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Performance and Wood Quality of *in Vitro* Propagated Hybrid Curly Birch (*Betula pendula* x *Betula pendula* var. *carelica* SOK.) Clones

By D. EWALD, G. NAUJOKS and H. PIEGERT[1])

Federal Research Centre for Forestry and Forest Products, Institute for Forest Genetics and Forest Tree Breeding, Eberswalder Chaussee 3, D-15377 Waldsieversdorf, Germany

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Abstract

Selected adult hybrid curly birch clones were multiplied by tissue culture. The donor trees originated from breeding experiments which sought to combine straight stem form and a strongly curled wood structure. Micropropagated plants were planted by machine in rows in a field trial. After ten years the height and breast height diameter of the clones were measured and the wood quality of two clones was assessed by felling sample trees to observe wood grain and veneer quality. Results show that tissue culture propagation methods are a valuable tool for the „true-to-type" propagation of birch with regards to outstanding wood qualities.

Key words: Betula pendula x *Betula pendula* var. *carelica* SOK., curly birch, micropropagation, field performance, veneer.

Introduction

Different types of valuable birch wood have been described in the literature (SCHROCK and SCHOLZ, 1953). Several attempts have been carried out to propagate selected adult trees of birch with certain wood qualities, but with only limited success (SCHOLZ, 1960). Intensive breeding experiments were conducted in the 1950's at the Institute for Forest Tree Breeding at Waldsieversdorf to combine stem straightness from selected white birch (*Betula pendula* ROTH) with curled wood quality of curly birch (*Betula pendula* var. *carelica* SOK.) selected from natural stands in Poland (SCHOLZ, 1963). Valuable curly-grained wood of birch is characterised by the inclusion of bark particles into the wood, visible as brown curls in vertical sections of the trunk or as rings or V-shaped figures in horizontal sections of the trunk.

These experiments showed that the combination of both characters (straight stems and curled wood) in one individual was a rare event. Thus, the production of large numbers of straight stemmed, curled wood plants via seed seems improbable. Although infrequently, some offspring did display the desired combination of stem straightness and curled wood along the whole stem. Cuttings obtained from topped (severely hedged) trees were used to vegetatively propagate the desired specimens with combined straightness and curled wood (MATSCHKE and SCHNECK, 1981). With the availability of suitable tissue culture methods for propagation of birch (CHALUPA, 1981, 1989; RYYNÄNEN and RYYNÄNEN, 1986; MATSCHKE *et al.*, 1987) there was an opportunity to produce large quantities of desirable curly birch if stem straightness and curled wood

[1]) Forest Office Güntersberge, Forsthaus Uhlenstein, D-06507 Siptenfelde, Germany

qualities were retained in tissue culture plantlets. This question is addressed in this report.

Material and Methods

Plant material

Among the offspring of former crossbreeding experiments (SCHOLZ, 1961) between shrub-like *Betula pendula* var. *carelica*, with excellent curled wood quality, and straight stemmed, fast growing *Betula pendula* individuals, four clones were selected which showed curled wood properties. Two clones (1/86 and 2/86) were selected in older field trials (10 to 20 years-old). The stem form in these two clones was known to be straight with a good diameter and curled grain along the whole stem. Two other clones (3/86 and 6/86) were selected from younger offspring (5 to 10 years-old) and were characterised by extremely curled wood.

Micropropagation

For the *in vitro* culture establishment, twigs with dormant buds were harvested from the crown of the original trees. Shoot segments (3 cm to 4 cm) were rinsed for a few minutes in 0.2% Euparen (fungicide by BAYER, 50% dichlorfluanide, w/v), then immersed in 0.25% mercuric chloride ($HgCl_2$) solution containing a few drops of Tween 80 for 17 minutes with gentle shaking, followed by three rinses with sterile distilled water. The bud scales were aseptically removed and the green shoot tips were placed vertically in the nutrient medium.

The nutrient medium was a modified LS medium (LINSMAIER and SKOOG, 1965) containing ⅓ strength macro and micro nutrients as well as vitamins supplemented with 1% sucrose, 0.1 mg/l kinetin, 0.8 mg/l 6-benzyl-aminopurine (BAP) and 0.004 mg/l 1-naphthalene acetic acid (NAA). As gelling agent 7.5 g/l SERVA agar (gel strength ~1000) was added. The explants were grown in test tubes (diameter 28 mm, 95 mm high) placed in a culture room maintained at 20°C to 22°C, illuminated with warm white fluorescent tubes at a light intensity of 1600 lux to 1700 lux (27.5 µmol to 28 µmol m^{-2} s^{-1}) during 16 hours per day.

After an initiation phase of up to three months callus formation started followed by adventitious and axillary shoot development. During subculture periods of one month good proliferating shoot clusters were used for multiplication. Elongated curly birch shoots were rooted on a medium following BOULAY (1979) with ½ strength macro and micro nutrients including vitamins, 0.5% sucrose and 0.1 mg/l indole-3-butyric acid (IBA) + 0.05 mg/l NAA and solidified with 7.5 g/l SERVA agar (gel strength ~1000). After 8 to 14 days the rooted plantlets were transferred to a standard soil:sand mixture (3:1) and kept under high air humidity maintained by mist irrigation under plastic foil cover. About 4 weeks later plantlets were acclimatised by successively reducing the air humidity and afterwards grown in the nursery up to 60 cm to 80 cm height.

Field trial establishment and data collection

A field trial was established with 2 year old micropropagated plants of the four selected curly birch clones. Trees were machine planted in rows (2 m x 1 m spacing). The number of individuals planted from each clone is given in *table 1*.

Table 1. – Number of curly birch plants grown in the field trial.

Clone label	Number of plants
1/86	635
2/86	181
3/86	477
6/86	40

The field trial was planted in 1985 in the Harz Mountains near the city Helbra in Germany. The soil of this area was contaminated from mining industries with heavy metals (Cu, Hg, As, Cd). After 10 years, the height of 20 randomly chosen plants of each clone was measured. The breast height diameter was measured only from two clones (1/86 and 2/86) which had good stem form. Average values, including the standard deviations, were calculated. Data were compared using the PROC GLM procedure (TUKEY-test) of the SAS program (SAS Institute Inc.). Two trees of clone 1/86 and 2/86 were harvested. The stems were cut into logs one meter in length and the wood quality (curled wood) was evaluated from top to bottom on the cut surface of each log (wood with the inclusion of bark spots – see *Figure 1C*).

Results and Discussion

After micropropagation and a period of 2 years in the nursery, plants of all clones showed similar growth (around one meter in height) and were characterised by straight stems (*Figure 1A*).

Ten years after establishing the field trial, each clone looked very uniform, although there were differences between the clones. The within clone uniformity suggested that machine planting is suitable for tissue cultured birch, but a direct comparison of machine versus hand planting experiments is needed to prove this point. The average performance, characterised by height and breast height diameter of the clones, is depicted in *figure 2*. The growth of the hybrid curly birch plants from tissue culture was evaluated using data presented by MEIER-DINKEL (1996) for an eight year-old field trial with micropropagated clones and seedlings of common birch. His plants showed an yearly increment of about one meter and a nearly linear growth behaviour. Starting with these results a height of 10 m for seedlings and micropropagated plants after 10 years in the field can be expected. The height performance of the best two curly birch clones (1/86 and 2/86) was lower (7.29 ± 0.44 m, 7.11 ± 1.01 m respectively) compared with the data of MEIER-DINKEL (1996) for selected birch clones and seedlings of *Betula pendula*. This seems to confirm former statements (SCHOLZ, 1961) that the curly wood quality is often an expression of a disturbed growth behaviour. The results obtained are nevertheless optimistic for practical use in a larger scale. Among the four clones used, those clones (3/86 and 6/86) selected earlier (5 to 10 years), in the absence of sufficient information concerning stem straightness, showed the shrub-like growth, typical for the original curly birch which was used as one parent in the crossbreeding experiments. This result was unexpected because the growth habit of the selected young plants from these clones, as well as the micropropagated plants, in the nursery did not differ from the other clones (1/86 and 2/86) with regard to stem straightness (*Figure 1A*). This result strongly suggests that only clones of proven superiority tested for long periods should be introduced into commercial micropropagation operations. The importance of a sufficient documentation of the growth parameters before introduction into tissue culture was also stressed for birch micropropagation by VIHERA-AARNIO (1994).

The two superior clones (1/86 and 2/86) were curled from top to bottom. The clone with the best growth (2/86) showed a wood quality different from the other one (1/86). Its wood was not as curled (*Figure 1C*, above) as the second clone. Brown bark spots were also included in the annual rings. The second clone (1/86) showed wedge-like inclusions in the wood which seem to

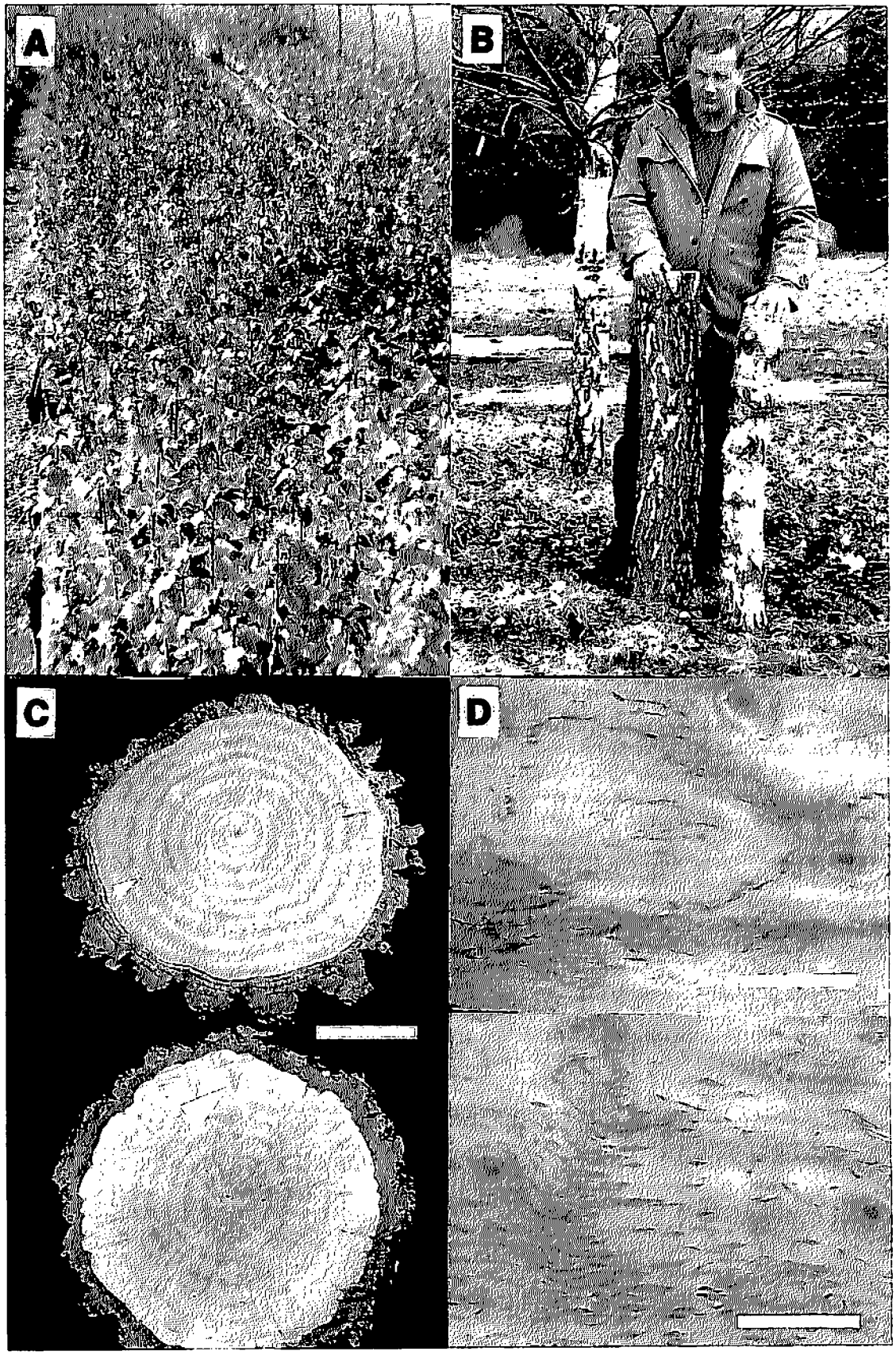

Figure 1. – A) Micropropagated curly birch plants in the nursery in September. B) Logs of ten-year-old curly birch clone 1/86 (right) and clone 2/86 (left). C) Wood quality of the two clones 1/86 (below) and 2/86 (above), in the horizontal cuts of the trunks, the small arrow marks brown spots, the broad arrow marks a V-shaped structure, bar represents 5 cm. D) Veneer samples of the two micropropagated clones 1/86 (below) and 2/86 (above), bar represents 5 cm.

Number of plants measured: 20. Columns with different letters are significantly different at $p < 0.05$ level (TUKEY-test)

Figure 2. – Performance of different micropropagated curly birch clones after ten years in a field trial (values are means including standard deviation).

originate from overgrown branches or twigs. This clone had a more bulbous outer surface of the stem. The basal logs were straight as in the case of ordinary birch (*Figure 1B*). These results from older selections show that micropropagated plantlets do retain the quality characteristics of the parent tree.

Curly birch was used as crossing partner for experiments about 40 years ago. Some trees selected from the older progeny trials are analysed in the EU project „Wood Quality in Birch" which is dealing with the characterisation and validation of wood properties in birch for industrial use and future breeding (running from 1.4.1999 till 31.3.2002, for more information see http://www.tv.slu.se/birch/).

Critically selected individual adult birch trees subjected to micropropagation may provide a practical means for producing large numbers of uniform high value wood products.

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