37～45

Effect of Gibberellins on the Promotion of Strobilus Production in *Larix leptolepis* GORD. and *Abies homolepis* SIEB. et ZUCC.

By

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Summary : The effect of gibberellin (GA) and girdling treatments on the promotion of strobilus production in *L. leptolepis* and *A. homolepis* grafts which were close to sexual maturity was investigated. For *L. leptolepis* grafts, treatments of $GA_{4/7}$, GA_4 and GA_3 applied separately were ineffective to promote strobilus production, but girdling treatments at the base of branches and stems had a promotive effect on strobilus production, especially for males. For *A. homolepis* grafts, treatments of $GA_{4/7}$ and GA_3 were slightly effective, but the promotive effect of $GA_{4/7}$ remained obscure in the additional test a year later. Girdling treatment at the branch base was not promotive. Some of the less-polar GAs are not promising for practical application to the seed orchards of *L. leptolepis* and *A. homolepis* as far as this experiment is concerned, but it is necessary to examine additional effects of GAs in combination with each other or with other cultural practices.

Introduction

Since it was first demonstrated that the exogenous application of GA_3 induced precocious flowering in young *Cryptomeria japonica* seedlings[7], many experiments have been made to verify the effectiveness of GAs in promotion of strobilus production with a large number of woody species. The results of many experiments indicated that the exogenous application of GA_3 was highly effective in promoting strobilus production and its effect was readily reproducible only in species within the *Taxodiaceae* and *Cupressaceae*[2,4,6,11,13,14]. Gibberellins other than GA_3, e. g., the $GA_{4/7}$ mixture, GA_4 and GA_9, were also effective in the *Taxodiaceae* and *Cupressaceae*, but their effect was equal to or less than that of GA_3 in a relative order of effectiveness, as tested on some species in the *Cupressaceae*[15]. Thus the exogenous application of GA_3 is now being used effectively for enhancing strobilus production and consequently for increasing seed production in many practical seed orchards of *Cryptomeria japonica* and sometimes in *Chamaecyparis obtusa* seed orchards in our country.

The exogenous application of GA_3 was, however, unsuccessful in inducing precocious flowering for most species of the *Pinaceae*[2,4,6,11,13,14], and until recently it was generally believed that the GA application was essentially ineffective in promoting strobilus production for the *Pinaceae*, although a slightly promotive but not repeatable effect was observed in young *Pinus mugo* seedlings by spraying 200 ppm GA_3 solution[21] and in young *Pinus densiflora* seedlings by spraying 300 ppm GA_3 or 500 ppm GA_7 (probably $GA_{4/7}$ mixture) solution[2～4].

Recently it was emphasized that the exogenous application of some less-polar GAs pro-

Received April 14, 1980 造 林—49 Silviculture—49

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moted strobilus production in several species within the *Pinaceae*, with the progress of studies of endogenous, less-polar GA-like substances characterized by bioassay or by chemical means, as reviewed by PHARIS and co-workers[14)16)]. Strobilus production was significantly promoted by treatments of the $GA_{4/7}$ mixture applied separately or often in combination with GA_3, GA_5, GA_9 and low levels of NAA in 2- or 4-year-old grafts and 4- or 6-year-old seedlings of *Pseudotsuga menziesii*[12)16)19)20)], in 3-year-old grafts of *Pinus taeda*[16)] and in 6-year-old seedlings of *Pinus contorta*[17)]. The success in promoting strobilus production in the *Pinaceae* was achieved with a relatively high dosage, e. g., up to 400 μg per branch (bud), by frequent application during a rather long period, e. g., biweekly from March to July, and often in combination with girdling as a pretreatment before the GA application. An attempt has been made to extend the success in several species of *Pseudotsuga* and *Pinus* to other commercially important species in the *Pinaceae*, as in several *Picea* species[14)].

As the exogenous application of some less-polar GAs would appear to be a promising technique for seed orchard management in the *Pinaceae* which required many years to attain full sexual maturity, the cooperative research on enhancing strobilus production for *Abies*, *Larix*, *Pinus* and *Chamaecyparis* species has been conducted with several research institutes in our country in 1977[8)]. The present paper is part of the cooperative research, and is concerned with the practical application of GAs in the seed orchards of *L. leptolepis* and *A. homolepis*.

Materials and methods

Experiments were made by using grafted ramets of *L. leptolepis* and *A. homolepis* plus tree clones from a central mainland source which were planted in the experimental seed orchards at the Nagano Branch Station, Kanto Forest Tree Breeding Institute, Komoro, Nagano (Photo. 1 A & B). Natural flowering had not been observed in *L. leptolepis* grafts before experiments were made, while a little natural flowering had often been recorded in *A. homolepis* grafts since 1967, although it was generally negligible[5)].

Nine-year-old grafts of 12 *L. leptolepis* clones, 5.78 (5.29 to 6.93) m in mean height and 8.5 (7.5 to 10.4) cm in mean D. B. H., and 14-year-old grafts of 10 *A. homolepis* clones, 4.24 (3.18 to 5.70) m in mean height and 9.0 (6.3 to 12.9) cm in mean D. B. H., were selected for treatments in 1977. In the following year, 2 clones, 4 in total, were selected again from good or poor flower bearers in each species which were confirmed by untreated controls in the test a year before. Two ramets of each clone were always used for the GA and girdling treatments on branches and two extra were used for the stem girdlings in 1977. The two others of the 4 clones selected were prepared for treaments in 1978.

Four or eight (only in 1977) main branches (first-order branches) of *L. leptolepis* grafts, which were 2.6 (2.3 to 2.8) cm in mean diameter at the branch base in 1977 and 2.5 (1.5 to 3.9) cm in 1978 and were selected from whorls below the central part of the crown, were used for treatments on branches (Photo. 2 A). Four main branches of *A. homolepis* grafts, which were 1.9 (1.6 to 2.5) cm in mean diameter at the branch base in 1977 and 2.1 (1.9 to 2.3) cm in 1978 and were selected from whorls of nearly the central part of the crown, were used for treatments on branches (Photo. 2 B). Main branches were always used in the regular order of treatments from the top, that is, controls, branch girdling, GAs applied by incising, and GAs applied by spraying.

Branch girdling was double overlapping, semicircular girdles about 0.5 cm wide at a distance of a half-diameter and 10 to 20 cm apart from the branch base. Stem girdling was thrice overlapping, semicircular girdles about 1 cm wide at a distance of a half-diameter and 50 to 70 cm apart from the stem base. Girdling treatment was examined as a control to ascertain the effectiveness of GAs.

Gibberellins $A_{4/7}$ mixture, A_3 and A_4 were tested separately, but not in combination with each other or with other plant growth substances. Gibberellins were applied by filling into small incisions at 20 mg per branch in 1977 and at 15 mg per branch in 1978, because of a harmful effect with falling leaves in some clones that was observed in 1977. Gibberellins $A_{4/7}$ (powder) and A_3 (powder) were directly introduced into small incisions 3 to 5 cm long in the upper side of the branch and 10 to 20 cm apart from the branch base in 1977. Gibberellins $A_{4/7}$ (powder) and A_4 (crystal) were put into small incisions by mixing with sodium carboxy methyl cellulose (CMC) paste in 1978. Gibberellins $A_{4/7}$ (crystal) were sprayed to the lowest branches with about 200 m*l* per branch of 100 mg/*l* aqueous solution containing 0.1% Tween-20. Crystal GAs were always used after diluting by a very little ethanol. Powder GAs melted away completely in the incisions in *L. leptolepis* branches, but about half of a given powder remained in *A. homolepis* branches 2 weeks after application.

Girdling and GA treatments in both species were made once during the period between early June and mid-July in 1977, with the exception of treatment by spraying, and also once in late June in 1978, when flower buds could be expected to differentiate.

Male and female strobili were counted separately on whole treated branches in the following late April, but the number of total buds was not counted in the present test. It is thought that the number of strobili per total buds including vegetative and reproductive ones on branches may be a better method for evaluating the effectiveness of treatments. The significant differences among treatments of strobilus production per branch were analyzed by the LSD test.

Experimental results

Larix leptolepis

Only one of 12 clones (Numazu-109) differentiated female strobili, but almost all of the clones bore male strobili in the untreated and incised controls in 1977. That year appears

Table 1. Effects of GA and girdling treatments on female and male strobilus production in 9-year-old grafts of 12 *L. leptolepis* clones (Results in 1977)

Treatments	Treatment dates	Female strobili per branch	Male strobili per branch
untreated control	—	0.3	115.6
incised control	June 2	0.3	130.5
GA_3	June 2	0.8	113.6
$GA_{4/7}$	June 2	0.4	130.2
$GA_{4/7}$	June 24	0.5	101.3
$GA_{4/7}$	July 14	0.8	69.7
$GA_{4/7}$ (spraying)	June 15 & July 14	1.0	114.8
girdling-branch	June 2	9.0	461.4
girdling-stem	June 2	13.3	389.2
LSD, 5%		13.2 (ns)	154.5

to have been the first flower bearing year of the grafts.

As compared with the untreated or incised controls, treatments of $GA_{4/7}$ and GA_3 applied by introducing powder into incisions at 20 mg per branch or $GA_{4/7}$ applied by spraying 100 ppm aqueous solution had little or no effect on the strobilus production of both sexes (Table 1). On different treatment dates during the period from June 2 to July 14, $GA_{4/7}$ were also ineffective to promote strobilus production. In addition to this, it should be noted to be excessive application at 20 mg per branch, by which new foliage became chlorotic and there was a visible toxic effect (defoliation) after 1 to 2 months in 5 clones, as marked in Fig. 1.

Girdling treatments at the base of branches and stems significantly ($p=0.05$) promoted male strobilus production (Table 1). Female strobilus production similarly increased and was greater than that in the untreated or incised controls, but differences between the controls

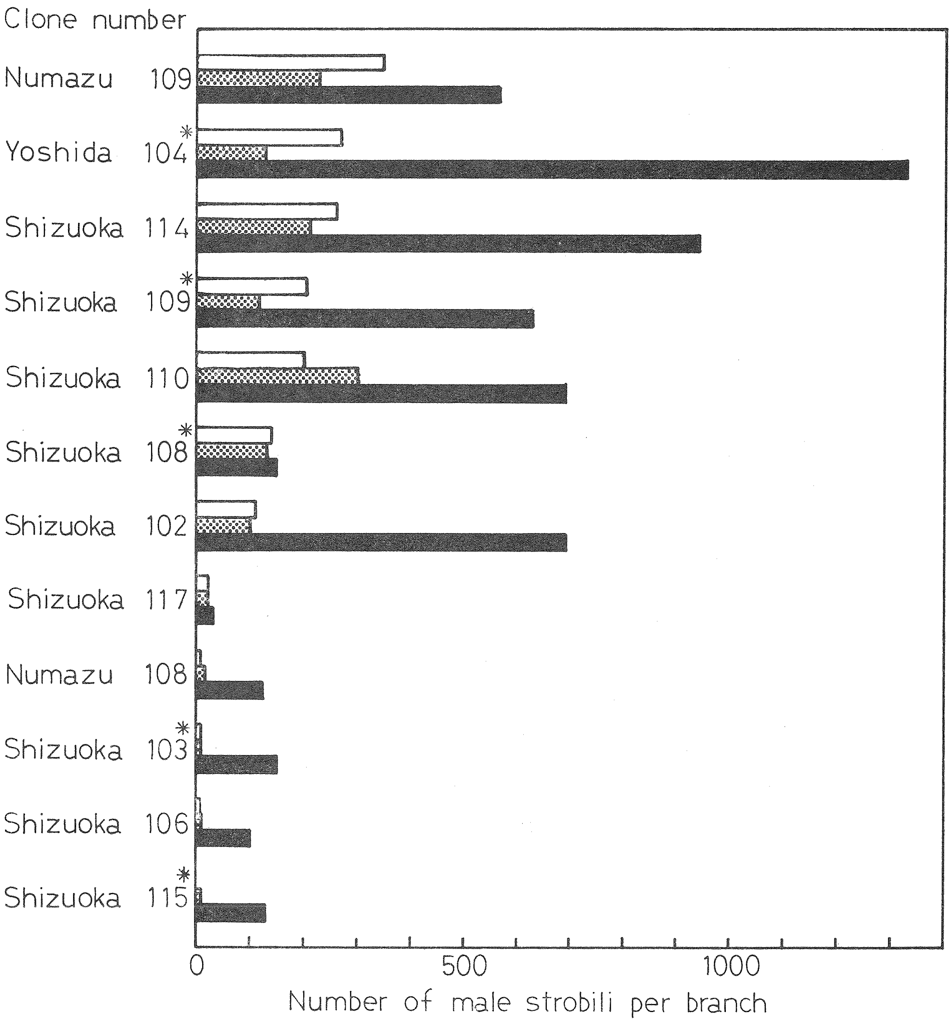

Fig. 1 Male strobilus production in each of 12 *L. leptolepis* clones (Results in 1977).

Open column : incised control, Dotted column : $GA_{4/7}$, Data are pooled for $GA_{4/7}$ applied at different treatment dates, Solid column : girdling-branch, * : Toxie effect (defoliation) by $GA_{4/7}$ application at 20 mg per branch.

Table 2. Effects of GA and girdling treatments on female and male strobilus production in 10-year-old grafts of 4 *L. leptolepis* clones with good or poor flower bearing (Results in 1978)

Treatments	Treatment dates	Female strobili per branch	Male strobili per branch
untreated control	—	2.5	137.8
$GA_{4/7}$	June 27	2.3	158.0
GA_4	June 27	4.5	154.8
girdling-branch	June 27	14.0	442.5

and girdling were not statistically significant, because the promotive effect on female strobilus production was limited only to some clones (Numazu-109, Yoshida-104 and Shizuoka-108). The response on male strobilus production to girdling was apparently affected by the level of male flower bearing in the incised (or untreated) control in each of the clones (Fig. 1), although the correlation between the incised control and girdling was not significant ($r=0.472$). The effect of girdling treatments to promote male strobilus production in *L. leptolepis* that was generally known through the past reports of MELCHIOR[9)10)] and many Japanese workers was sure to have a high relation to a natural or inherent predisposition to flower bearing in each of the clones, but an actually promotive effect by girdling in each of the clones was subjected to wide fluctuations, as indicated by no significant correlations.

From continued observations in the following year, using grafts of 4 clones with good (Numazu-109 and Shizuoka-110) or poor (Numazu-108 and Shizuoka-103) flower bearing in 1977, nearly the same responses of strobilus production to GA and girdling treatments were recognized (Table 2), as in the results of the test a year before. Treatments of $GA_{4/7}$ and GA_4 applied by introduction into incisions at 15 mg per branch had little effect on the strobilus production of both sexes. Girdling treatment at the branch base, however, promoted female and male strobilus production.

Similar results on strobilus production by treatments of $GA_{4/7}$ and girdling in *L. leptolepis* grafts have also been obtained in one of the cooperative experiments at the Tohoku Forest Tree Breeding Institute (MIKAMI, S. et al., pers. comm.).

Abies homolepis

Only two of 10 clones (Numazu-101 and Shizuoka-107) differentiated female strobili and

Table 3. Effects of GA and girdling treatments on female and male strobilus production in 14-year-old grafts of 10 *A. homolepis* clones (Results in 1977)

Treatments	Treatment dates	Female strobili per branch	Male strobili* per branch
untreated control	—	0.5	1.0
girdling-branch	June 3	0.6	0.7
GA_3	June 3	1.1	3.0
$GA_{4/7}$	June 3	2.1	4.6
LSD, 5%		2.0 (ns)	5.3 (ns)

*: Counted as number of clusters.

Table 4. Effects of GA and girdling treatments on female and male strobilus production in 15-year-old grafts of 4 *A. homolepis* clones with good or poor flower bearing (Results in 1978)

Treatments	Treatment dates	Female strobili per branch	Male strobili* per branch
untreated control	—	0	0.5
girdling-branch	June 27	0	1.0
$GA_{4/7}$	June 27	0.3	0.8
GA_4	June 27	0	0.3

* : Counted as number of clusters.

also two clones (Numazu-101 and Yabuhara-103) bore male strobili in the untreated control in 1977. The number of strobili of both sexes in the untreated control was negligible.

The same clones named in the untreated control mainly bore strobili for the GA and girdling treatments. The girdling treatment at the branch base had little effect on the strobilus production of both sexes (Table 3), although male strobilus production seemed to slightly increase by girdling at the stem base from an observation made at the same time. Treatments of $GA_{4/7}$ and GA_3 applied by introducing powder into incisions at 20 mg per branch apparently promoted male and female strobilus production, as compared to the untreated control (Table 3). Gibberellins $A_{4/7}$ were more effective than GA_3, but differences between the untreated control and both GAs were not statistically significant in both sexes.

According to the additional test a year later, using grafts of 4 clones with good (Numazu-101 and Shizuoka-107) or poor (Numazu-104 and Komagane-101) flower bearing in 1977, there was no evidence for the promotive effect on strobilus production by treatments of $GA_{4/7}$ and GA_4 applied by introduction into incisions at 15 mg per branch (Table 4). The promotive effect of GAs on strobilus production was not consistent between test years and so remained obscure. This uncertainty may be attributed to the difference of environmental conditions between test years, as well as the inadequacy of the position of the main branches selected within the crown.

Further aspects of GA application to the *Larix* and *Abies*

The exogenous application of $GA_{4/7}$, GA_4 and GA_3 was ineffective to promote strobilus production in *L. leptolepis* grafts which were close to sexual maturity, even at a relatively high concentration as shown by a visible toxic effect in 1977. GA application, however, seemed to be slightly effective in *A. homolepis* grafts, but the promotive effect of $GA_{4/7}$ remained obscure in the additional test a year later.

From the observations of this experiment, less-polar GAs are not promising for the practical application to the seed orchards of *L. leptolepis* and *A. homolepis* at the present time, but the possibility of the promotion of strobilus production by GA application is undeniable. Gibberellins $A_{4/7}$, A_4 and A_3 are applied separately in this experiment, but not in combination with each other or with other cultural practices, as recently reported in *Pseudotsuga menziesii* seedlings[18] and *Pinus taeda* grafts[1]. Gibberellins are also applied once on a date during the supposed period of flower bud differentiation in this experiment, but not by frequent application for a rather long period, as shown by an example of biweekly application of GAs over

12 weeks from March to June[19]. It is, therefore, necessary to examine in *L. leptolepis* and *A. homolepis*, whether an additional effect of GAs can be expected or not, as GAs are applied in combination with each other or with girdling and other cultural practices.

Acknowledgement

The authors wish to express their thanks to Mr. K. Hatiya and Dr. S. Asakawa, Forestry and Forest Products Research Institute, for undivided advice concerning the manuscripts. To members of the Nagano Branch Station, Kanto Forest Tree Breeding Institute, appreciation is expressed for their assistance. The cooperation of Kyowa Hakko Kogyo, Co. Ltd., Tokyo, in providing samples of the $GA_{4/7}$ mixture and GA_4 is gratefully acknowledged.

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* : In Japaness with English summary, ** : Only in Japanese.
Titles of papers in parentheses are tentative translations from the Japanese originals by the present authors.

Explanation of plate

Photo. 1. Grafts of plus tree clones examined in the experimental seed orchards.

A : *L. leptolepis* seed orchard established in 1970, 0.45 ha consisting of 178 ramets of 29 plus tree clones at an initial planting distance of 5 m.

B : *A. homolepis* seed orchard established in 1965, 0.75 ha consisting of 452 ramets of 25 plus tree clones at an initial planting distance of 4 m.

Photo. 2. An example of the position of main branches within the crown used for treatments.

A : *L. leptolepis* graft

B : *A. homolepis* graft

Main branches (first-order branches) within the range of the arrows in the plates were used in each species.

カラマツ，ウラジロモミでのジベレリンによる着花促進の効果

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摘　　要

スギ，ヒノキ科の多くの樹種では，ジベレリン（以下GAと略記する）とくにGA_3が花芽分化を促進し，再現性のある効果を示すことが明らかになっているが，マツ科の樹種ではGAによる花芽分化の促進は期待できないと考えられてきた。最近，極性の低いGA（$GA_{4/7}$，GA_4，GA_9など）を用いた一連の実験で，マツ科の樹種でもGAが花芽分化を促進する可能性のあることが明らかになったので，わが国に生育するマツ科の樹種で，極性の低いGAが着花促進にどの程度の効果を示すのか，また実用化への可能性をもつものかを確かめることにした。

本試験では，関東林木育種場長野事業場構内に設定されているカラマツ，ウラジロモミ採種園内のツギキクローン（カラマツ9年生12クローン，ウラジロモミ14年生10クローン）を用い，昭和52，53年の2年間にわたって，GA（$GA_{4/7}$，GA_3，GA_4）と環状剥皮処理による着花促進の効果を調べた。

カラマツでは，1主枝あたり20mgあるいは15mgの$GA_{4/7}$，GA_3，GA_4の枝基部での包埋処理，および1主枝あたり約200mlの100ppm $GA_{4/7}$の散布処理で，雌，雄花ともに着花促進の効果が認められなかった。しかし，枝あるいは幹基部での環状剥皮処理は，著しく着花を促進し，とくに雄花が多量に着生した。ウラジロモミでは，1主枝あたり20mgの$GA_{4/7}$，GA_3の枝基部での包埋処理が，雌，雄花ともに着花を促進したが，1主枝あたり15mgの$GA_{4/7}$，GA_4の包埋処理による継続試験では，明確な促進効果が認められなかった。樹冠内での処理枝の位置と処理時期，方法について，今後の検討が必要である。枝基部での環状剥皮処理では，着花を促進する効果が認められない。

本試験の結果によれば，カラマツ，ウラジロモミで，一部の極性の低いGAによる実用的レベルでの着花促進は期待できないと考えられるが，異種のGAの混合，あるいは他の物理的処理との併用による付加的な促進効果について，さらに詳細に調べる必要があると考える。

1980年4月14日受理
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Photo. 1

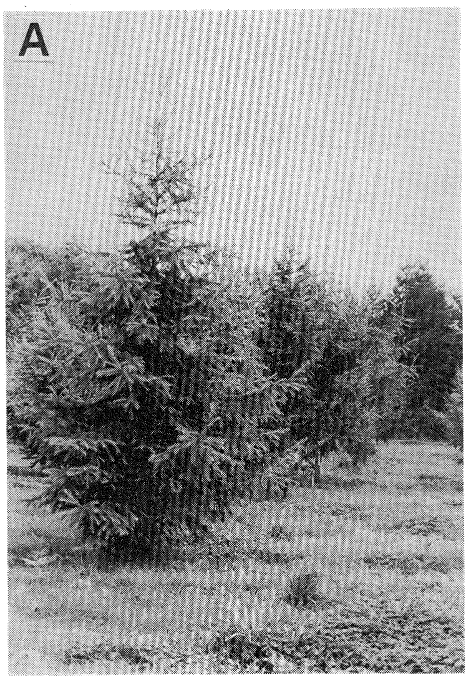

Photo. 2