

## Old colchicine-induced polyploid materials of *Betula pendula* Roth and *Betula pubescens* Ehrh.

K. Pieninkeroinen and T. Valanne

Department of Biology, University of Turku, SF-20500 Turku, Finland

### Introduction

The members of the genus *Betula* form a particularly significant group of broad-leaved trees in Eurasia and North America. Certain birch species, e.g., *B. pendula*, *B. pubescens* and *B. papyrifera*, are valuable sources of wood and great importance is attached to breeding work aimed at their economic improvement.

Polyploidy induced by colchicine treatment in *B. pendula* and *B. pubescens* has been reported by Johnsson and Eklundh (1940), Schröck (1951) and Valanne (1972). The rate of growth of induced *Betula* polyploids has been observed to be inferior to that of the diploid parental trees (Johnsson, 1956; Eifler, 1955; 1967). It has also been reported that triploid *B. pendula* trees obtained from cross-pollination of colchicine tetraploid and diploid trees did not grow faster than the diploid control plants (Johnsson, 1956). The aim of this study was to reveal possible breeding and evolutionary trends in *B. pendula* and *B. pubescens*.

### Materials and Methods

The material was part of about 2000 individually numbered ca 25 yr old *B. pendula* and *B.*

*pubescens* trees, which originated from extensive colchicine experiments with different birch species carried out in Finland (Valanne, 1972). The birches from the experiments are located on the island of Seili (60°12' N, 21°55' E), at the Experimental Station of the Finnish Forest Research Institute, Punkaharju (61°43' N, 29°25' E), at Päilahti by Orivesi (61°37' N, 24°29' E) and in the Botanical Garden of the University of Turku, Ruissalo (60°26' N, 22°10' E). From the materials of Punkaharju, Orivesi and Turku, 50 *B. pendula* and 50 *B. pubescens* trees were taken for this study.

The chromosome counts were carried out according to the method of Hömmö and Särki-lahti (1986), using young leaves. Arnott's (1959) method of clearing leaves was used to study stomata. For the measurement of wood fibres and vessels, wood tissue was macerated according to the method for broad-leaved trees.

### Results

The retarding effect of colchicine on the height growth of birches is strong. In the colchicine-induced polyploids, both the height and the diameter (dbh) were significantly smaller than in the colchicine-treated chromosomally normal trees (Table I). Some diploid *B. pendula* trees reached a height of nearly 20 m, while the highest polyploids were under 14 m. The colchi-

**Table I.** The mean ( $\pm$  SE) height and dbh in chromosomally normal and colchicine-polyploid trees of *B. pendula* and *B. pubescens*.

	n	Height (m)	F	dbh (cm)	F
<i>B. pendula</i>					
2x = 28	24	15.5 $\pm$ 1.0		20.2 $\pm$ 1.3	
4x = 56	26	8.1 $\pm$ 0.6	41.3***	11.5 $\pm$ 0.9	32.1***
<i>B. pubescens</i>					
4x = 56	7	15.2 $\pm$ 0.3		19.9 $\pm$ 0.6	
6x, 7x, 8x	43	7.4 $\pm$ 0.3	97.6***	10.1 $\pm$ 0.6	39.7**

Differences tested with analysis of variance, significance: \*\*\*  $P < 0.001$ , \*\*  $P < 0.01$ .

**Table II.** The mean ( $\pm$  SE) length of guard cells of stomata, wood fibres and vessels of chromosomally normal and colchicine-polyploid trees of *B. pendula* and *B. pubescens*.

	Stomata ( $\mu$ m)	Wood fibres ( $\mu$ m)	Wood vessels ( $\mu$ m)
<i>B. pendula</i>			
2x = 28	31.7 $\pm$ 0.2	664.6 $\pm$ 5.6	458.0 $\pm$ 4.5
4x = 56	44.2 $\pm$ 0.3***	892.0 $\pm$ 4.0***	635.2 $\pm$ 3.3***
<i>B. pubescens</i>			
4x = 56	37.6 $\pm$ 0.3	687.9 $\pm$ 10.6	467.9 $\pm$ 6.7
6x, 7x, 8x	54.0 $\pm$ 0.2***	924.6 $\pm$ 3.6***	687.4 $\pm$ 2.8***

Differences tested with analysis of variance, significance: \*\*\*  $P < 0.001$ .

cine treatment caused branching. No statistically significant difference was found in branch numbers between the groups.

The petioles of polyploid *B. pendula* trees were thicker than those of diploid ones, containing more parenchymatous and vascular tissue. In the tetraploids, the cell size of the parenchymatous tissue was greater in relation to other cell types than in the diploid trees. In the polyploid trees, the sclerenchymatous tissue around the vascular bundle seemed to be more abundant and its cells often had thinner walls than in the diploids.

The guard cells of the stomata were significantly longer in the induced polyploids than in the colchicine-treated chromosomally normal trees (Table II). The lengths of the guard cells of the normal trees were on an average 70% of those of the induced polyploids.

In the colchicine polyploids, the wood fibres and vessels were statistically significantly longer and the vessels wider than in the chromosomally normal trees (Table II). The mean lengths of the fibres and vessels of polyploid *B. pendula* were several tens of micrometres smaller than those of polyploid *B. pubescens*. In some *B. pubescens* trees with the longest wood fibres, the mean length exceeded 1 mm. The shortest wood fibres were recorded in some diploid *B. pendula* trees, the mean value being under 0.6 mm.

## Discussion and Conclusion

Retardation of the height growth is a typical effect of colchicine in the *Betula* species studied (Table I). This has already

been seen in the initial stages of saplings (Johnsson and Eklundh, 1940; Valanne, 1972). On the other hand, the cell size of colchicine polyploids is greater (Table II), and the leaves and leaf organs are greater than at the diploid level.

The chromosome number of a considerable part (33 trees) of the *B. pubescens* material is heptaploid with  $7x = 98$ , and only a small number of the trees have the expected  $8x = 112$  (6 trees). The large proportion of heptaploid trees suggests that the chromosome set of *B. pubescens* consists of  $42 + 14$  chromosomes ( $3x + x$ ), and that in the colchicine treatments 42 chromosomes have been duplicated, while 14 chromosomes are unchanged. In our experiments with birches of the subsection *Nanae* (*B. glandulosa* and *B. nana*, both  $2n = 28$ ,  $x = 14$ ), no colchicine polyploids have been obtained. In the literature, it has been suggested that the *B. pubescens* genome contains a genome of the subsection *Nanae* (e.g., Walters, 1968). The abundance of trees with  $7x = 98$  occurring in our *B. pubescens* material supports this suggestion.

## References

- Arnott J. (1959) Leaf clearings. *Turtox News* 37, 192-194
- Eifler I. (1955) Künstliche polyploidie-erzeugung bei *Picea abies* und *Betula verrucosa*. *Z. Forstgen. Forstpflanzenzücht.* 4, 162-166
- Eifler I. (1967) Anwendungsmöglichkeiten der polyploidie-züchtung in der forstwirtschaft. *Arch. Forstwes.* 16, 515-528
- Hömmö L. & Särkilahti E. (1986) A method of counting chromosomes of hardwood trees using root tips and young leaves. *Can. J. For. Res.* 16, 401-403
- Johnsson H. (1956) Auto- and allotriploid *Betula* families, derived from colchicine treatment. *Z. Forstgen. Forstpflanzenzücht.* 5, 65-70
- Johnsson H. & Eklundh C. (1940) Colchicinbehandling som metod vid växtföredling av lövträd. *Sven. Papperstidn.* 43, 355-360, 373-377
- Schröck O. (1951) Stimulierende wirkung des colchicins bei der keimung und wachstum der sämlinge. *Züchter*, 21, 142-149
- Valanne T. (1972) Colchicine effects and colchicine-induced polyploidy in *Betula*. *Ann. Acad. Sci. Fenn. Ser. A4 Biol.* 191, 1-28
- Walters S.M. (1968) *Betula* in Britain. *Proc. Bot. Soc. Br. Isl.* 7, 179-180