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Genetic variability of bilaterally symmetrical fruits of Norway maple in function of species biodiversity conservation

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Abstract: Parkway trees in urban environments have a pivotal ecological role, primarily because they modify microclimatological parameters. Norway maple is a suitable species for these purposes since it is very adaptable to city conditions. This characteristic of Norway maple allows urban trees to be used as a basis for biodiversity conservation. Bilaterally symmetrical fruits of Norway maple are particularly convenient for the exploration of developmental stability and the role of genetic and environment in variability of morphological characteristics of Norway maple. Variability change through different conditions could be used for creation of a scale that could serve to predict morphology of Norway maple in different environments. The differences between fruits collected from 4 locations show great variability of the species, but the size and appearance of the fruits are in accordance with literature data and do not deviate in any group of samples. Despite similar normal distributions of groups, the fruits differ significantly, although these variations are not consequences of a cause-and-effect connection between environment and appearance of fruits. Variability of samples taken from the urban environment does not differ from variability of samples taken from natural habitats. The large variability of Norway maple is expected since it has a wide ecological amplitude as well as a small plasticity, which is typical of species adapted to shadowy conditions. The great variability of fruit morphology within each individual is typical of Norway maple, so these could be used as representatives of the population in which they are grown. In this paper it is shown that the differences found between fruits are the result of genetic variations mosaic; that is, they are different ecotypes of Norway maple, since bilaterally symmetrical fruits do not have significantly different left and right sides, which shows the developmental stability of Norway maple.

Key words: *Acer platanoides* L., genotype, morphogenetic control, Norway maple, polymorphism

1. Introduction

Urban growth is identified as a grave danger to biodiversity, but at the same time urban regions may conserve an array of species and contribute to conservation and biodiversity (Sukopp and Werner, 1983; Gilbert, 1989; Pyšek, 1993; McKinney, 2002). Nevertheless, distant urban ecosystems cannot completely replace the function of systems of habitats that are close to the natural state (Kowarik et al., 2011). Despite significant human influence, urban tree communities (including species for which it is not the natural habitat) are determined by the same climatic factors that shape indigenous plant communities (Ramage et al., 2013).

Parkway trees are of great importance for the urban environment, primarily because of their climatological and meteorological influence (Meier and Scherer, 2012), and they are exposed to a relatively high level of stress, which results in shortened life expectancy (Sæbø et al., 2005). The selection of taxa for these purposes is of key importance in urban planning in order to optimize management of expenses and human comfort (Leuzinger and Körner, 2010). The wrong choice can cause problems such as pavement impairment, tangling in communication lines, or traffic distractions (Gerhold and Porter, 2000).

Acer platanoides L. is a species of an attractive form. It is susceptible to a small number of entomological and phytopathological diseases, performs well on poor soils, and is resistant to pollutants, and therefore it is the best species for urban parkways (Nowak and Rowntree, 1990). For this reason, it is among the most frequent in city alleys (Iles and Vold, 2003). Norway maple is characterized by an abundant fruiting and hence spreads swiftly. It has high photosynthetic potential, and it is shade-tolerant and grows quickly (Kloeppel and Abrams, 1995). The lack of nutrients does not result in decrease in the leaf size, nor does it affect biomass (Black-Samuelsson and Andersson, 2003). Among a group of 10 nonnative species in Helsinki and Vantaa in Finland, unfavorable conditions

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affected Norway maple growth and development the least (Lehvavirta and Hannu, 2002). The purpose of this paper is to examine the differences in morphology between fruit samples taken from trees in 3 natural habitats and samples from trees of urban parkways in order to determine whether morphology of this species changes as a consequence of urban environmental conditions.

Variations in plant appearance develop as a result of many factors. Apart from genetic plasticity, they are influenced by availability of nutrients, air temperature, phenology, age, environmental characteristics, and concentration of carbon dioxide. Therefore, it is not known how variations in morphology change through ecological scales, which prevents us from establishing a scale of characteristic variations in relation to different environmental aspects (Messier et al., 2010). Morphologic research of trees from urban areas and from natural habitats could contribute to the creation of such scales and help understand the environmental and genetic roles in the morphology of species. The role of phenotypic plasticity is different, partly due to fitness of plants and partly due to genotype (Pigliucci, 2001). Although phenotype depends on hereditary factors whose manifestation is directly linked to environment, population dynamics, adaptability of species, and evolution of plant character (Rossiter, 1996), close relations of morphology with the environment are characteristic of species of a small ecological amplitude that are specialized to certain habitats. Species of a wide ecological amplitude are more tolerant of environmental changes and their morphology is not influenced by environment to such an extent, but rather is a consequence of genetic variability (Geng et al., 2012). Shade-tolerant species are generally of less genetic plasticity, although an increase in nutrient availability reduces differences in plasticity (Portsmuth and Niinemets, 2007). Somatic mutations of plants can be inherited by natural mechanisms of sexual and asexual reproduction. Long life expectancy, a large number of clones, and a complete regeneration of buds each year enable individual plants or clones to evolve. Plants can also develop a mosaic of genetic variations. This evolution also results in fine adjustment to local conditions of environment, which creates ecotypic variations (Whitham and Slobodchikoff, 1981).

It is proven that morphology of certain plant parts is primarily determined by general characteristics of species more than was formerly believed (Poorter and Rozendaal, 2008). No evidence was found for the influence of unfavorable growing conditions on developmental stability of Norway maple's appearance. Morphology variations of large number of species can be shown within only one representative. Latter research confirmed these findings and supported scientists' tendency to divert focus from species-based ecology to plant part-based ecology (Messier et al., 2010). In the previous research of Norway maple parkway trees, it was determined that variations of leaf morphology differed significantly depending on the location from which the samples were taken. Samples were taken from 3 natural habitats and from an urban environment. Still, there was no cause-and-effect relation between environment and leaf morphology (the connection was 3%). This variability is, therefore, the consequence of genetic variability of species (Simović et al., 2013). Due to its wide distribution, Norway maple is likely to have large genetic variations, and distant climatic populations could develop as a response of plants to different environmental conditions (Yao and Tigerstedt, 1995; Westergaard, 1997; Joyce et al., 2002).

Bilaterally symmetrical fruits typical of Norway maple are especially convenient for research of genetic variability. The measure of the left and right sides of bilaterally symmetrical characteristics offer 2 repeated measurements of the same developmental process, as they developed at the same time in the same environmental conditions. Hence, the assessment of repeatability of developmental process is possible as it gives insight into genotype capacity to control developmental stability (Whitlock, 1996). Small deviations from perfect symmetry of fruit are considered to be the result of genotype influence or disruption of environmental conditions (Møller and Swaddle, 1997). The points listed above can be examined within any sample individual and samples taken from different populations could be used for the examination of the environmental role in morphological variability. This would also show whether there is a difference between variation in morphology of samples taken from an urban environment and samples taken from natural habitats.

2. Material and methods

A sample of 1600 trees of approximate age was used for the research of morphological characteristics of Norway maple fruits. From each tree, a sample of 100 fruits was taken from the southern part of the lower third of the canopy.

The fruits were collected in 2013 from 4 locations: parkways in the Belgrade city center (from 400 trees), and from 400 trees from each of 3 locations in cadastral municipality Rudnik in department numbers 63, 64, and 73. The locations in Rudnik belong to the same climate and the large sample provides a representative model that covers differences in microclimate.

The analysis included the fruit length (cm), the length of achenes (cm), the length of fruit wings (cm), the largest width of wings (cm), the smallest width of wings (cm), and the inner and outer angle between the wings (°).

The rate of central tendency and average values were calculated with IBM SPSS Statistics as well as the differences between individuals and populations, which were determined by multiple analysis of variance and Scheffe's test.

3. Results

The morphometrical analysis of Norway maple fruits included 11 characteristics of fruits within samples of 40,000 fruits from each of 4 locations: the fruits from parkway trees in the Belgrade city center and from trees in Rudnik (departments 63, 64, and 73).

For all of the 11 parameters, at each location dispersions of samples were similar; that is, standard deviations and variances were similar. Limit values of fruit size were different but results were grouped in a similar vein (Tables 1–4).

The multiple analysis of variance was used for the determination of differences between groups.

The fruit size varied from 3.8 cm to 10.3 cm, and the average fruit length of Norway maple was 6.8 cm. The average length of achenes was 1 cm and 1.1 cm. The average length of wings was 3.5 cm and 3.4 cm, the largest width was 1.2 cm, and the smallest width was 0.9 cm. The value of outer angle varied from 100° to 180° and the average value was 144°, and the value of the inner angle varied from 90° to 160° with an average value of 132° (Table 5).

The differences between samples taken from 4 locations were significant at the level of $P < 0.01$ (Table 6). However, there was no cause-and-effect connection between environments from which the samples were taken and these differences ($R^2 < 1\%$). Variation of samples taken

Table 1. Statistical parameters for fruits of trees grown in Belgrade.

Parameter	N	Min.	Max.	Mean	Std. deviation
Fruit length	40,000	3.80	10.30	6.9215	1.10054
Length of the left achene	40,000	0.70	6.10	1.1287	0.17759
Length of the right achene	40,000	0.60	11.00	1.1093	0.16911
Length of the left wing	40,000	1.00	5.70	3.5502	0.60145
Length of the right wing	40,000	0.80	5.50	3.5214	0.59491
The largest width (left wing)	40,000	0.60	3.30	1.2281	0.20710
The largest width (right wing)	40,000	0.50	1.90	1.2183	0.21270
The smallest width (left wing)	40,000	0.00	8.00	0.9455	0.13682
The smallest width (right wing)	40,000	0.10	9.00	0.9446	0.17584
Outer angle	40,000	101.00	180.00	144.7584	7.76349
Inner angle	40,000	90.00	160.00	131.9221	7.50498
Valid N (list-wise)	40,000				

Table 2. Statistical parameters for fruits of trees grown in Rudnik (department 63).

Parameter	N	Min.	Max.	Mean	Std. deviation
Fruit length	40,000	3.80	10.30	6.8115	1.04629
Length of the left achene	40,000	0.70	6.10	1.1183	0.15877
Length of the right achene	40,000	0.60	11.00	1.1033	0.17892
Length of the left wing	40,000	1.00	5.70	3.4870	0.56452
Length of the right wing	40,000	0.80	5.50	3.4550	0.55938
The largest width (left wing)	40,000	0.60	3.30	1.2348	0.20200
The largest width (right wing)	40,000	0.50	3.30	1.2262	0.20227
The smallest width (left wing)	40,000	0.00	8.00	0.9537	0.14645
The smallest width (right wing)	40,000	0.10	9.00	0.9505	0.16051
Outer angle	40,000	101.00	175.00	143.2143	5.58269
Inner angle	40,000	90.00	160.00	131.7760	5.78758
Valid N (list-wise)	40,000				

Table 3. Statistical parameters for fruits of trees grown in Rudnik (department 64).

Parameter	N	Min.	Max.	Mean	Std. deviation
Fruit length	40,000	3.80	10.30	6.7380	1.01209
Length of the left achene	40,000	0.70	6.10	1.1036	0.15483
Length of the right achene	40,000	0.60	11.00	1.0876	0.18885
Length of the left wing	40,000	1.00	5.70	3.4464	0.54871
Length of the right wing	40,000	0.80	5.50	3.4170	0.54243
The largest width (left wing)	40,000	0.60	3.30	1.2223	0.20090
The largest width (right wing)	40,000	0.50	3.30	1.2131	0.19886
The smallest width (left wing)	40,000	0.00	8.00	0.9450	0.13748
The smallest width (right wing)	40,000	0.10	9.00	0.9435	0.16981
Outer angle	40,000	101.00	175.00	143.6885	5.48805
Inner angle	40,000	90.00	160.00	132.1970	5.70195
Valid N (list-wise)	40,000				

Table 4. Statistical parameters for fruits of trees grown in Rudnik (department 73).

Parameter	N	Min.	Max.	Mean	Std. Deviation
Fruit length	40,000	3.80	10.30	6.7746	1.02071
Length of the left achene	40,000	0.70	6.10	1.1135	0.15485
Length of the right achene	40,000	0.60	11.00	1.0974	0.17288
Length of the left wing	40,000	1.00	5.70	3.4666	0.55109
Length of the right wing	40,000	0.80	5.50	3.4354	0.54719
The largest width (left wing)	40,000	0.60	3.30	1.2285	0.19948
The largest width (right wing)	40,000	0.50	3.30	1.2195	0.19939
The smallest width (left wing)	40,000	0.00	8.00	0.9503	0.14890
The smallest width (right wing)	40,000	0.10	9.00	0.9476	0.15941
Outer angle	40,000	101.00	175.00	143.3929	5.52201
Inner angle	40,000	90.00	160.00	132.0248	5.71753
Valid N (list-wise)	40,000				

from the urban environment did not differ from variations of samples taken from natural habitat.

The morphology of samples from each of the 4 locations was different: for fruit length, $F = 230$, $df = 3$, $P < 0.01$; for the length of the left achene, $F = 167$, $df = 3$, $P < 0.01$; for the length of the right achene, $F = 108$, $df = 3$, $P < 0.01$; for the length of the left wing, $F = 252$, $df = 3$, $P < 0.01$; for the length of the right wing, $F = 263$, $df = 3$, $P < 0.01$; for the largest width of the left wing, $F = 25$, $df = 3$, $P < 0.01$; for the largest width of the right wing, $F = 28$, $df = 3$, $P < 0.01$; for the smallest width of the left wing, $F = 33$, $df = 3$, $P < 0.01$; for the smallest width of the right wing, $F = 14$, $df = 3$, $P < 0.01$; for the outer angle, $F = 503$, $df = 3$, $P < 0.01$; and for the inner angle, $F = 32$, $df = 3$, $P < 0.01$.

Scheffe's test confirmed that differences were not the consequence of environmental factors ($P > 0.01$ for all of the 11 parameters of fruit appearance).

4. Discussion

Norway maple fruit size was within the limit values listed in literature (Vukićević, 1982), with an average value of 6.8 cm for the whole sample. The size of achenes was 1 cm to 1.1 cm, the wings were 3.5 cm and 3.4 cm, with the largest width of 1.2 cm and the smallest of 0.9 cm. These values were similar for all 4 groups of samples and are in accordance with literature data for size and appearance of Norway maple fruit. The same was proven with the analysis of outer and inner angles of fruit wings,

Table 5. Statistical parameters for the whole fruit sample.

Parameter	N	Min.	Max.	Mean	Std. deviation
Fruit length	160,000	3.80	10.30	6.8114	1.04772
Length of the left achene	160,000	0.70	6.10	1.1160	0.16204
Length of the right achene	160,000	0.60	11.00	1.0994	0.17777
Length of the left wing	160,000	1.00	5.70	3.4876	0.56816
Length of the right wing	160,000	0.80	5.50	3.4572	0.56273
The largest width (left wing)	160,000	0.60	3.30	1.2284	0.20244
The largest width (right wing)	160,000	0.50	3.30	1.2193	0.20343
The smallest width (left wing)	160,000	0.00	8.00	0.9486	0.14255
The smallest width (right wing)	160,000	0.10	9.00	0.9465	0.16655
Outer angle	160,000	101.00	180.00	143.7635	6.19438
Inner angle	160,000	90.00	160.00	131.9800	6.22725
Valid N (list-wise)	160,000				

Table 6. Multiple analysis of variance. Different letters in a column show significant difference.

Effect		Value	F	Hyp. df	Error df	Sig.
Intercept	Pillai's trace	0.999	12,657,901.119^{b}	11.000	159,986.000	0.000
	Wilks' lambda	0.001	12,657,901.119^{b}	11.000	159,986.000	0.000
	Hotelling's trace	870.307	12,657,901.119^{b}	11.000	159,986.000	0.000
	Roy's largest root	870.307	1,2657,901.119^{b}	11.000	159,986.000	0.000
Location	Pillai's trace	0.050	247.340	33.000	479,964.000	0.000
	Wilks' lambda	0.950	251.169	33.000	471,348.893	0.000
	Hotelling's trace	0.053	254.961	33.000	479,954.000	0.000
	Roy's largest root	0.050	733.607^{c}	11.000	159,988.000	0.000

whose average values were 144° for outer and 132° for inner angle.

Standard deviations and variance were different for all of the 11 characteristics of Norway maple fruits, but were distributed similarly.

Results of multiple analysis of variance showed that characteristics of Norway maple fruits differed significantly depending on the location from which samples were taken ($P < 0.01$), and that variation of samples taken from the city center of Belgrade did not differ from variation of samples taken from 3 locations in Rudnik. This confirms that unfavorable conditions of urban environments do not influence developmental stability of Norway maple appearance.

Urban growth could indeed contribute to biodiversity conservation since the differences between Norway maple fruit characteristics in an urban environment in Belgrade and natural habitats in Rudnik are primarily the result of genetic variability and not the environment; respectively, there was no cause-and-effect connection between location and the differences in sample variability ($R^2 <$ 1%). These differences are in accordance with previous research (Kowarik et al., 2011) showing that urban populations could not replace natural habitats when it came to biodiversity conservation.

Norway maple is already recognized as a species suitable for urban parkways since the unfavorable city conditions have minor effect on this species. Differences between fruits (as well as leaves (Simović et al., 2013)) of urban trees and trees from natural habitats were significantly different but were not the consequence of different environments for any of the 11 parameters of fruits ($P >$ 0.01). Variations in fruit appearance were smaller than those in leaf appearance of the same samples. Preservation of Norway maple morphological characteristics of fruit in urban environments justifies its use for these purposes. The use of species adaptable to urban environments for parkways contributes to better optimization between costs of city landscapes preservation and positive effects on urban microclimate.

Variability that appears in different populations may be the consequence of large genetic variability that is characteristic for all species of a wide ecological amplitude, among which is Norway maple. It is also one of the species adaptable to shadowy conditions, meaning smaller genetic plasticity. Differences determined between populations may be ecotypic variations and large variability within each tree may be the result of a mosaic of genetic variations, which, according to previous research, appear due to a large number of clones, long life expectancy, and regeneration of buds (Whitham and Slobodchikoff, 1981). The results confirm diversity in fruit appearance, and statistical methods show that it is not the consequence of different environments.

Due to large variability between trees, there is a tendency for genetic variability to be examined in one individual as a representative of the species. The study results verify that there are differences between populations and, therefore, selected trees could be representatives of populations from which they were taken for samples from both urban environments and natural habitats. For this kind of research, Norway maple fruits are notably suitable due to their bilateral symmetry. This type of fruit offers 2 values of the same characteristic of the plant part developing in identical conditions, so stability of developmental processes can be determined according to these values. The differences in tested samples between the left and right side are minor (in the average length of achene and the average length of wings they are 1 mm, and the largest and the smallest widths of fruits are identical for both the left and right side) for samples from the urban environment as well as for samples from natural habitats.

In this paper, it is shown that analyzed morphometrical characteristics of Norway maple fruits differ significantly depending on the location from which samples were taken. However, statistical analysis proved that there was no cause-and-effect connection between environment and these characteristics. The variability of fruits in the urban environment did not deviate from the variability of fruits from natural habitats. Therefore, urban populations are also considered suitable for sustainability of biodiversity of species and Norway maple has proven itself to be a suitable species for urban parkways.

Limit values of fruit dimensions in the sample populations of Norway maple did not deviate from literature data. The variability of measured characteristics was large and different for all of the 11 measured characteristics, but these values were distributed in the same vein. The variability of morphometrical characteristics of Norway maple fruits is connected to genetic variability or even a mosaic of genetic variability that appears within a single tree. Small genetic plasticity and large variability of Norway maple are expected since it is a species of wide ecological amplitude that also adapts to shadowy environments. The differences between populations from various locations may be due to different ecotypic variations of the species, but the large variability is characteristic of species and so these differences cannot be used as the determining factor of different phenotypes. Variation of fruit morphology of samples from the urban environment was not due to unfavorable conditions, but was rather the consequence of genetic variability of the species.

Norway maple fruits are bilaterally symmetrical and therefore suitable for examination of developmental processes, i.e. the influence of genetics and environment. The differences between the left and right sides of the fruit are minor.

These results on the variability of Norway maple fruit characteristics are a basis for selection of genotypes important for application in forestry, landscape architecture, horticulture, and foundation of cultures. These results could also be used as basis for creation of a scale of Norway maple characteristic variations in relation to different environmental aspects.

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