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ARTICLES

Influence of Parent Tree Characteristic on Propagation of Hybrid Aspen by Root Cuttings

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Abstract

The influence of parental tree's age on root cutting sprouting capacity was studied. It was found that the age of a parental tree had a great effect on sprouting capacity of aspen's and its hybrids root cuttings. The age showed a strong and significant correlation with the percentage of sprouted root cuttings. Younger parental trees yielded a higher percentage of sprouted root cuttings.

The results also showed that root cutting diameter had an influence on offspring biometric parameters. A hypothesis that larger root cuttings accumulate higher amounts of nutrients and thus stimulate plant development was raised. This hypothesis was later confirmed by an actual finding that larger offspring are produced from larger root cuttings under uniform growth conditions.

The results showed that higher sprouting capacity could be expected when the *P. tremula x P. tremuloides* hybrid rather than that of *P. tremula x P. alba* was used for root cutting preparation. It was found that sprouting percentage of *P. tremula x P. tremuloides* root cuttings reached 15% while that of *P. tremula x P. alba* root cuttings was just 9%.

Key words: aspen, root cutting, seedlings, height, diameter

Introduction

Naturally, aspen reproduces by seed and root sprouts. Aspen fruits abundantly almost every year, however, due to a very short seed viability period only small portion of the seed germinate and seedlings survive. The seed remain viable for 1-2 weeks only. Imbibed seed can stay viable for just 2-3 days (Farmer et al. 1963).

Aspen can be propagated on a large scale by micropropagation. Micropropagation is rapid, but laborious and technically demanding, requires hi-tech facilities, and is thus a relatively costly method. Expenses play a very important role in a mass reproduction, preventing large scale commercial use of this technique (Pulkkinen 2001).

Aspen propagation by woody stem sections might be applied as well; however, stem cuttings strike roots very hardly and slowly. Experiments to propagate aspen by non-woody stem sections have been made as well (Farmer et al. 1963); however, this method is not applied in forestry.

Propagation of aspen by root cuttings might be an efficient and inexpensive method of vegetative propagation which does not require complicated and expensive technology. Root cuttings might be used to propagate all tree species if shoots sprout from root cuttings buds (Farmer 1962). Therefore, propagation by root cuttings might be an alternative method compared with an expensive and elaborate micropropagation method.

Vegetatively propagated aspen trees are identical to the parental tree. If, however, the seedlings are raised from the seed, high genetic and even phenotypic variation might occur among them.

The largest demand for aspen wood is by the paper industry. To produce specialty papers, an aspen wood of uniform quality and of desired characteristics will be needed. Application of vegetative propagation methods allows exploitation of genetic variation and planting pure clones in the plantations (Zsuffa 1992).

Success of cultivation from root cuttings depends on a broad span of factors. When the roots are intact, auxins synthesized in the buds of the top branches

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are transported to the roots where they terminate root bud burst. This auxin to cytokinin ratio is easily broken by a stress like fire or tree harvesting (DeBell 1990, Zasada and Phipps 1990).

To stimulate this process various measures can be applied. Physiological tree status has an impact on rooting and sprouting efficiency too. During the period of leaf bud break the sprouting capacity is reduced considerably.

Wood properties can influence success of vegetative propagation as well, therefore, not all clones are suitable for mass propagation by root cuttings (Pulkkinen 2001). Among clones of hybrid poplar high variation in growth of both branch and root system as well as in wood production are found (Zsuffa 1992). The same variation is observed when these clones are propagated by root cuttings.

Stock plants selected for propagation must be healthy and vigorous; diseases and virus infections can decrease growth of the shoot and roots and reduce the amount of root cutting production. The ability of root cuttings to regenerate from unhealthy stock plants can be poor (Hartmann et al. 2002.). The water balance of the stock plant can affect the regeneration of cuttings too.

Manipulating the propagation conditions, it may be possible to improve the regeneration process. To improve rooting of stem cuttings, several species of stock plants have been grown in low light intensities (Eliasson and Brunes 1980). Certain rooting inhibitors have been found to be more abundant in tissues grown in intense light compared to tissues grown at lower levels of light. However, irradiance to the stock plants must be sufficient, because vigorously growing stock plants can produce more cuttings with high rooting ability than suffering plants can (Eliasson and Brunes 1980).

Age of the stock plant influences the ability of root cuttings to regenerate as well as the number of cuttings obtained (Hartmann et al. 2002). Due to their generally larger root system and longer shoot, older stock plants produce root cuttings more abundantly than younger plants. However, root cuttings from older plants may have lower potential to form adventitious roots than those taken from younger plants (Hackett 1988).

N. Stenvall et al. (2004) found that variation in sprouting can be influenced by the parental tree's age and genotype and that this variation reached 94% when aspen was propagated by root cuttings. Sprouting capacity of the most productive clones can exceed twice that of the less productive ones. The highest and lowest sprouting rate of the clones reached 67% and just 28%, respectively. The same experiment showed that sprouting of root cuttings was influenced by parental tree age also. It was found that root cut-

tings from younger parental trees showed a higher sprouting percentage; however, no statistically significant differences were obtained. As a final conclusion of their study, the authors recommended using 2-year old parental trees as a source of root cuttings for propagation.

The optimal propagation conditions for efficient and rapid shoot and root formation of root cuttings may differ greatly (Hartmann et al. 2002). The growth medium must be aerated, provide oxygen for the cuttings, but it must provide moisture for the root cuttings also (Hartmann et al. 2002). High air humidity helps to maintain sufficient moisture and decreases the risk of drying in root cuttings. If the root cuttings become dry for any length of time, the root cuttings will not root, even if they are later rehydrated (Hartmann et al. 2002).

Among environmental conditions air temperature has a large impact on aspen rooting and sprouting capacity too. High air temperature increases the loss of water from the leaves of stem cuttings, it also tends to promote development of the shoot before roots are formed in cuttings. (Hartmann et al. 2002).

The substrate's temperature has a large impact on aspen rooting capacity too. Increasing substrate's temperature by heating it artificially is a widely used measure in horticulture to root branch and root cuttings (Hartmann et al. 2002). High soil temperature may induce root initiation in cuttings, but lower soil temperatures are generally beneficial for elongation of roots (Hartmann et al. 2002).

According to Stenvall et al. (2005), aspen showed the highest rooting efficiency when the substrate temperature reached 30°C degrees. The authors also obtained statistically significant differences in rooting efficiency between 18°C and 30°C of substrate's temperature.

Rooting capacity of root cuttings was also influenced by other factors like plant hormones (the ration of auxines and cytokinines), amount of carbohydrates, plant physiological status and environmental conditions (Farmer et al. 1962, Hartmann et al. 2002).

Methods

Relationships between root cutting sprouting efficiency as influenced by parental tree age, type of aspen hybrid, and root diameter were calculated and analyzed. Studies were performed in the nurseries of the Dubrava and Anykščiai forest enterprises.

To evaluate the influence of age of the stock plant on the ability of root cuttings to regenerate, the cuttings were taken from hybrid aspen parental trees growing in the Vaišvydava forest district of the Du-

brava forest enterprise and park of Girionys (Table 1). Additionally, as a source for root cutting collection, 1-year-old parental trees were grown in the nursery of Anykščiai forest enterprise. The clones used in this

cuttings taken from the older trees showed a considerably lower sprouting percentage. An average sprouting of the root cuttings for 24-year-old hybrid aspen was 11%. Root cuttings taken from 39-year-old hybrid

Table 1. The plantations of hybrid aspen parental trees used for root cuttings in Dubrava forest enterprise

| No | Forest district | No of compartment | No of site | Year of establishment | Area, ha | Site type | Sum of temperature | Latitude | No of trees used for cuttings |
|----|-----------------|-------------------|------------|-----------------------|----------|---------------------|--------------------|----------|-------------------------------|
| 1 | Vaišvydava | 39 | 14 | 1983 | 2.0 | Oxalidone-morosum | 2,200-2,300 | 53°N | 23 |
| 2 | Park Girionys | 64 | | 1968 | 0.4 | Hepalito-oxalidosum | 2,200-2,300 | 53°N | 17 |

study were hybrid aspens originating from crosses between *Populus tremula* males from Lithuania (latitudes 53°N) and *P. tremuloides* females from Canada (latitudes 50°N). The clones were selected from a group of aspen clones that have been propagated in Lithuania for several years. These clones have also been micropropagated on a large scale. The root cuttings were taken in early April before budburst from healthy roots, diameter 3–15 millimetres, and cut into pieces 3 centimetres long.

For this study the following experimental variants were investigated:

1. root cuttings taken from 1-year old parental trees;
2. root cuttings taken from 24-year old parental trees;
3. root cuttings taken from 39-year old parental trees;

The number of root cuttings taken for each variant varied from 1,000 to 2,100 units.

Root cuttings from 24- and 39-year-old trees were taken by following the roots from mother tree a couple of metres before finding narrow roots. The effect of location of the root cutting (Stenvall et al. 2006) on the regeneration ability of hybrid aspen was not tested in this experiment.

Root cuttings were placed into plastic bags or mixed with peat and stored in moist cool basements where the temperature did not exceed +40°C.

To evaluate the influence of type of aspen hybrid on root cutting sprouting efficiency, the root cuttings were collected from the trees of two aspen hybrids: a *P. tremula x P. tremuloides* hybrid was compared with *P. tremula x P. alba* hybrid. The trees of both hybrids were 24years old. The number of root cuttings taken for each variant was 2,500 units.

To evaluate an influence of diameter of the root cuttings on the development of sprouts, root cuttings were taken from 39-year-old parental trees with the following experimental variants:

1. root cutting diameter up to 5 mm;
2. root cutting diameter 6-10 mm;
3. root cutting diameter over 10 mm.

The diameter at the midpoint of each root cutting was measured. The number of root cuttings taken for each variant varied from 1,300 to 2,000 units.

The experiments were performed in a greenhouse with relative humidity maintained at about 90% and temperature kept at about 18-22°C under natural light conditions. The root cuttings were planted into greenhouse beds in early May. Bed media were made of a mixture of the upper peat and sand at the proportion of 1:1. Root cuttings were placed horizontally one by another in rows leaving a space of 10 cm between the rows. Then the root cuttings were back-filled with a 3 cm deep layer of the same mixture. The experiments were irrigated with sprayers, no fertilizers were used.

Test plots of 1 m² were laid out in the beds. Each test variant consisted of 6 such test plots. The number of sprouted root cuttings was counted during the growth period at the intervals of 5 days, and assessments of sprout biometrical parameters (height and diameter) were performed at the end of the growing season. Five average sprouts were selected per plot. They were dug and the dry weight of various plant parts was evaluated.

The root cutting was considered to have sprouted when one or more buds of the cutting had begun to form a new shoot. The sprouting percentages were calculated per variable group by dividing the number of sprouted cuttings by the sum of all cuttings of the group.

The data were analyzed by ANOVA statistical methods and the results are presented in tables and figures.

Results

It has been found that the age of a parental tree had a great effect on sprouting capacity of aspen's and its hybrids root cuttings. The age has shown a strong and significant correlation with the percentage of sprouted root cuttings (Fig. 1). Younger parental trees have yielded in a higher percentage of sprouted root cuttings.

The highest sprouting percentage was shown by the root cuttings taken from 1-year old parental trees. Mean sprouting percentage of such root cuttings reached 52%. This is a relatively high percentage which can ensure successful mass sprout production. Root

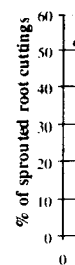


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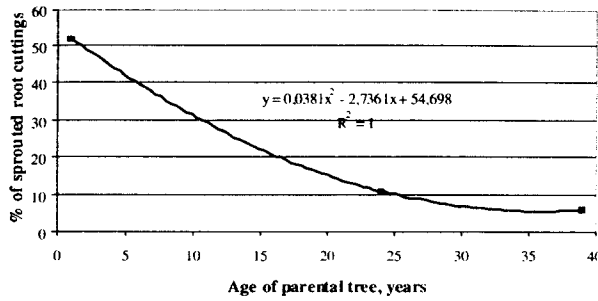


Figure 1. Regression of sprouting percentage as a function of age of the parental tree

aspen trees showed even less sprouting with just 6% of the root cuttings having sprouted.

A regression between age of a parental tree and root cutting sprouting can be expressed by a second degree polynomial equation:

$$y = 0.0381x^2 - 2.7361x + 54.698$$

A relationship between root cuttings sprouting capacity and age of parental tree was reported by Hackett (1988) as well. However, the relationship he found was linear and not polynomial.

Despite the fact that increased age of a parental tree resulted in lower sprouting percentage, only a limited number of root cuttings for mass sprout production can be prepared from 1-year-old trees. A higher number of root cuttings for propagation can be taken from an older tree than young one; however, due to the low sprouting percentage this option might not always be economically feasible. Therefore, young parental trees remain the most suitable source of root cuttings for vegetative aspen propagation because the sprouting percentage of their root cuttings is higher. This finding was confirmed by Stenvall et al. (2004) conclusion that the root cuttings from 2-year old parental trees should be taken for hybrid aspen propagation. Despite the fact that our study did not include 2-year-old parental trees, the regression equation between the age of parental tree and sprouting percentage showed that their sprouting percentage would not have differed significantly.

The type of aspen's hybrid used as a root cuttings source had an influence on sprouting of cuttings as well (Fig. 2). The results showed that a higher sprouting capacity could be expected with the *P. tremula x P. tremuloides* hybrid rather than the *P. tremula x P. alba* hybrid when used for root cutting preparation. It was found that sprouting percentage of *P. tremula x P. tremuloides* root cuttings reached 15% while that of *P. tremula x P. alba* root cuttings was just 9%. In both cases sprouting percentage was not high but

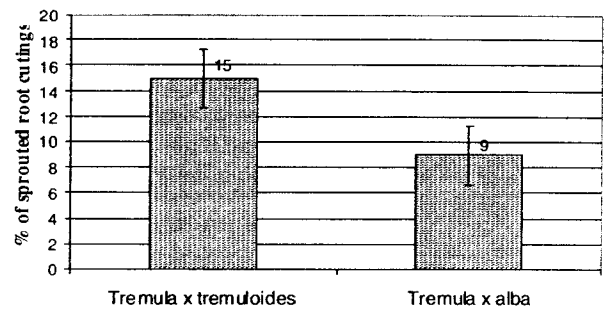


Figure 2. The influence of type of aspen hybrid used as a root cuttings source on sprouting of cuttings

this was also influenced by the fact that root cuttings were taken from 24-year-old parental trees whose sprouting capacity did not exceed 10%. Nevertheless, the obtained statistically significant differences in sprouting capacity between these two hybrids clearly illustrate existing differences.

These results can be compared to those of Pulkkinen (2001) and Haapala et al. (2004). Individual aspen hybrids showed specific differing wood properties that influenced success of vegetative propagation. Variation in growth of shoots and root system of individual clones of a hybrid aspen found by Zsuffa (1992) might be even higher among individual hybrids differing in growth productivity. Therefore, vegetative sprouts from the root cuttings of these hybrids also show the same variation.

Analysis of variance was applied to determine influences on root cutting sprouting capacity. The results showed that the age was the factor that had a major influence on sprouting capacity. Its effect reached almost 90% while the influence of hybrid's type was much weaker and reached approximately 10% (Table 2).

Table 2. Anova on the effects of age of parental tree and type of aspen hybrid used as a root cuttings source on sprouting of cuttings

| Source | MS | df | F | P-value | F crit |
|----------------------|----------|-----|--------|---------|--------|
| Age of parental tree | 40.66667 | 2 | 4.2244 | 0.009 | 3.212 |
| Aspen hybrid | 4.166667 | 1 | 4.328 | 0.019 | 3.212 |
| Error | | 982 | | | |

The results also showed that root cutting diameter had an influence on offspring biometric parameters. A hypothesis that larger root cuttings accumulate higher amounts of nutrients and thus stimulate sprouting of root cuttings and plant development was raised. This hypothesis was later confirmed by an actual finding that larger offspring do grow from larger root cuttings under uniform growth conditions.

Thicker root cuttings produced thicker sprouts (Fig. 3). Mean diameter of aspen sprouts at the root collar reached 3.6 mm when root cuttings were thin (up to 5 mm in diameter). Mean diameter of the sprouts that grew from the root cuttings 6-10 mm and over 10 mm thick at the height of root collar were 4.5 mm and 6 mm, respectively.

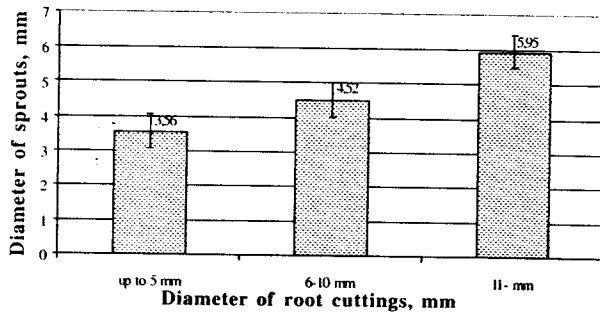


Figure 3. The influence of diameter of root cuttings on the diameter of aspen sprouts

When root cuttings were thicker, their sprouts were taller too (Fig. 4). Under uniform conditions the shortest sprouts grew from the thinnest (up to 5 mm thick) root cuttings. Their mean height was 28 cm. Sprouts of larger root cuttings were taller. 6-10 mm and over 10 mm thick root cuttings generated sprouts with average heights of 34 cm and 46 cm, respectively. ANOVA analysis of variance confirmed that the differences in biometric parameters of aspen sprouts that grew from the root cuttings of various thicknesses were statistically significant (Table 3).

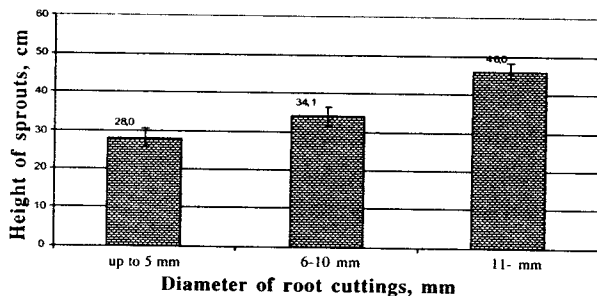


Figure 4. The influence of diameter of root cuttings on the height of aspen sprouts

Table 3. Anova on the effect of diameter of root cuttings on the height and the diameter of aspen sprouts

| Source | MS | df | F | P-value | F crit |
|----------|----------|-----|----------|---------|----------|
| Diameter | 38.33626 | 2 | 14.44901 | 0.00001 | 3.058928 |
| Height | 2170.687 | 2 | 8.956565 | 0.0002 | 3.059376 |
| Error | 242.3571 | 143 | | | |

Despite the fact that larger root cuttings produce larger planting material, over 15 mm thick root cuttings should not be used for propagation. The major limiting factor in selecting planting material for root cuttings is sprouting capacity which is determined by a number of root buds. The thicker the aspen's root is the lower the number of buds that can burst (Zsuffa 1992).

Conclusions

1. Age of hybrid aspen's parental tree had a significant relationship with the number of sprouting root cuttings. Root cuttings from younger parental trees showed a higher sprouting capacity.
2. Root cuttings of a *P. tremula* x *P. tremuloides* hybrid showed better sprouting capacity than those of *P. tremula* x *P. alba* hybrid.
3. Larger diameter root cuttings produced larger offspring.

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ВЛИЯНИЕ ХАРАКТЕРИСТИК МАТЕРИНСКИХ ДЕРЕВЬЕВ ОСИНЫ ГИБРИДНОЙ НА РАЗМНОЖЕНИЕ ЕЁ КОРНЕВЫМИ ЧЕРЕНКАМИ

В. Сухоцкас

Резюме

Целью данного исследования является изучение влияния возраста материнского дерева осины гибридной на укоренение корневых черенков. Установлено, что возраст материнского дерева осины и её гибридов имеет существенное влияние на укоренение корневых черенков. Возраст материнского дерева имеет достоверную корреляцию с процентом укоренившихся черенков. Более молодые материнские деревья осины способствуют увеличению процента укоренившихся черенков.

Результаты исследования также показали, что диаметр корневого черенка влияет на биометрические параметры всходов. Практика подтвердила гипотезу, что черенки большего диаметра скапливают более высокое содержание питательных веществ, и это в свою очередь стимулирует развитие всходов. При одинаковых условиях окружающей среды более крупные саженцы развиваются из черенков большего диаметра.

Более высоким процентом укоренения отличаются корневые черенки *P. tremula* x *P. tremuloides* гибрида, по сравнению с гибридом *P. tremula* x *P. alba*. Укоренение корневых черенков *P. tremula* x *P. tremuloides* в данной работе составило 15%, в то время как *P. tremula* x *P. alba* только 9%.

Ключевые слова: осина, корневые черенки, всходы, высота, диаметр

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