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Full Length Research Paper

Phenotypic stability of *Acer pseudoplatanus* cv Atropurpureum traits as a baseline of breeding process

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Aiming at the conservation and improvement of valuable traits and intensified production of *Acer pseudoplatanus* cv Atropurpureum planting material for domestic and foreign market, a series of experiments was established to monitor the phenotype expression and stability of the character of purple leaf underside, the study results showed the continuous presence of the analysed character in the progeny of all mother trees, in higher or lower percentages, which can be used as a starting point for mass production of this cultivar, both by generative and vegetative methods.

Key words: cv Atropurpureum , expression, half-sib lines, phenotype stability, purple undersides, seedling production, sycamore maple.

INTRODUCTION

Biological resources and biological diversity confer a great many benefits on society. These benefits are most easily recognisable when embodied in a commodity of market value. In this case, social benefits can be proxied by the process people are willing to pay (Pregernig, 2005).

During the last decades, forestry in Europe has shown a growing awareness of the high importance of a specific group of tree species known as noble hardwoods. The acknowledgement of their eminent silvicultural, economic and/or ecological values led to their upgrading from a former status of 'minor' species.

Out of this awareness arose a major concern to safeguard the existing resources of these species (isolated individuals, relict populations, small-scale homogeneous stands). Because one cannot protect what one does not know, substantial efforts have been needed, in the first place, to identify the still existing resources (De Cuyper, 2007).

Conservation and improvement of valuable traits of noble hardwood species applying research and breeding methods approach with the final result of intensive mass production of breed individuals can be considered as a tool for social, economic and overall valorisation of biodiversity.

For rarely occurring species, grafting of trees and establishment of plantations of grafts in seed orchards or clonal archives are suggested to increase the effective population size (Eriksson, 2001).

Based on numerous researches, it can be inferred that a substantial number of forest tree characters significant for breeding are determined by cytoplasmatic structures. Of all the cytoplasm organelles, the best studied is the effect of plastids and mitochondria on phenotype characters and the role and effect of plastids in extra-chromosomal inheritance is successfully applied in the cultivar synthesis in floriculture, horticulture and forestry. The spontaneous occurrence of the green leaf colour change into violet, yellow or purple is the consequence of anthocyanin presence in the cell sap of epidermal cells, or of carotenoid presence in chromoplasts (Sari et al., 1989). Pronounced phenotype attractiveness of such individuals led to the synthesis of many cultivars used in mass production of ornamental trees and shrubs. Previous experimental research shows that the character of leaf colour is predominantly inherited from mother, which is designated as matriclinous inheritance and indicates the cytoplasmatic inheritance of this trait. However, in rare cases and in low %, the plastids of normal pigmentation, as well as

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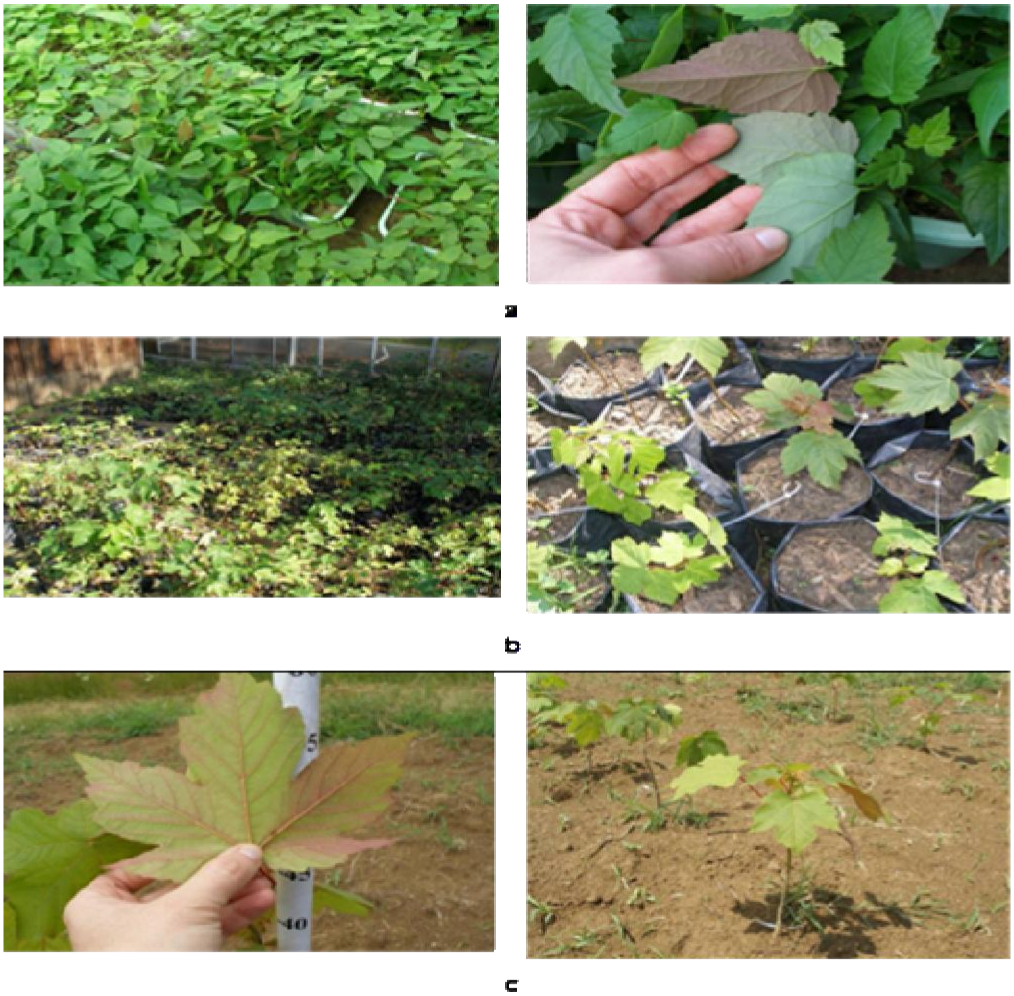

Picture 1. Experiments in the greenhouse of the faculty of forestry in Belgrade (a) in the nursery of the faculty of forestry in Belgrade (b) and in the field, surroundings of Ljig, Serbia (c).

different chlorophyll mutations, can be transmitted to the progeny via pollen. This trait characterises some species in the genus *Epilobium*, where 3% of the progeny expresses the plastids of the male parent, as well as the species *Pelargonium zonale* in which this % ranges up to 30% (Isajev and Šija i -Nikoli , 2003).

MATERIAL AND METHODS

Sycamore maple (*Acer pseudoplatanus* L.) is one of the most valuable species of our noble hardwoods. Based on results of previous research (Fukarek, 1961; Gudeski, 1984; Tripi , 1988; Bojovi , 1989; Iveti and Tucovi , 2003; Šija i -Nikoli et al., 2006), it can be concluded that this is a species with a pronounced ecological plasticity and high genetic potential, exemplified by a great number of cultivated forms. *Acer pseudoplatanus* L. cv Atropurpureum , cv Briliantissimum , cv Variegatum , cv Leopoldi , cv Flavescens , cv Flavovariegatum , cv Worleei (Bojovi , 1989). From the aspect of ornamentalness, one of the most frequent cultivars in urban green spaces and tree rows is cv Atropurpureum also known as Spaethii or Purpureum . It is distinguished by dark green leaf adaxials and purple undersides, which gives the crown an extraordinarily attracttive appearance.

This research, aiming at the analysis of the expression and phenotype stability of characters of the cultivar Atropurpureum in the earliest stages of ontogenesis, was conducted on 9 half-sib lines representing the progeny of the selected trees in the area of Belgrade. Starting from the reference data (Stilinovi , 1987), showing that the sown cv. Atropurpureum seeds produce 50% of individuals with purple undersides and 50% individuals with green undersides, the sowing experiment started in the autumn 2005. The expression and stability of purple undersides in the progeny was monitored in the experiments established in the greenhouse, in the nursery and in the field, Picture 1.

Seeding was conducted in autumn 2005, in 60 x 40 cm plastic containers, into the supstrat mixture of peat, sand and mold (2:2:1), in the greenhouse with controlled system of irrigation. 3 months after germination, plants have been allocated to open area conditions. In spring 2006, 1 year old seedlings were replanted into plastic bags and 1 year after, the field test was established from 2 year old plants.

The leaf underside colour was analysed on the sample of 50 plants per line, at the ages of 1, 2 and 3 years, at the beginning (immediately after leaf formation) and at the end of the growing

Table 1. % of plants with green, purple and variegated undersides in different half-sib lines of sycamore maple, at the end of the first growing season.

Line	Average number of leaves per plant	Leaves with green underside (%)	Leaves with purple underside (%)	Leaves with variegated underside (%)
1	5.52	10.15	85.87	3.98
2	4.70	49.79	46.81	3.40
3	4.82	12.86	81.33	5.81
4	4.86	50.21	44.44	5.35
5	4.78	0.00	94.14	5.86
6	5.58	81.72	17.20	1.08
7	5.58	74.55	24.37	1.08
8	6.02	6.98	90.37	2.65
9	5.14	46.30	52.14	1.56

Table 2. % of plants with green, purple and variegated undersides in different sycamore maple half-sib lines: A - at the beginning of the second growing season and B - at the end of the second growing season.

Line	A			B		
	Plants with green undersides (%)	Plants with purple undersides (%)	Plants with variegated undersides (%)	Plants with green undersides (%)	Plants with purple undersides (%)	Plants with variegated Undersides (%)
1	3.33	83.34	13.33	10.88	81.12	8.00
2	60.00	30.00	10.00	60.00	36.67	3.33
3	23.33	63.34	13.33	13.33	80.00	6.67
4	33.33	50.00	16.67	33.33	66.67	0
5	3.34	66.66	30.00	3.33	90.00	6.67
6	76.67	16.67	6.66	90.00	3.33	6.67
7	80.00	13.33	6.67	80.00	13.33	6.67
8	6.67	83.33	10.00	10.00	80.00	10.00
9	56.67	36.67	6.66	63.33	20.00	16.67

season (right before leaf fall). The collected data showed the % of plants with purple undersides, plants with green undersides and plants with variegated undersides within the analysed half-sib lines. Cluster analysis was performed in program “Statistika 6.0”, based on data on plants with purple underside %, in the aim of determining the genetic similarity, that is, distance among the analysed half-sib lines, based on the data collected in all 3 years of research.

RESULTS

The results of the analysis of phenotype expression of sycamore maple purple/green leaf undersides in 9 half-sib lines in the earliest stage of ontogenesis are presented in Table 1.

The research performed at the end of the first growing season shows a clear differentiation of lines into those with the plants with a higher % of green undersides (lines 6 and 7), those with purple undersides (lines 1, 3, 5 and 8) and those with approximately equal % of plants in both categories (lines 2, 4 and 9), Table 1. Plants with variegated leaves occurred in almost all lines, in relatively low % (from 1.08 to 5.86%).

Phenotype expression of purple/green leaf undersides at the beginning (A) and at the end of the growing season (B), at the ages of 2 and 3 years, is presented in Tables 2 and 3. The study results show the phenotype stability of the expression of purple undersides of the lines 5, 1, 3 and 8, expressed in all 3 study years and during the growing seasons, with average values 82.83, 80.30, 79.20 and 75.94%, respectively. Also, phenotype stability of the expression of green undersides was continuous in the lines 6 and 7, with average values 82.80 and 80.91%. The lines 9 and 2 show the increasing % of plants with green undersides with the age and during the growing season. The study results are additionally illustrated by the dendrogram cluster analysis, Figure 1, which shows clearly the 2 homogeneous groups: group 1, consisting of

Table 3. % of plants with green, purple and variegated undersides in different sycamore maple half-sib lines: A - at the beginning of the third growing season and B - at the end of the third growing season.

Line	A			B		
	Plants with green undersides (%)	Plants with purple undersides (%)	Plants with variegated undersides (%)	Plants with green undersides (%)	Plants with purple undersides (%)	Plants with variegated undersides (%)
1	11.76	82.36	5.88	25.29	68.83	5.88
2	70.00	30.00	0	70.00	30.00	0
3	13.34	86.66	0	15.33	84.67	0
4	53.33	33.33	13.34	73.33	16.67	10.00
5	3.33	90.00	6.67	26.66	73.34	0
6	-	-	-	-	-	-
7	83.33	13.33	3.34	86.67	10.00	3.33
8	20.00	64.00	16.00	30.00	62.00	8.00
9	80.00	16.67	3.33	83.33	10.47	6.20

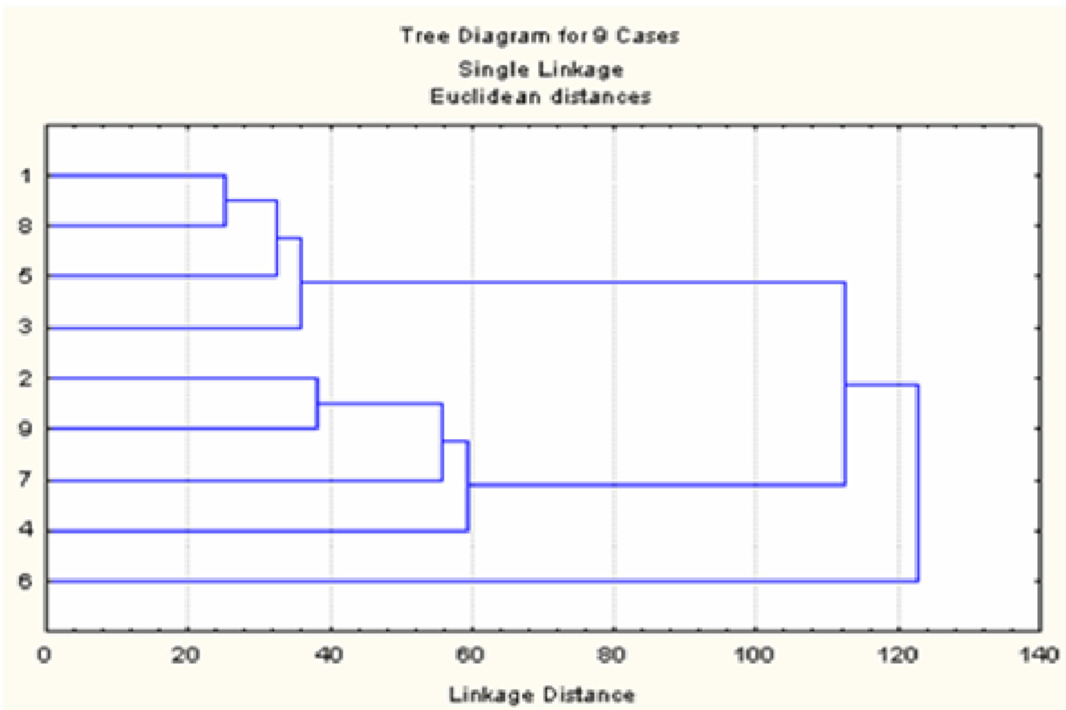

Figure 1. Cluster analysis dendrogram based on the analysis of phenotype expression of leaf underside colour during 3 growing seasons.

the lines with prevailing plants with purple undersides (1, 8, 5 and 3) and group 2, consisting of the lines 2 and 9, which subsequently group with lines 7 and 4 and whose plants mainly have green undersides.

DISCUSSION

The main directions of the reconstruction of the existing species and the creation of the new cultivated forms (cultivars) consist of the detection and definition of the desired characters, selection of initial material, identification of qualitative and quantitative parameters, study of the methods of inheritance of the selected characters and mass production of the improved planting material (Šija i - Nikoli et al., 2006).

Aiming at the intensified production of *A. pseudoplatanus* L. cv Atropurpureum planting material for domestic and foreign market, a series of experiments was established to monitor the phenotype expression and stability of the character of purple leaf underside. The study results show the constant presence of the analysed character in the progeny of all mother trees in different %. The progeny of mother trees 1, 3, 5 and 8 is characterised by a continuously high % (about 80%) of purple leaf undersides, so we can infer its genetic determination, which is also approved with the dendrogram of cluster analyses showing 1 homogeneous group of these mother trees connected on relatively short distance. In future, these trees should be used as the source of genetically good-quality seed material for mass production of this cultivar. Taking into account the high germination % of seeds of mother trees 2, 4, 6 and 7 (60 - 91%), (Šija i - Nikoli et al., 2006), whose progeny predominantly forms good-quality plants with green undersides, in future these trees can be used as the source of seeds for mass production of rootstocks, for which the scions should be taken from the trees 1, 3, 5 or 8, by the recommended method of budding dormant or active buds (Isajev and Šija-i -Nikoli , 2003), which will result in the greater part (80%) of the generative progeny with the desired charac-ters.

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