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Effect of GA_{4/7} on flowering of pruned and unpruned seedlings of Scots pine (*Pinus sylvestris* L.)

Abstract

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Influence of GA_{4/7} was studied on the flowering of pruned and unpruned 13-year-old seedlings of Scots pine. GA_{4/7} injection significantly stimulated female and male strobili initiation while pruning enhanced the number of lateral long-shoots formed. On the basis of these results a combined treatment increasing the number of flowers was proposed.

Additional keywords: Flower induction, GA injection, long-shoot formation, GA × pruning interaction.

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INTRODUCTION

Trials with hormonal stimulation of flowering in Scots pine were only done on vegetatively propagated individuals, i.e. on grafts growing in seed orchards and/or clone archives. The literature on this subject was recently reviewed by Chałupka (1991).

Artificial regeneration of Polish Scots pine forests is partially based on seeds collected from standing trees in qualified seed stands. Phenotypical individual selection of plus trees is also done in such seed stands and seedling seed orchards are established from the generative progeny of these trees (Kocięcki, 1980). Thus the problem of abundant flowering in seedling seed orchards arises. For that reason experiments on the stimulation of flowering in Scots pine seedlings were started.

MATERIALS AND METHODS

Two experiments were conducted in 1988 on 13-year-old Scots pine seedlings (half-sib progenies). In the first experiment GA_{4/7} was applied to

slightly pruned trees. Three largest first order branches were selected for pruning in 1986 and 1985 stem whorls, and three variants of pruning were tested: P_0 – unpruned branch, P_{25} and P_{50} – removing leading 1988 shoots on the branches when they achieved respectively 25% and 50% of the total length of their 1987 growth increment (Fig. 1).

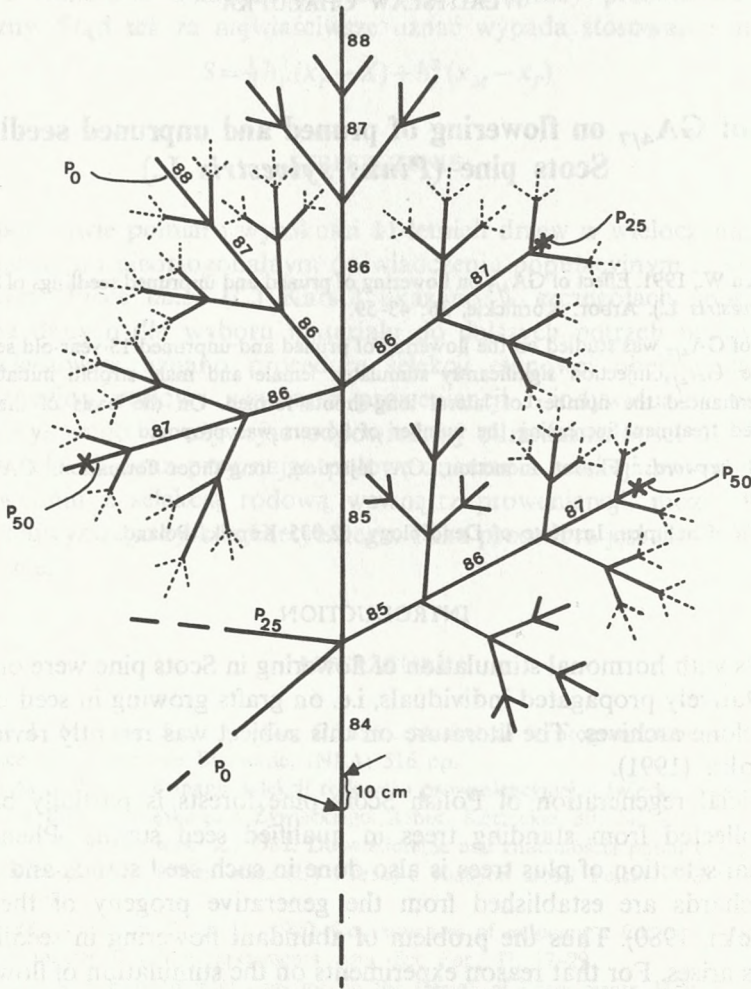


Fig. 1. Schematic diagram of the observed crown part in treated Scots pine trees. P_0 , P_{25} and P_{50} – pruning variants (explanation in the Materials and Methods); X – removed '88 shoots; – 1989 shoots taken for evaluation of analysed characters; 84, 85, 86, 87, 88 – year's growth increment of internodes and respective whorls. Arrows indicate place of injections

In the second experiment $GA_{4/7}$ was applied to unpruned trees. Similarly as in the above described experiment three first order branches in the same stem whorls were selected for testing the results of $GA_{4/7}$ application.

In both experiments GA_{4/7} was dissolved in ethanol and injected once on July 7th at a concentration of 160 mg ml⁻¹. The injection was done into two bored holes on opposite sides of the trunk, using a micropipette (Fig. 1). The amount of GA_{4/7} solution injected to one hole was 250 μ l. Thus 500 μ l of solution was applied per tree, i.e. 80 mg of GA_{4/7}. Control trees obtained 500 μ l of ethanol.

Observations of flowering were done in May 1989 in both experiments on shoots formed in 1988 and extended in 1989 on two 1988 and 1987 branch whorls (Fig. 1). An analysis of variance was used for statistical evaluation of data concerning three characters: (1) total number of currently elongating lateral long-shoots, (2) number of female strobili per 100 shoots, and (3) percentage of shoots with male strobili. The percentage data were subjected to arcsine transformation before analysis (Snedecor, 1956).

RESULTS

PRUNED TREES

Results of analysis of variance for the experiment with pruned trees are presented in Table 1.

NUMBER OF CURRENTLY ELONGATING LATERAL-SHOOTS

Only slight pruning in the P₂₅ variant significantly increased the total number of 1989 shoots formed in 1988, i.e. in the year the treatment was done (Fig. 2). This means that the P₅₀ pruning was done too late to influence the formation of long-shoot primordia.

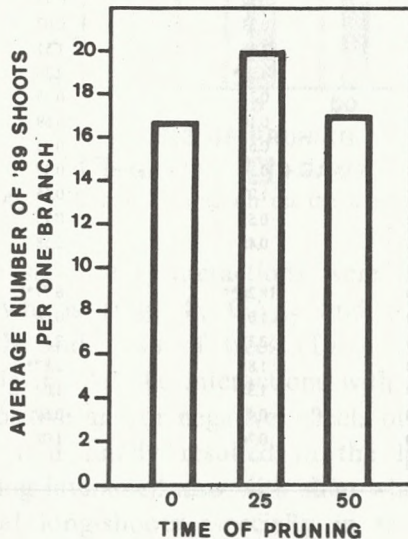


Fig. 2. Effect of pruning on the number of '89 long-shoots

Table 1

The influence of GA_{4/7} on flowering of pruned Scots pine seedlings. Results of analysis of variance. Values of F: ** – significant at 0.01 level; * – significant at 0.05 level

Source of variation	Degrees of freedom	Total number of long-shoots elongating in '89	Number of female strobili per 100 '89 shoots	Percentage of '89 shoots with male strobili
Total	239			
Variables	119	3.32**	1.67	1.88
Pruning (P)	2	8.94**	0.11	0.53
GA _{4/7} (G)	1	0.01	59.64**	4.87*
Stem whorls (W)	1	10.31**	5.51*	4.04
Branch whorls (B)	1	41.41**	0.52	57.16**
Rows (R)	4	35.76**	3.75*	8.85**
P × G	2	5.19*	0.23	1.90
P × W	2	4.11*	2.04	0.58
P × B	2	1.80	0.51	0.99
P × R	8	1.39	0.93	0.46
G × W	1	0.31	4.91*	0.01
G × B	1	6.74*	0.18	3.27
G × R	4	6.13**	4.17*	4.16*
W × B	1	1.67	0.16	0.08
W × R	4	2.75	1.41	0.71
B × R	4	3.18*	2.29	5.13**
P × G × W	2	1.41	2.37	2.18
P × G × B	2	0.59	0.32	0.13
P × G × R	8	1.76	0.94	0.87
P × W × B	2	0.97	0.96	1.32
P × W × R	8	2.17	0.98	0.88
P × B × R	8	0.32	0.48	0.14
G × W × B	1	0.31	0.07	0.00
G × W × R	4	2.36	1.51	2.74
G × B × R	4	4.30*	2.36	1.66
W × B × R	4	0.31	0.55	1.05
P × G × W × B	2	0.10	0.69	0.43
P × G × W × R	8	0.63	0.73	0.57
P × G × B × R	8	0.56	0.49	0.26
P × W × B × R	8	1.07	0.65	0.72
G × W × B × R	4	0.53	0.75	0.78
P × G × W × B × R	8	0.48	0.58	1.03
Trees in R and G variants (T _{GR})	10	18.20**	6.77**	13.38**
P × T _{GR}	20	1.08	0.68	0.89
W × T _{GR}	10	2.32	2.14	1.49
B × T _{GR}	10	1.88	2.87*	6.71**
P × W × T _{GR}	20	1.38	1.04	0.64
P × B × T _{GR}	20	0.47	0.69	0.38
W × B × T _{GR}	10	0.76	1.08	0.40
P × W × B × T _{GR} (Error)	20			

The differences in the number of 1989 elongating shoots between the 1986 and 1985 stem whorls (37.8 vs. 33.6 average per one 1st order branch), and 1988 and 1987 branch whorls (15.7 vs. 20.0) were strongly significant (Table 1). This is the result of a natural ageing process and branching pattern of the trees. Also the differences between randomly selected rows of trees and between single trees within rows and GA_{4/7} variants were highly statistically significant (Table 1).

An interesting result seems to be a significant interaction between pruning and GA_{4/7}. Gibberellins applied without pruning or together with pruning done early (P₂₅), increased the number of long-shoots formed, while the GA_{4/7} combined with the later pruning (P₅₀) diminished the value of this character (Fig. 3).

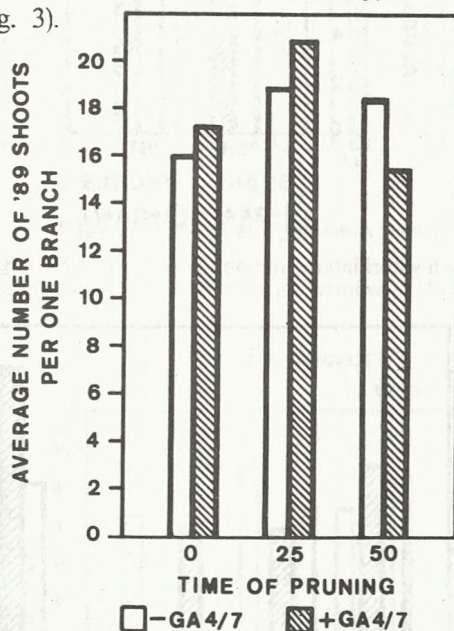


Fig. 3. Effect of GA_{4/7} × Pruning interaction on the number of '89 long-shoots

Other statistically significant interactions were also observed between GA_{4/7} and branch whorls (Fig. 4), GA_{4/7} and rows of trees (Fig. 5), GA_{4/7}, branch whorls and rows of trees (Table 1), and branch whorls and rows of trees (Fig. 6). All the interactions with GA_{4/7} were connected with very variable (positive and/or negative) effects of GA_{4/7} on the number of 1989 long-shoots that finally resulted in the lack of overall GA_{4/7} effect (Table 1). Pruning interacted also with stem whorls (Fig. 7), increasing the number of lateral long-shoots especially in the 1986 whorls, i.e. in the upper crown of the treated trees.

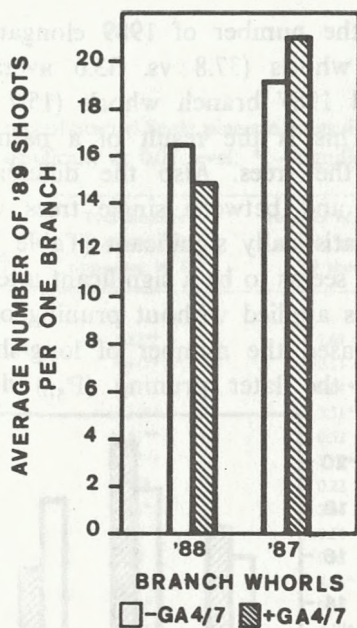


Fig. 4. Effect of $GA_{4/7} \times$ Branch whorl interaction on the number of '89 long-shoots in pruned trees

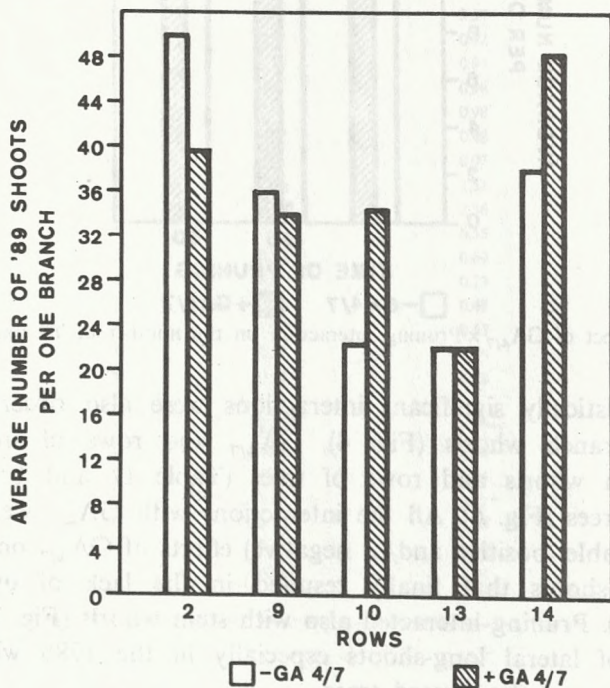


Fig. 5. Effect of $GA_{4/7} \times$ Rows of trees interaction on the number of '89 long-shoots in pruned trees

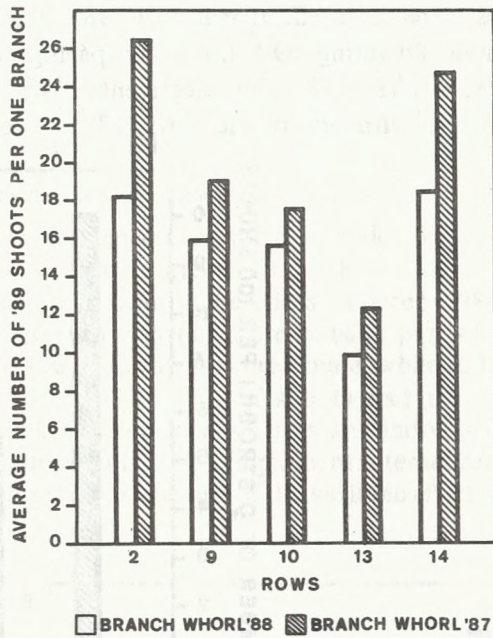


Fig. 6. Effect of Rows \times Branch whorl interaction on the number of '89 long-shoots in pruned trees

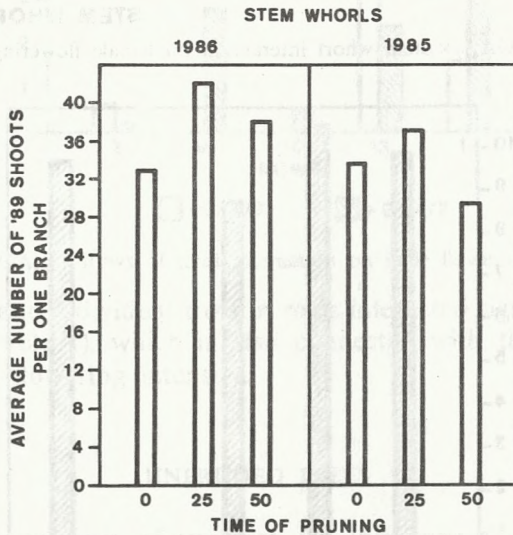


Fig. 7. Effect of Pruning \times Stem whorl interaction on the number of '89 long-shoots

NUMBER OF FEMALE STROBILI PER 100 SHOOTS

A highly significant effect of GA_{4/7} injection was observed on the female flowering of treated Scots pine seedlings (Table 1). The average number of female strobili per 100 shoots was 7.83 after GA_{4/7} treatment versus 0.15 in the

control. Gibberellins were more effective in the 1985 stem whorl, increasing the intensity of female flowering 69.5 times comparing to only 48.1 times in the upper 1986 whorl (Fig. 8). The effectiveness was also differentiated among the rows of trees with one of the rows (13) not responding (Fig. 9).

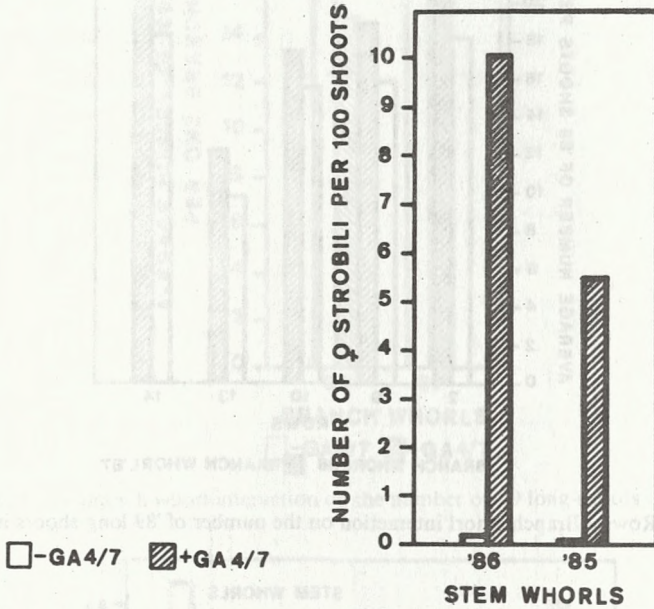


Fig. 8. Effect of $GA_{4/7} \times$ Stem whorl interaction on female flowering in pruned trees

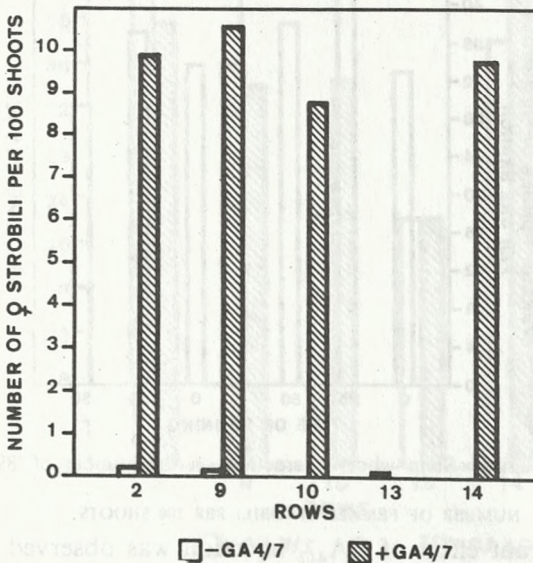


Fig. 9. Effect of $GA_{4/7} \times$ Rows of trees interaction on female flowering in pruned trees

The natural difference in the intensity of female flowering between the 1986 and 1985 stem whorls (5.16 vs. 2.82 respectively) was statistically confirmed. Significant differences between rows of trees and between individual trees in rows, and significant interaction between branch whorls and individual trees in rows was also observed (Table 1).

PERCENTAGE OF LATERAL SHOOTS WITH MALE STROBILI

GA_{4/7} injection influenced positively male flowering increasing the percentage of shoots with male strobili from 0.8% in the control to 1.8% in the treated trees. Highly significant differences between 1988 and 1987 branch whorls were also observed. On the more outside part of the crowns (branch whorls 1988) only 0.1% of shoots flowered male, while in the inside part of the crowns (branch whorls 1987) 3.6% of shoots had male strobili.

Natural variations between rows of trees and individual trees in rows were also statistically confirmed (Table 1). Significant interaction between GA_{4/7} and rows of trees was observed as a result of the variable effect (positive or negative) of GA_{4/7} (Fig. 10).

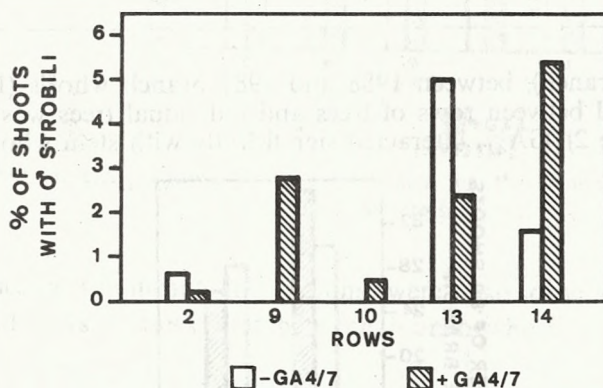


Fig. 10. Effect of GA_{4/7} × Rows of trees interaction on male flowering in pruned trees

Rows of trees and individual trees in rows interacted significantly with the branch whorls (Table 1), which is also connected with the individual tree variation in male flowering intensity.

UNPRUNED TREES

Results of the analysis of variance for this experiment are presented in Table 2.

NUMBER OF CURRENTLY ELONGATING LATERAL LONG-SHOOTS

High natural variation in the number of currently elongating shoots in 1989 between 1986 and 1985 stem whorls (32.6 vs. 27.4 average per

Table 2

The influence of $GA_{4/7}$ on flowering of unpruned Scots pine seedlings. Results of analysis of variance. Values of F: ** – significant at 0.01 level; * – significant at 0.05 level

Source of variation	Degrees of freedom	Total number of long-shoots elongating in '89	Number of female strobili per 100 '89 shoots	Percentage of '89 shoots with male strobili
Total	79			
Variables	39	6.27**	8.87**	10.70**
$GA_{4/7}$ (G)	1	1.24	35.79**	129.75**
Stem whorls (W)	1	18.56**	1.97	44.16**
Branch whorls (B)	1	51.93**	47.44**	55.37**
Rows (R)	4	7.26**	22.39**	7.93**
G × W	1	8.48*	4.84	15.34**
G × B	1	0.00	13.67**	15.34**
G × R	4	17.89**	16.21**	17.06**
W × B	1	2.30	0.06	0.52
W × R	4	4.32*	2.36	1.33
B × R	4	2.45	10.78**	3.70*
G × W × B	1	1.36	0.42	0.14
G × W × R	4	2.17	1.22	2.88
G × B × R	4	3.86*	6.87**	5.92*
W × B × R	4	0.78	0.60	0.29
G × W × B × R	4	1.48	0.05	0.01
Trees in R and G variants (T_{GR})	10	4.48*	11.59**	9.31**
$W \times T_{GR}$	10	1.31	0.80	1.86
$B \times T_{GR}$	10	3.44*	6.03**	3.22*
$W \times B \times T_{GR}$ (Error)	10			

one 1st order branch), between 1988 and 1987 branch whorls (12.8 vs. 17.2 respectively), and between rows of trees and individual trees was statistically confirmed (Table 2). $GA_{4/7}$ interacted significantly with stem whorls (Fig. 11),

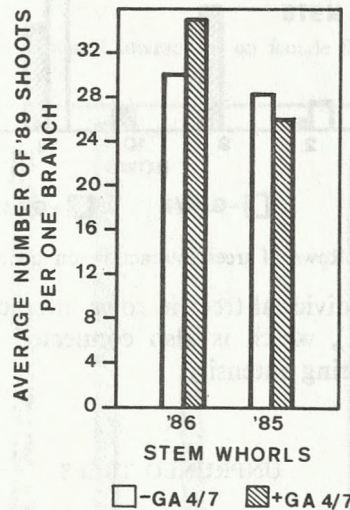


Fig. 11. Effect of $GA_{4/7}$ × Stem whorl interaction on the number of '89 long-shoots in unpruned trees

and rows of trees (Fig. 12), and with branch whorls and rows of trees (Table 2). The effect of $GA_{4/7}$ in these interactions was very variable

(positive or negative) which resulted in lack of overall GA_{4/7} influence on the number of 1989 shoots, similarly as in the experiment with pruned trees (Table 1).

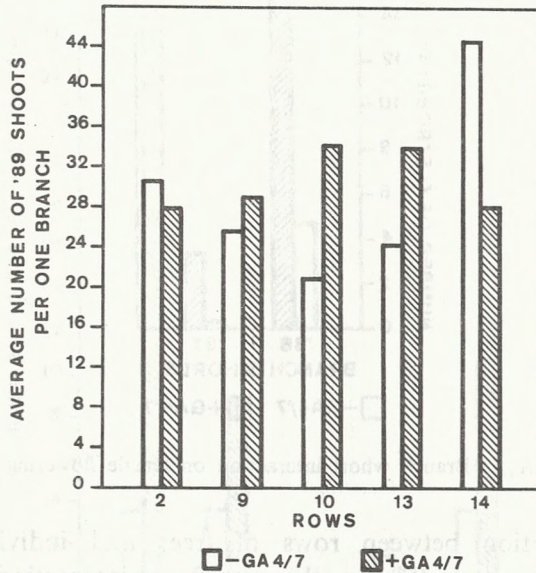


Fig. 12. Effect of GA_{4/7} × Rows of trees interaction on the number of '89 long shoots in unpruned trees

Statistically significant interactions were also observed between stem whorls and rows of trees, and between branch whorls and individual trees (Table 2).

NUMBER OF FEMALE STROBILI PER 100 SHOOTS

The GA_{4/7} treatment significantly increased the intensity of female flowering on unpruned trees (Table 2), increasing the number of female strobili per 100 shoots from 2.5 in the control to 10.7 in the injected trees. Significantly more strobili were observed in the outside branch whorl 1988 than on the inside 1987 one (11.4 and 1.9 respectively). The GA_{4/7} treatment interacted significantly with branch whorls being more effective on the inner 1987 whorl (Fig. 13). Other significant interactions were observed between GA_{4/7} and rows of trees (Fig. 14), and between GA_{4/7}, branch whorls and rows of trees (Table 2).

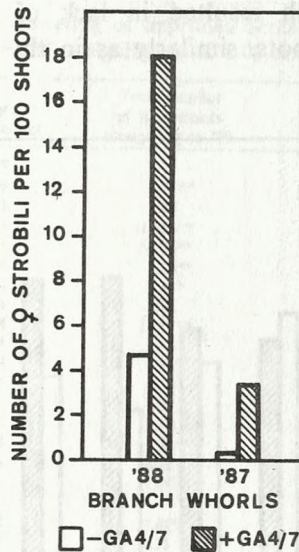


Fig. 13. Effect of $GA_{4/7} \times$ Branch whorl interaction on female flowering in unpruned trees

Natural variation between rows of trees and individual trees was statistically significant, and some other significant interactions were observed too (Table 2).

PERCENTAGE OF LATERAL SHOOTS WITH MALE STROBILI

The $GA_{4/7}$ treatment significantly influenced male flowering intensity increasing the percentage of shoots with male strobili from 0.4% to 11.5%. More of male strobili were located on the lower 1985 stem whorl than on the 1986 one (8.1% vs. 1.5%), and on the inside 1987 branch whorls than on the outside 1988 ones (8.5% vs. 1.3%).

Significant interactions were observed between $GA_{4/7}$ and stem whorls (Fig. 15), between $GA_{4/7}$ and branch whorls (Fig. 16), and between $GA_{4/7}$ and rows of trees (Fig. 17). In all these cases $GA_{4/7}$ promoted male flowering but with different effectiveness.

The intensity of male flowering was also significantly differentiated between rows of trees, and this was reflected in the interaction between branch whorls and rows, and between $GA_{4/7}$, branch whorls and rows of trees. Individual tree variation in the intensity of male flowering and the interaction between branch whorls and individual trees were also statistically confirmed (Table 2).

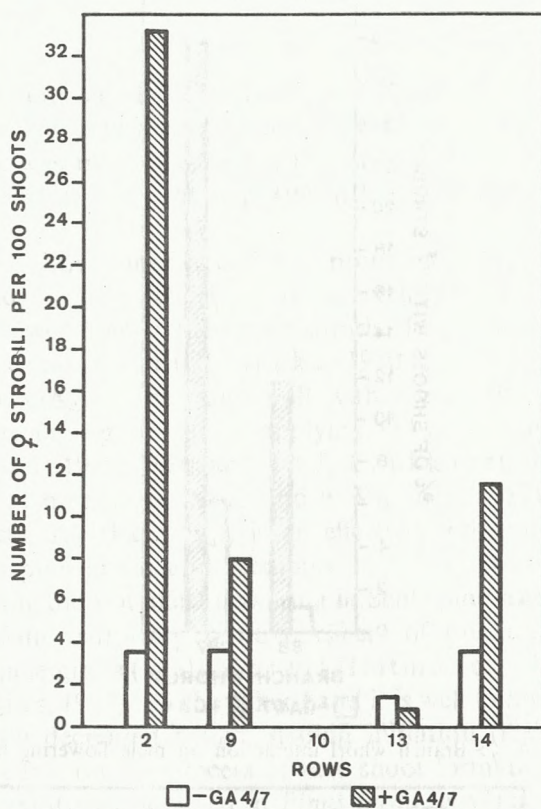


Fig. 14. Effect of GA_{4/7} × Rows of trees interaction on female flowering in unpruned trees

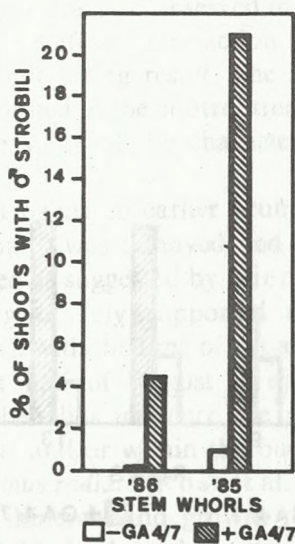


Fig. 15. Effect of GA_{4/7} × Stem whorl interaction on male flowering in unpruned trees

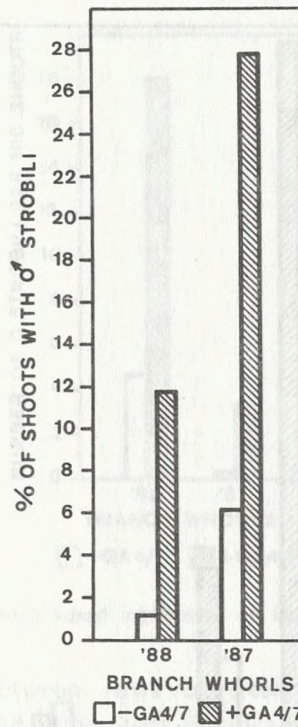


Fig. 16. Effect of $GA_{4/7} \times$ Branch whorl interaction on male flowering in unpruned trees

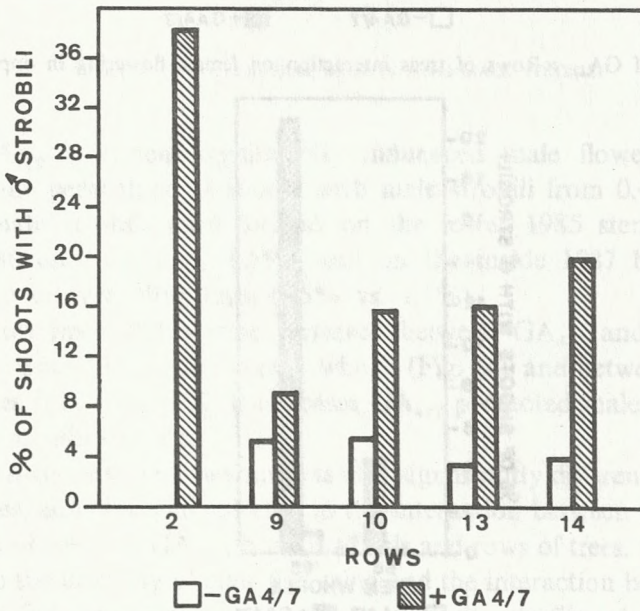


Fig. 17. Effect of $GA_{4/7} \times$ Rows of trees interaction in male flowering in unpruned trees

DISCUSSION

The GA_{4/7} injection significantly stimulated female and male flowering both in the pruned and unpruned 13-year-old seedlings of Scots pine. In the pruned trees GA_{4/7} was more effective in the promotion of female flowering than in the unpruned trees, and the opposite situation was noticed in the case of male flowering.

In the experiment with pruned trees GA_{4/7} promoted female flowering more effectively in the lower stem whorl where normally the number of female strobili initiated is lower than in the upper whorls. The same was observed in the crown of Scots pine grafts (Chałupka, 1980).

These effects of GA_{4/7} correspond well with the results of experiments conducted on Scots pine grafts when applying GA_{4/7} through spraying or injection (Chałupka, 1978, 1980 and 1987; Luukkanen 1980 and 1981; Luukkanen and Johansson, 1981a and b; Wesoły, 1987). Basing on all these results one can say that GA_{4/7} is an effective and well checked floral promotor in Scots pine grafts and seedlings.

The slight pruning did not affect flowering in Scots pine seedlings (Table 1). There are some data indicating positive effects of this treatment on the flowering of Scots pine grafts (Melchior and Heitmüller, 1960; Melchior, 1961; Danusevičius, 1987). On the other hand it is well known that intensive pruning significantly decreased female strobili initiation (Retkes, 1969).

GA_{4/7} had no effect on the process of new shoot formation in Scots pine seedlings. Similar result was obtained in *Pinus taeda* by Greenwood (1981). Only the earlier pruning (the P₂₅ variant) significantly increased the number of new lateral shoots formed, as was also observed in *Pinus ponderosa* (Giertych 1964). In this context the significant interaction between pruning and GA_{4/7} injection seems to be an interesting result. The applied GA_{4/7} increased the number of lateral shoots formed in the control trees and in those pruned earlier (P₂₅), and diminished the value of the character when applied to the later pruned trees (P₅₀) (Fig. 3).

One can presume that owing to earlier pruning of the branch leader the main sink for photosynthates was removed and their distribution within the branch whorls was changed as suggested by Giertych (1964), while the GA_{4/7} injected at the beginning of July supported the effect of pruning. Such suggestions correspond well with the time of initiation of long-shoot primordia, which starts in the first half of August (Hejnowicz, 1982). Also some indications exist that gibberellins influence the partitioning of photosynthates within the whorl of shoots and/or within the buds of *Abies balsamea* (Little and Loach, 1975), and *Pinus radiata* (Ross et al., 1984). It is also possible that other substances mainly minerals and growth regulators transported in the xylem become more available for lateral shoots after pruning. Such effect was

shown in grafts of *Picea abies* when covering them with polythene significantly increased the level of some mineral nutrients (Chałupka and Fober, 1990).

The results obtained and discussed above suggest a new procedure for the stimulation of flowering in Scots pine. The stimulation seemed to be more effective when removing of branch leaders was done at the beginning of shoot elongation. This treatment should be promotive for the formation of more lateral shoot primordia in the first growing season. The second step of the treatment would be GA_{4/7} injection at the proper time of the next growing season to induce more female and/or male strobili primordia on the increased number of lateral shoots previously induced by slight pruning. Such procedure would be worth checking in other experiments.

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Wpływ GA_{4/7} na kwitnienie przycinanych i nie przycinanych drzew sosny zwyczajnej (*Pinus sylvestris* L.)

Streszczenie

Mieszanina giberelin A_{4/7} podawana była poprzez iniekcję do pni 13-letnich sosen, których korony były lekko przycinane lub pozostawały nietknięte. W obu wariantach gibereliny istotnie zwiększyły liczbę zawiązanych kwiatów zarówno żeńskich, jak i męskich. Samo przycinanie pędów w koronie nie wpłynęło na kwitnienie, zwiększyło natomiast istotnie liczbę nowo formowanych bocznych długopędów. Na podstawie tych wyników zaproponowano dwustopniowe traktowanie sosny w celu zwiększenia liczby kwiatów: w pierwszym roku – delikatne przycinanie koron w celu zwiększenia liczby długopędów, natomiast w następnym roku – podawanie giberelin w celu indukowania zawiązków kwiatowych na tych długopędach.

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