Jour. Korean For. Soc. 84(4): 489 494, 1995

Shortening of Breeding Cycle by Forced Flowering in Forest Trees I. Induction of Flowering by Cultural Treatments in Seedlings and Grafts of *Betula pendula* Roth and *Betula platyphylla* var. *japonica* Hara<sup>1</sup>

Soo Hwan Ryu<sup>2</sup>, Yong Dong Ryo<sup>2</sup>, Mun Kyu Choi<sup>2</sup>, Won Kyu Choi<sup>2</sup>, Jae Woo Shim<sup>2</sup>, Keuk Soo Chung<sup>2</sup> and Min Sup Chung<sup>3</sup>

開花誘導에 의한 林木育種싸이클의 短縮 I. 자작나무와 은자작나무 幼苗에 있어서 裁培環境 調節에 의한 開花誘導<sup>1</sup> 류수환<sup>2</sup>·여영동<sup>2</sup>·최문규<sup>2</sup>·최원규<sup>2</sup>·실재우<sup>2</sup>·정국수<sup>2</sup>·정민선<sup>3</sup>

#### ABSTRACT

Seedlings and grafts of *Betula pendula* and *Betula platyphylla* var. *japonica* were grown under partially controlled environments in a greenhouse and in a plastic-greenhouse installed inside a laboratory. Plant growth conditions such as photoperiod, temperature, nutrient supply were partially controlled to enhance the vegetative and reproductive growth of the birch seedlings and grafts.

By the treatments twenty and seventy one percents of the seedlings, respectively, for the *Betula pendula* and *Betula platyphylla* var. *japonica* developed visible floral organs between 250 to 508 days after seeding. By the same treatments eighty and fifty three percents of the grafts, respectively, for *Betula pendula* and *Betula platyphylla* var. *japonica* developed visible male catkins between 51 to 497 days and female catkins between 365 to 396 days after grafting.

Breeding cycle of birch species can be reduced to a great extent by the induction of precocious flowering at early stages of seedling and graft development.

Key words: Betula spp. flowering induction.

### 要 約

자작나무 및 은자작나무 種苗와 接木苗의 營養生長과 生殖生長을 促進시키기 위하여 溫室과 실험실에 설치된 비닐하우스 內에서 日長,溫度,養分供給 등을 部分的으로 調節하여 促成裁培 處理를 하였다. 이와 같은 生長促進 處理에 의해 자작나무와 은자작나무는 播種 後 250-508일 사이에 각각 71% 및 20%의 幼苗가 암꽃 또는 수꽃을 形成하기 시작하였다. 또한 자작나무와 은자작나무는 接木 後 51일 내지 497일 사이에 각각 53%와 80%의 接木苗가 수꽃을 發育시켰으며 암꽃이 發育을 시작하기까지는 365일 내지 396일이 所要되었다.

幼苗에 대한 生長促進 處理에 의해 短期間 內 開花誘導에 成功 함으로써 林木의 改良을 위한 育種 싸이클과 新品種의 種子생산 期間을 劃期的으로 短縮시킬 수 있는 契機를 마련하였다.

<sup>「</sup> 接受 1995年 7月 31日 Received on July 31, 1995

<sup>&</sup>lt;sup>2</sup> Donghae Pulp Co. Ltd.(All retired now except K. S. Chung)

<sup>&</sup>lt;sup>3</sup> College of Agriculture, Kyungpook National University, 702-701, Taegu R. O. Korea

### INTRODUCTION

One of the most important factors that has to be considered in an active, ongoing tree improvement program is time required for every breeding cycle and for seed production. The progress in major parts of the tree breeding procedures can be achieved feasibly within a short period of time by inputting greater manpower and efforts. However, production of genetically improved seeds is impossible until the parental trees, that have been selected for their genetic potentialities, begin to flower.

Many of the important tree species do not commonly attain sexual maturity for a few to many years. For commercial seed production in a short period, forest tree breeders will not select parental trees flowering at early ages which might be associated with reduced timber production. But they will select parental trees that grow vigorously until late ages before they begin to flower and try to induce flowering at early ages by controlling physiological processes of the trees to shorten the generation intervals and so speed up breeding programs.

Up to date, cultural and growth regulator treatments are the major methods used to induce precocious flowering in perennial tree species (Greene & Lord 1978, McLaughlin & Greene 1984, Ho 1988, Ross 1990, Sheng, & Wang 1990, Greenwood *et al* 1991, Harrison & Slee 1991, McLaughlin & Greene 1991a, 1991b, Wilkinson & Richards 1991). One of the prominent works for the induction of precocious flowering in forest trees has been accomplished and practised in commercial scale by Finnish tree breeders since early 1970's(Anonymous 1976).

In 1970, research workers at Foundation For Forest Tree Breeding in Finland induced flowering successfully in *Betula pendula* seedlings in a plastic-greenhouse with controlled environment temperature control, nutrients supply, and carbon dioxide enrichment in the ambient air etc. In the following year twenty four of the sixty(40%), in the two to four year-old birch

seedlings that had been treated for flowering induction produced about 1.5kg of seeds(about 4.5 millions in number)(Lepisto 1973). The third year of flowering induction in 1972, fifty five(92%) of the sixty birch seedlings produced about 23kg of seeds(about 47 millions of seeds). The results enable the tree breeders at Foundation For Forest Tree Breeding to establish two-clone seed orchards in plastic-greenhouses and produce large quantity of newly bred valuable hybrid birch seeds within a few years and thus shortened the breeding cycle of birch species dramatically.

The aim of this study was to find out a proper way for the induction of precocious flowering in *Betula pendula* and *Betula platyphylla* var. *japonica* with similar method developed by the Finnish research workers under Korean environmental conditions and to produce a large quantity of improved birch seeds within a few years.

# MATERIALS AND METHODS

Seedlings and grafts of *Betula pendula*(Finnish seed source) and *B. platyphylla* var. *japonica* (Korean and Japanese seed sources) were grown in a plastic-greenhouse installed inside a laboratory at Kyungpook National University in Taegu city(35° 53′ N, 128° 37′ E) and in a greenhouse at Samcheuk(37° 22′ N, 129° 06′ E) Kwangwondo province.

The seedlings in the plastic-greenhouse and greenhouse were grown under 20-hour day(about 12 to 14-hour natural incident light plus 6-8 hours of supplemental light mainly with high pressure sodium lamps(80%), and mercury lamps (20% in electrical wattage) during night. Total radiant flux by the supplemental light was approximately 160-196 J/m²/sec.(maximum 250 J/m²/sec. at the top of tree crown) at tree crown level. Temperatures in the plastic-greenhouse and greenhouse fluctuated considerably during the night depending on the outdoor temperatures. But temperature during the induction period was maintained to 15-18% (min. 12%) at night and 24-30% (max. 38%) during the day.

To enhance the seedling growth, carbon dioxide concentration of ambient air in the plastic greenhouse and the greenhouse was elevated up to 800 1200 ppm by gas burners throughout the period of seedling growth until the seedlings develop reproductive organs visibly.

The birch seedlings were transplanted into plastic pots(33cm both in diameter and height) when they were about 10cm in height, and regularly watered with nutrient solution contain ing the following ingredients:

KNO <sub>3</sub>	5mM
H <sub>3</sub> BO <sub>3</sub>	0.046mM
KH <sub>2</sub> PO <sub>4</sub>	lmM
ZnSO <sub>4</sub> ······	0.08mM
Ca(NO <sub>3</sub> ) <sub>2</sub>	4mM
CuSO <sub>4</sub> ·····	0.003mM
MgSO <sub>4</sub>	3mM
FeEDTA	0.003mM
(NH <sub>4</sub> ) <sub>2</sub> MoO <sub>24</sub>	0.001mM

When height of the seedlings reached about one meter which believed to be a proper develop mental stage for flowering induction the same kind of nutrient solution but with reduced KNO<sub>3</sub> (3mM) and increased KH<sub>2</sub>PO<sub>4</sub>(3mM) was supplied every other day thereafter.

## RESULTS AND DISCUSSION

It has been shown that the growth of birch seedlings can be greatly enhanced by controlling the environmental factors in which the seedlings are growing(Krizek *et al* 1969, Krizek & Zimmerman 1973). Major favorable conditions for

the fast growth of birch seedlings are high day/night(30/18°C) temperatures with high light intensity(26.9 klx. or higher light intensities) during the day, sufficient nutrient supply and carbon dioxide enrichment of the ambient air(400 2,000 ppm). Some of the birch species and/or races may require long day conditions(14 hour or longer) for rapid growth of the seedlings.

The growth of birch seedlings and grafts at present work was very rapid in response to the controlled environmental conditions, particularly to high temperatures during the day and continuous nutrient supply with elevated carbon dioxide concentrations in the ambient air. Though the precise measurements were not made, the growth of *B. pendula* seedlings was visibly faster than that of *B. platyphylla* var, *japonica* under long day(20 hour photoperiod) conditions.

Betula pendula seedlings began to develop floral organs between 250 508 days after seeding during the first induction cycle(Table 1). Four(20%) of the twenty treated seedlings developed male catkins and only 5% of the treated seed lings developed female catkins after 440 days from seeding.

In case of *B. platyphylla* var. *japonica* five (71.4%) of the seven treated seedlings developed only male catkins between 309 410 days after seeding(Table 2). In this case the rate and duration of flowering induction appeared to be an acceptable level for practical scale for shortening the breeding cycle of birch species provided that the induced seedlings produce female catkins as well as male ones in successive years.

Induction of flowering for the grafts of B. pendula appeared even more effective than the

Table 1. Flowering induction for the seedlings of Betula pendula

Tree number	Date of seeding	Date of the first catkin development	Date of flowering	Number of catkins	Days required for the first visible catkin development from seeding
288-1	1989. 1. 3	1989, 9. 10 M.	1990, 4. 2	5	250 days
1	,	1990, 3. 19 F.	1990, 4. 2	6	440 days
288-2	"	1990, 5. 26 M.		1	508 days
288-3	"	1990, 5. 25 M.		5	507 days
288 - 4	"	1990, 5. 25 M.		12	507 days

M.: male catkin F: female catkin percentage of seedlings that developed male catkins:  $4/20 \times 100\% = 20\%$ 

Table	2.	Flowering	induction	for	the	seedlings	of	Betula	platyphylla	var.	japonica.
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Tree of number	Date seeding	Date of the first catkin development	Number of catkins	Days required for the first visible catkin development from seeding
P0- 1	1989, 4. 26	1990, 6, 10 M	1	410 days
PJ17	9 9 0	1990, 3. 1 M	23	309 days
PJ 18	у у н	1990, 6. 4 M	12	404 days
P11~3	n n n	1990, 6. 7 M	1	407 days
P11-5	и и и	1990, 6, 4 M	21	404 days

M: male catkins

percentage of seedlings that developed male catkins:  $5/7 \times 100 = 71.4\%$ 

**Table 3.** Flowering induction for the grafts of Betula pendula.

Tree number	Date of grafting	Date of the first catkin development	Date of flowering	Number of catkins	Days required for visible catkin development from seeding
Pn 1-5	1989, 3, 20	1989, 5, 10		2 M	51 days
Pn 1-10	1989, 3, 20	1990, 3, 19	1990, 4. 1	2 F	365 days
Pn 5~ 2	1989, 7.20	1990. 4. 10	1990. 4. 7	34 M	264 days
Pn 5- 5a	1989, 11. 6	1990, 1. 6		3 M	61 days
Pn 5- 5b	n n n	1990, 6, 28		1 M	234 days
Pn 6 7	y n n	1990, 4, 10		1 M	155 days
Pn 7= 1	1989, 2.13	1989, 7, 30		3 M	167 days
Pn 7- 2	n n	1990. 5. 27		12 M	103 days
Pn 7- 5a	и и и	1990, 5, 10	1990, 4. 1	1 M	86 days
Pn 7- 5b	u u u	1990, 5, 28		2 M	104 days

M: male catkin, F: female catkin

percentage of grafts that developed male and/or female catkins 9/10 100% = 90.0%

Table 4. Flowering induction for the grafts of Betula platyphylla var. japonica.

Tree number	Date of grafting	Date of the first catkin development	Date of flowering	Number of catkins	Days required for the first visible catkin development from seeding
P 31-5	1989, 2, 16	1990. 3. 19F	1990. 4. 2	6	396 days
P 31 7	1989. 2. 16	1989. 9. 10M	=	18	206 "
		1990. 3. 19F	1990. 4. 2	4	396 "
P 31- 8	1989. 2. 18	1990. 6. 10M	-	19	477 "
P 32- 7	1989. 3. 14	1990. 6. 4M		5	447 "
P 39- 8a	1989. 2. 21	1990. 5. 27M		21	460 "
P 39= 8b	1989. 2. 21	1990. 7. 3M		36	497 "
P 39= 9	1989. 2. 21	1990. 6. 10M	-	1	474 "
P 39-11	1989. 2, 21	1990, 6, 28	-	19	492 "
P 39-12	1989. 2. 21	1990. 5. 27M		19	460 "
P 40-35	1989. 2. 20	1990. 5. 25M		24	459 "

M: male catkin

F: female catkin

percentage of grafts that developed male and/or female catkins:  $10/15 \times 100\% = 66.7\%$ .

seedlings of the same species(Table 3). Ninety percent of the treated grafts developed male and/or female catkins within a year. Some of the grafts developed male catkins in two months and female ones in a year. Although there was no statistical difference(P=0.20 for induction rate) due to small number of sample sizes the rate and duration of flowering induction in grafts of *B. platyphylla var. japonica* appeared to be lower and longer than those of *B. pendula*(Table 4). In all cases the seedlings and grafts of *B. pendula* and *B. platyphylla* var. *japonica* developed female catkins when the seedlings were moved out and exposed to natural conditions outside(10 to 14.2-hour photoperiods).

According to Mr. Lauri Kärki at Foundation For Forest Tree Breeding(personal communication in 1976) some of the birch seedlings grown under controlled environment in a plasticgreenhouse developed male catkins when the height of seedlings reached 150-200cm at age of seventy seven days. However, it took two hundreds undreds and fifty, and three hundreds and nine days, respectively for B. pendula and B. platyphylla var. japonica under controlled environmental conditions at present work in Korea. Only some of the treated grafts of B. pendula developed male catkins between 51 to 86 days after grafting at present work. Comparing natural conditions during the treatments for flowering induction between Finland and Korea. the quantity of incident solar energy with optimal level appears to be one of the major factor in shortening the juvenile period of birch seedlings. The quantity of incident solar energy, effective for plant growth during the summer in Finland is much greater than in Korea.

Two clones(grafts) of *Betula pendula* treated for flowering induction(just one cycle treatment for eight months) did not flower up to age five to six. One of those clone began to flower at age six and the other at age seven after they had been treated with succinic acid 2,2-dimethylhydrazide for two successive years before flowering. Thus it appears that some of the birch genotypes require longer induction period than eight months for a successful flowering induction.

Maintaining optimum conditions for flowering induction in a plastic-greenhouse during the hot summer season in this region is technically difficult and thus the seedlings that had been treated for flowering induction were moved

outside for two months during July to August. Probably, this kind of interruption during the induction period appeared to delay flowering induction of the birch seedlings.

In conclusion, flowering induction simply by cultural treatments in greenhouse and/or plastic-greenhouse is possible at early developmental stages of seedlings and grafts in *Betula pendula* and *Betula platyphylla var. japonica*. This can be done with a reasonable cost and permissible efficiency under Korean climatic conditions.

### ACKNOWLEDGEMENTS

We are very grateful to Professor Emeritus Max Hagman(former research worker at Finnish Forest Research Institute) and Susumu Mikami, Tohoku Research Center, Forestry and Forest Product Research Institute in Japan who prepared seed materials for *B. pendula* and *B. platyphylla* var. *japonica* respectively for this study. We gratefully acknowledge Risto Hagqvist, M. Lepisto, and Jouni Mikola, Foundation For Forest Tree Breeding in Finland who prepared seed materials of *B. pendula* and useful informations for this study.

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